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Norwegian Scientific Committee for Food Safety

Risk assessment of mycotoxins in cereal grain in Norway

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Summary

Cereals can become infected by fungi that contaminate the grain with toxic secondary metabolites, called mycotoxins. Mycotoxins in cereal grain can pose a risk to both human and animal health. Long-term intake of mycotoxins found in Norwegian cereal grains can, among other effects, cause reduced immune responses, reduced growth and affect reproduction. In the European Economic Area (EEA), maximum limits (MLs) have therefore been determined for mycotoxins in food and recommended for mycotoxins in feed.

In the light of constantly new knowledge on toxicity and the rise in the level of mycotoxin contamination during recent years, in a letter dated September 21st 2010, the Norwegian Food Safety Authority requested the Norwegian Scientific Committee for Food Safety (VKM) to undertake a risk assessment on mycotoxins in cereal grain. The risk assessment should cover the fungi and toxins present in grain and grain-related products available on the Norwegian market (both domestic and imported, food and feed, including fish feed). In addition to an evaluation concerning human and animal health, the risk assessment should consider the different factors affecting fungal infection and mycotoxin production in cereal grain, both in the field and during storage. The risk assessment will be used as a scientific background for risk management related to agricultural practices, food and feed production and to modify MLs in food and feed. In addition to contributing to the constant development of regulations in the EU, the Norwegian Food Safety Authority will communicate the risk to producers in agriculture and industry.

In order to answer the request from the Norwegian Food Safety Authority, VKM appointed three project groups consisting of both VKM members and external experts, covering the different topics of food and human health, feed and animal health, and plant health. The reports from the project groups have been evaluated and approved by the relevant VKM panels. The final opinion, the current document, has been approved by the Scientific Steering Committee of VKM.

These are the main conclusions in this opinion, and the main data gaps identified:

Plant health

The most important mycotoxin-producing fungi infecting cereals during the growing season (field fungi) in Norway belong to the genus *Fusarium*. The most important mycotoxin-producing fungi that infect cereal grain during storage (storage fungi) are species of *Aspergillus* and *Penicillium*. The most relevant mycotoxins produced by these fungi are presented in the table:

	Genus	Mycotoxins
Field fungi	*Fusarium*	beauvericin, deoxynivalenol (DON), enniatins, fumonisins, HT-2 toxin, moniliformin, nivalenol, T-2 toxin, zearalenone
Storage fungi	*Aspergillus*	aflatoxins, ochratoxin A
	Penicillium	ochratoxin A

Deoxynivalenol (DON) is the most commonly produced mycotoxin in the field in Norway and other temperate areas. Ochratoxin A is the storage mycotoxin of main concern in Norwegian-produced grain.

DON is found in practically all samples of crude cereal grain, compound feed for animals and cereal food products such as flour, bran and oats flakes. T-2 and HT-2 toxins are also widespread, particularly in unprocessed oats, but also in other grains such as barley.

Data from the last decade show a strong increase in the mean concentration of DON in crude grains of oats. An increased median concentration of DON can also be seen in wheat during this period. During the same period *Fusarium graminearum*, one of the most important producers of DON, has been detected at a much higher level than previously. As opposed to DON, there are no clear trends in the occurrence of the T-2 and HT-2 toxins in crude grains during the last decade.

During the last ten years, *Fusarium* infections of cereal seed have increased by more than 100 % in oats, barley and spring wheat, compared with the three previous decades. Precipitation in the flowering period and during late summer before harvest promotes the occurrence of mycotoxins in cereals. In the last five growing seasons there has been more precipitation than normal in the flowering period for cereals in Norway. If such weather conditions are representative of the future climate, then we can expect significantly increased problems with mycotoxins in cereals in the years to come.

One of the main challenges for cereal cultivation in Norway is an extensive cereal monoculture, with limited use of crop rotation, combined with reduced tillage. Under humid conditions these factors contribute to increased occurrence of mycotoxin-producing fungi. An integrated approach, with a combination of crop rotation, ploughing, growing cereal varieties with the best available resistance, and fungicide treatment at flowering, is a strategy for controlling *Fusarium* head blight and reducing mycotoxin contamination of cereals.

Several reports show lower mycotoxin-content in organic cereals compared to conventional production. Other studies report no difference between the two growing systems. Organic farming is an alternative strategy for limiting *Fusarium*-infection and mycotoxin production in Norwegian cereals.

To improve cropping practices that can reduce the mycotoxin development, there is a need for new and robust insight into the role of the various factors contributing to mycotoxin contamination in cereals, such as host plants, fungal interations, crop rotation, tillage, fertilization, use of pesticides, regional and local topography, land use heterogeneity, distances to lakes/rivers, and soil types and local weather conditions. The effects of mechanical, chemical, or biological treatments of cereal crop residues on survival and development of *Fusarium* inoculum should be further studied. More knowledge on *Fusarium* epidemiology is needed to improve mathematical models to predict the risk of *Fusarium* and mycotoxin development in cereals. It needs to be further clarified whether resistance to DON producers corresponds with resistance to producers of T-2 and HT-2 toxins.

Feed and animal health

Pig is particularly sensitive to mycotoxins compared to other domestic animals. DON reduces performance and animal welfare and there is high risk for such effects due to its considerable occurrence in cereal grains. The current analysis results of DON in feed for pigs show mean concentration around 0.4 mg/kg, a level that may reduce feed intake and performance in the pig barns. However, the effect levels vary considerably in various feeding studies with growing pigs Lowest Observed Adverse Effect Levels (LOAELs) from 0.35 to 2 mg/kg.

The risk for adverse effects of most other mycotoxins (T-2 and HT-2 toxins, zearalenone, fumonisins and aflatoxins) is regarded as low to negligible in pigs.

For ochratoxin A, EFSA has derived a LOAEL for nephrotoxicity at 0.2 mg/kg diet which is about 1000 times higher than a mean level of ochratoxin A in cereals in Norway. However, more recent findings show reduced sperm quality in boars at 0.003 mg/kg as well as reduced

performance of growing pigs at 0.025 mg/kg diets indicating a moderate to high risk for effects.

The sensitivity for mycotoxins in poultry may vary considerably for different species and production forms but is in general less than for pigs.

The sensitivity for mycotoxins in ruminants is generally low. However, disturbed rumen metabolism due to e.g. excessive amounts of compound feed may increase the sensitivity to mycotoxins. Horses show moderate sensitivity to a range of mycotoxins. As the major horse diet is roughage and horse owners show more care with using oats for horses, the exposure of mycotoxins via cereals is limited.

The effect data base for mycotoxins in rabbit is scarce but available data indicates a moderate sensitivity to a range of mycotoxins.

The knowledge on effects of mycotoxins in dog and cat is rather low, but the risk seems in general to be low.

For fish the risk for effects is considered as negligible based on the maximum theoretical concentrations of the mycotoxins in the feed for salmon and rainbow trout and the effect data available. However, the risk for aflatoxin effects in rainbow trout is considered moderate. No effect data for aflatoxins are available for Atlantic salmon.

VKM has pointed out several data gaps related to animal health. Amongst the most important ones is to gain better knowledge of factors influencing the broad range for level of critical effect of DON in pig and the indication of increased sensitivity to DON, T-2/HT-2 and ochratoxin A and possibly other mycotoxins in intensively fed ruminants. Occurrence data for various mycotoxins in compound feed for most animal species and in feed ingredients as maize should be provided. More knowledge on mycotoxins in stored feed for terrestrial and aquatic animals is also necessary.

Food and human health

The dietary exposure to DON and zearalenone, respectively, were estimated based on occurrence data in Norwegian cereal products and consumption data from national dietary surveys. VKM chose to use the lowest and highest mean mycotoxin concentrations of the four years (2008-2011) for each of the four flour products: sieved wheat flour, milled wheat flour, wheat bran and oat flakes. The exposures were compared with the internationally derived tolerable daily intakes (TDIs) for the respective toxins.

The contribution from animal–derived food items to the dietary intake of mycotoxins is low and of little significance compared to the intake from plant sources.

DON is the main mycotoxin of concern in Norwegian grain. It is the most prevalent mycotoxin in Norwegian grain and is present in virtually all samples of flour and oat flakes.

The estimated mean and high (95-percentile) exposures to DON in years with low mean concentration of DON in the flour, respectively, were in the range of, or exceeded the TDI by almost 2 times in 1-year-old infants and 2-year-old children. In years with high mean DON concentration, the high (95-percentile) exposures exceeded the TDI up to 3.5 times for 1-, 2-, 4- and 9-year-olds. VKM concludes that exceeding the TDI at mean or high exposures to DON in infants and children is of concern, although the TDI is not a threshold for toxicity. The estimated dietary intakes of DON in adolescence and in the adult population are equal to

or below the TDI and are therefore not a health concern. Acute exposure to DON is of no concern.

The estimated intakes of zearalenone are below the TDI for all age groups. Exposure to zearalenone and nivalenol is of no concern for all age groups.

The dietary intakes of sum of T-2 and HT-2 toxins could not be estimated due to the high number of samples below the limit of detection. Scenarios were made to illustrate the potential intakes of sum of T-2 and HT-2 toxins.

The scenarios are considered to represent an overestimation. These scenarios indicate that the dietary intake of the sum of T-2 and HT-2 toxins in 1- and 2-year-olds may exceed the TDI, while the 4-year-olds with high exposure have an intake in the range of the TDI. According to the exposure scenarios, the exposures to the sum of T-2 and HT-2 toxins in 9- and 13-year-olds are below the TDI. Furthermore, both the mean and high exposures in adults are below the TDI. VKM concludes that according to the performed exposure scenarios, the dietary intake of the sum of T-2 and HT-2 is potentially of concern for the youngest age groups.

The dietary exposure to nivalenol in Norway could not be estimated due to a high number of samples that were below the limit of detection. The scenarios for intake of nivalenol were below the TDI for all age groups. The intake of nivalenol is of no concern for all age groups.

The mycotoxins fumonisins, ochratoxin A and aflatoxins in grain products are considered to be of no concern.

No assessment of the emerging mycotoxins enniatins, beauvericin and moniliformin in grain could be made due to the lack of occurrence data, as well as toxicity data of these toxins. VKM recognizes the presence of moniliformin, enniatins and beauvericin in Norwegian grains, which potentially might be of risk for human health.

Several data gaps have been identified. The occurrence data for the most important mycotoxins, DON and the sum of T-2 and HT-2 toxins, in Norwegian food products are scarce. A more systematic surveillance with optimized methods should be performed, especially focussing on products with high wheat and oat contents. Further, knowledge of combined exposure of mycotoxins is limited. The consumption of maize-based products and rice in Norway has increased in recent years, substituting potatoes that are less used. Thus, Norwegian consumers might be exposed to maize-specific mycotoxins at higher extent than before. The monitoring of maize-based products for toxic metabolites such as aflatoxin, zearalenone and fuminosin, but also for DON, appears to be urgently needed.

Concluding remarks

In cereals, the most important mycotoxin-producing fungi are *Fusarium* species, which infect cereals during the growing season and cause yield loss and mycotoxin contamination of the grain. With the current occurrence of mycotoxins in grains in Norway VKM identified deoxynivalenol (DON) as the main mycotoxin of concern for human and animal health.

- Pig is in comparison to other domestic animals, particularly sensitive to DON. At current levels of DON in pig feed there is a high risk that DON reduces the pigs´ performance and welfare.

- For humans, the calculated exposure shows that infants and children exceed the TDI for DON from consumption of flour and oat flakes. Exceeding the TDI is of concern, although the TDI is not a threshold for toxicity.

Because of these findings and since cereal grain is an important feed and food ingredient VKM is of the opinion that steps to reduce levels of DON in feed and foods are warranted.

- VKM identified a number of agricultural factors influencing the *Fusarium* infection rate and consequently the mycotoxin production such as crop rotation, ploughing, resistance of cereal varieties, and fungicide treatment at flowering.

- VKM also notes that given the content of mycotoxins in flour for use in Norway attention should also be paid to the content of mycotoxins in imported grains.

- Special attention should be paid to cereals intended for infants and children.

Although there is a large annual variation in the occurrence of DON, VKM notes that during the last decade, parallel to an increased precipitation during the flowering period, there has been a strong increase in the infection rate and occurrence of DON in oats and wheat. Future climate change in Norway, with increased temperature and possibly increased precipitation during the flowering period, would imply a significant increase in problems with *Fusarium* infection and occurrence of mycotoxins in cereals in the years to come.
VKM also identified important gaps in knowledge and data with respect to plant production, especially concerning *Fusarium* infection rates and protective measures against infection. Also, data on occurrence and data on toxicity of mycotoxins, particularly in some domestic animal species and for emerging toxins, are lacking.

Norsk sammendrag

Korn kan infiseres av muggsopp som forurenser kornet med sekundære metabolitter som kalles mykotoksiner (muggsoppgifter). Mykotoksiner i korn kan utgjøre en helserisiko for både mennesker og dyr. Over tid kan inntak av mykotoksinene som finnes i norsk korn blant annet føre til nedsatt immunforsvar, redusert vekst og effekter på reproduksjon. Det er derfor satt grenseverdier for mykotoksiner i næringsmidler og anbefalte grenseverdier i fôr i EØS. Regelverksutviklingen er løpende og ny kunnskap kan gi nye og/eller reviderte grenseverdier.

I lys av stadig ny kunnskap om toksisiteten til mykotoksiner og en økt forekomst av mykotoksiner i korn de seneste årene, ba Mattilsynet i september 2010 Vitenskapskomiteen for mattrygghet (VKM) om en risikovurdering av mykotoksiner i korn.

Risikovurderingen tar for seg muggsopp og mykotoksiner som finnes i korn og kornprodukter på det norske markedet (norskprodusert og importert, mat og fôr, inklusive fiskefôr). I tillegg til å risikovurdere mykotoksinene med hensyn til humanhelse og dyrehelse gjennomgår VKM ulike faktorer som påvirker muggsoppdannelsen og mykotoksinproduksjonen i korn, både under vekst ute i felt og under lagring.

Risikovurderingen er vitenskapelig bakgrunnsmateriale for risikohåndtering av jordbrukspraksis, produksjon av mat og fôr og for å vurdere om det er behov for å endre grenseverdier i mat og fôr. I tillegg til det løpende regelverksutviklingsarbeidet i EU, ønsker Mattilsynet å kunne gi innspill til næringen om risiko på dette feltet.

For å besvare oppdraget fra Mattilsynet har VKM oppnevnt tre prosjektgrupper bestående av medlemmer av VKM og eksterne eksperter som dekker fagområdene for plantehelse, fôr og dyrehelse, mat og humanhelse. Rapportene fra de tre prosjektgruppene har blitt vurdert og godkjent av relevante faggrupper i VKM. Endelig uttalelse, risikovurderingen av mykotoksiner i korn, er vurdert og godkjent av VKMs hovedkomité.

Nedenfor oppsummeres hovedkonklusjonen og de viktigste kunnskapshullene i risikovurderingen:

Plantehelse

De viktigste mykotoksinproduserende muggsoppene som infiserer kornet i vekstsesongen (feltsopp) i Norge tilhører slekten *Fusarium*. På lager er arter i slektene *Aspergillus* og *Penicillium* de viktigste mykotoksinprodusentene.

Tabellen viser de mest relevante mykotoksinene produsert av disse muggsoppene:

	Slekt	Mykotoksiner
Feltsopp	*Fusarium*	beauvericin, deoksynivalenol (DON), enniatiner, fumonisiner, HT-2 toksin, moniliformin, nivalenol, T-2 toksin, zearalenon
Lagringssopp	*Aspergillus*	aflatoksiner, okratoksin A
	Penicillium	okratoksin A

Deoksynivalenol (DON) er det vanligste mykotoksinet som produseres i felt i Norge og i andre tempererte strøk. Okratoksin A er det lagrings-mykotoksinet vi er mest bekymret for i norsk korn.

DON forekommer i nesten alle prøver av norsk korn, kraftfôr og kornprodukter til mat som mel, kli og gryn. Toksinene T-2 og HT-2 er også vanlige, spesielt i ikke-prosessert havre, men også i bygg.

Data fra siste tiårsperiode viser en kraftig økning i gjennomsnittlig konsentrasjon av DON i havre. I samme periode har det også vært økt konsentrasjon av DON i hvete. Forekomsten av

muggsoppen *Fusarium graminearum,* en av de viktigste DON-produsentene, har også økt det siste tiåret. Men i motsetning til DON har forekomsten av T-2 og HT-2 ikke økt i perioden.

Fusarium-infeksjonen i havre, bygg og vårhvete har økt med mer enn 100 % de siste ti år sammenlignet med de forrige tretti år. Nedbør i blomstringsperioden og på ettersommeren før høsting fremmer forekomsten av mykotoksiner i korn, og nedbøren i blomstringen har vært over normalen de siste fem vekstsesonger. Dersom en slik værtype er representativt for framtidig klima, kan vi vente økte problemer med mykotoksiner i korn i åra fremover.

Monokultur av korn, med manglende vekstskifte kombinert med redusert jordarbeiding, er en av de store utfordringene i norsk korndyrking. I et fuktig klima er dette faktorer som bidrar til økt forekomst av mykotoksinproduserende sopp. En integrert produksjon som kombinerer vekstskifte, pløying, dyrking av mest mulig resistente sorter og sprøyting med soppmidler i blomstringen er en strategi for å bekjempe *Fusarium*-angrep og redusere mykotoksininnholdet i kornet.

Det er flere rapporter som viser at økologisk dyrka korn har lavere innhold av mykotoksiner enn konvensjonelt dyrket korn. Andre undersøkelser viser samme nivå ved de to dyrkingssystemer. Økologisk produksjon er en alternativ strategi for å begrense *Fusarium*-angrep og mykotoksinproblemene i norsk korn.

Bedre dyrkingspraksis for å redusere mykotoksininnholdet i korn krever ny og bedre innsikt i betydningen av forskjellige faktorer som påvirker mykotoksinproduksjonen i korn. Eksempler på faktorer som bør undersøkes bedre er: Samspill mellom vertplante og sopp, vekstskifte, jordarbeiding, gjødsling, bruk av plantevernmidler, regional og lokal topografi og arealbruk, avstand til vassdrag og innsjøer, jordtype og mikroklima. Effekter av mekanisk, kjemisk og biologisk behandling av planterester på *Fusarium*-smitte bør studeres nærmere. Det er behov for bedre innsikt i *Fusarium*-epidemiologi for å kunne videreutvikle matematiske modeller som predikerer risiko for *Fusarium*-angrep og mykotoksiner i korn. Det er også viktig å undersøke om resistens mot DON-produserende muggsopp også gir resistens mot muggsopp som produserer T-2 og HT-2 toksin.

Dyrehelse

Griser er spesielt sensitive for mykotoksiner sammenlignet med andre husdyr. DON reduserer ytelse og dyrevelferd, og det er høy risiko for slike effekter på grunn av betydelige forekomster i korn. Analyseresultater av DON i fôr til gris viser gjennomsnitts - konsentrasjon rundt 0,4 mg/kg som er et nivå som kan redusere fôrinntak og ytelse i grisehuset. Men nivåene som gir effekt varierer betydelig i ulike fôrings- studier med slaktegris (LOAELs - de laveste observerte skadelige effektnivåer - 0,35 til 2 mg/kg).

Risikoen for effekter av de fleste andre mykotoksiner (HT-2 og T-2-toksin, zearalenon, fumonisin og aflatoksiner) regnes som lav til ubetydelig for gris.

For okratoksin A har EFSA utledet en LOAEL for nyretoksisitet på 0,2 mg/kg fôr som er ca. 1000 ganger høyere enn gjennomsnittlig nivå av okratoksin A i korn i Norge. Nyere funn viser redusert sædkvalitet hos råner ved nivåer på 0,003 mg/kg, så vel som redusert ytelse hos slaktegris ved 0,025 mg/kg, noe som indikerer en moderat til høy risiko for effekter.

Sensitiviteten for mykotoksiner hos fjørfe kan variere betydelig for forskjellige arter og produksjonsformer, men sensitiviteten er generelt lavere hos fjørfe enn hos griser.

Sensitiviteten for mykotoksiner hos drøvtyggere er generelt lav. Men forstyrret vom-metabolisme på grunn av for eksempel store mengder kraftfôr kan øke opptaket og dermed

risikoen for effekter av mykotoksiner. Hest viser moderat senstitivitet for en rekke mykotoksiner. Siden hesten spiser mest høy, og hesteeierne er blitt forsiktige med å gi havre, er eksponeringen av mykotoksiner via korn begrenset.

Effektstudier av mykotoksiner i fôret til kanin er mangelfull, men tilgjengelige data indikerer en moderat sensitivitet for en rekke mykotoksiner.

Kunnskap om effekter av mykotoksiner i fôret til hund og katt er relativt lav, men risikoen synes generelt å være lav.

For fisk er risikoen for effekter ansett som ubetydelig basert på tilgjengelige effektstudier og maksimale teoretiske konsentrasjoner av mykotoksiner i fôr til laks og regnbueørret. Imidlertid synes sannsynligheten for effekter av aflatoksin i fôret til regnbueørret å være moderat. Ingen effektstudier for aflatoksiner i fôret til atlantisk laks er tilgjengelig.

VKM har påpekt flere kunnskapshull knyttet til mykotoksiner og dyrehelse. Blant de viktigste er å få bedre kunnskap om faktorer som påvirker effektnivået av DON i fôret til gris, samt at det ser ut til å være økt sensitivitet hos drøvtyggere for DON, HT-2/T-2 og okratoksin A og eventuelt andre mykotoksiner ved intensiv fôring. Forekomstdata for ulike mykotoksiner i kraftfôrblandinger til de fleste dyrearter, og i fôringredienser som mais bør skaffes. Det er også viktig å få bedre kunnskap om mykotoksiner i lagret fôr i fjøs og oppdrettsanlegg.

Humanhelse

Beregninger av hvor mye man får i seg av mykotoksinene DON og zearalenon gjennom kosten ble basert på forekomstdata i norsk mel og inntak av mat og drikke i de nasjonale kostholdsundersøkelsene. VKM valgte å bruke den laveste og høyeste gjennomsnittlige konsentrasjonen av mykotoksiner for siktet hvete, sammalt hvete, hvetekli og havregryn i løpet av en 4 års periode (2008-2011). Mengdene man får i seg av hvert toksin (dvs. eksponeringen) ble sammenlignet med de internasjonalt vedtatte tolerable daglige inntakene (TDI) for toksinene.

Mykotoksiner kommer hovedsakelig fra vegetabilske kilder i kosten. Til sammenligning er bidraget fra animalske produkter lavt og av liten betydning.

Det viktigste mykotoksinet som gir grunn til bekymring i norsk korn er DON. DON er det mest vanlig forekommende mykotoksinet i norsk korn og er til stede i nesten alle prøver av mel og havregryn.

I år med lav gjennomsnittlig DON konsentrasjon i melet var de estimerte gjennomsnittlige og høy (95-persentil) eksponeringene for DON blant 1 og 2 åringer henholdsvis i samme størrelsesorden eller nesten to ganger høyere enn TDI. I år med høy gjennomsnittlig DON konsentrasjon i melet overskred høyeksponentene (95-persentilene) blant 1, 2, 4 og niåringer TDI med opptil 3,5 ganger. VKM konkluder med at overskridelse av TDI ved gjennomsnittlig og høyeksponering for DON blant barn er av bekymring, selv om TDI ikke er en terskel for toksisitet. De beregnede kostinntakene av DON blant ungdom og voksne tangerer eller er lavere enn TDI og representerer derfor ingen helsebekymring. Det er ingen bekymring tilknyttet akutt eksponering for DON.

De beregnede kostinntakene av zearalenon er lavere enn TDI i alle aldersgrupper og eksponering for zearalenon representerer ingen bekymring.

Kostinntaket av summen av T-2 og HT-2 toksiner kunne ikke beregnes ettersom innholdet var under deteksjonsgrensen i et stort antall av prøvene. Det ble gjort scenarioberegninger for å

illustrere mulige inntak av summen av T-2 og HT-2 toksinene. Scenarioene vurderes til å representere en overestimering. De antyder at kostinntaket av summen av T-2 og HT-2 toksiner blant 1 og 2 åringer kan overskride TDI, mens høyeksponentene blant 4 åringer har et inntak som tangerer TDI. Scenarioene viser videre at eksponeringen blant både niåringer, 13 åringer og voksne er under TDI. VKM konkluderer at i henhold til scenarioberegningene kan kostinntaket av summen av T-2 og HT-2 toksiner muligens representere en bekymring blant de yngste aldersgruppene.

Kostinntaket av nivalenol kunne ikke beregnes ettersom innholdet var under deteksjonsgrensen i et stort antall av prøvene. Scenarioberegninger viste at inntaket av nivalenol er under TDI for alle aldersgrupper og representerer ingen bekymring.

Mykotoksinene fumonisin, okratoksin A og aflatoksin er ikke vurdert til å være av bekymring i kornprodukter.

Mykotoksinene enniatiner, beauvericin og moniliformin kunne ikke vurderes grunnet mangel på både forekomst- og toksisitetsdata. VKM mener at tilstedeværelsen av disse mindre kjente toksinene i norsk korn muligens kan representere en bekymring.

Det er identifisert mange kunnskapshull. Det er mangelfulle forekomstdata for de mest vanlige mykotoksinene, DON og summen av T-2 og HT-2 toksiner, i norske matprodukter. Mer systematisk overvåking med optimaliserte metoder bør utføres, og med særlig fokus på produkter med høyt innhold av hvete og havre.

Det er begrenset kunnskap om konsekvensene av eksponering for flere mykotoksiner samtidig (kombinert eksponering).

I de senere årene kan det tyde på at inntaket av maisbaserte produkter og ris har økt i Norge. Norske konsumenter kan derfor bli eksponert for maisspesifikke mykotoksiner i høyere grad enn tidligere. Det synes å være et stort behov for overvåking av maisbaserte produkter med hensyn til aflatoksin, zearalenon og fumonisin, men også for DON.

Avsluttende bemerkninger

De viktigste produsentene av mykotoksiner i korn er sopper i slekten *Fusarium*. Disse infiserer kornet i løpet av vekstsesongen og forårsaker redusert utbytte i tillegg til at kornet forurenses med mykotoksiner. Ut i fra dagens forekomst av mykotoksiner i norsk korn fastslår VKM at det er deoksynivalenol (DON) som utgjør størst bekymring for menneskers og dyrs helse.

- Sammenliknet med andre husdyr er grisen spesielt sensitiv for DON. Med dagens nivåer av DON i grisefôr er det en høy risiko for at DON reduserer grisens ytelse og velferd.

- En beregning av eksponeringen av mennesker gjort ut i fra bruk av mel og havregryn viser at spedbarn og barn overstiger TDI (tolerabelt daglig inntak) for DON.

På grunn av disse funnene, og siden korn er en viktig ingrediens i fôr og mat, er VKM av den oppfatning at det er nødvendig å iverksette tiltak for å redusere nivåene av DON i fôr og mat.

- VKM identifiserte en rekke faktorer ved norsk korndyrking som påvirker nivået av *Fusarium*-infeksjon og dermed også produksjonen av mykotoksiner i kornet. Slike

faktorer er vekstskifte, pløying, resistens hos kornsorter, og sprøyting med soppmidler i blomstringen.

- VKM påpeker også at man på grunn av innholdet av mykotoksiner i mel som brukes i Norge bør være oppmerksom på nivået av mykotoksiner i importert korn.

- Spesiell oppmerksomhet bør rettes mot korn beregnet for spedbarn og barn.

Selv om det er store årlige variasjoner i forekomsten av DON, påpeker VKM at det i løpet av det siste tiåret, parallelt med økt nedbør i blomstringen, har vært en økning i *Fusarium*-infeksjon og forekomst av DON i havre og hvete. Fremtidige klimaendringer i Norge med økende temperatur og muligens økte nedbørsmengder i løpet av blomstringen kan føre til betydelig økte problemer med *Fusarium*-infeksjon og forekomst av mykotoksiner i korn i årene som kommer.

VKM har også identifisert viktige kunnskapshull og mangel på data med hensyn til planteproduksjon, spesielt når det gjelder infeksjonsrate og beskyttelsestiltak mot *Fusarium*. Også data om forekomsten og giftigheten av mykotoksiner, særlig for enkelte husdyr og noen av de mindre kjente toksinene, mangler.

Keywords

Mycotoxins, cereal grain, fungi, Deoxynivalenol (DON), T-2 and HT-2 toxins, *Fusarium*, *Aspergillus*, *Penicillium*, plant health, feed, animal health, temperal trends, tolerable daily intakes (TDI), exposure, occurrence data, climate change, storage conditions

Some definitions

A **plant variety** is a plant grouping within a single botanical taxon of the lowest known rank, which grouping can be

- defined by the expression of the characteristics resulting from a given genotype or combination of genotypes,
- distinguished from any other plant grouping by the expression of at least one of the said characteristics, and
- considered as a unit with regard to its suitability for being propagated unchanged.

A **cereal variety** is a genetically very homogenous product from many generations of backcrossing and selection to give uniform agronomic characters such as height, disease resistance, maturity, yield and quality.

Abbreviations

ARfD (Acute Reference Dose)
EFSA (the European Food Safety Authority)
FEB (*Fusarium* ear blight)
FHB (*Fusarium* head blight)
FDK (*Fusarium* damaged kernels)
JECFA (Joint WHO/FAO Expert Committee on Food Additives)
LOAEL (Lowest Observed Adverse Effect Level)
LOD (limit of detection)
LOEL (Lowest Observed Effect Level)
NOAEL (No Observed Adverse Effect Level)
NOEL (No Observed Effect Level)
PMTDI (Provisional Maximum human Tolerable Daily Intake)
TDI (Tolerable Daily Intake)
VKM (the Norwegian Scientific Committee for Food Safety)

Type A trichothecenes
DAS (diacetoxyscirpenol)
HT-2 (HT-2 toxin: 15-Acetoxy-3α,4β-dihydroxy-8α-(3-methylbutyryloxy)-12,13-epoxytrichothec-9-ene)
T-2 (T-2 toxin: (2α,3α,4β,8α)-4,15-bis(acetyloxy)-3-hydroxy-12,13-epoxytrichothec-9-en-8-yl 3-methylbutanoate)

Type B trichothecenes
DON (deoxynivalenol)
3-acetyl DON (3-acetyl-deoxynivalenol)
15-acetyl DON (15-acetyl-deoxynivalenol)
FUSC (fusarin C)
NIV (nivalenol)

Other mycotoxins
AFB1 (aflatoxin B1)
AFB2 (aflatoxin B2)
AFG1 (aflatoxin G1)
AFG2 (aflatoxin G2)
AFM1 (aflatoxin M1)
AFM2 (aflatoxin M2)
BEA (beauvericin)
ENN (enniatins)
FUM (fumonisins)
MON (moniliformin)
OTA (ochratoxin A)
ZON (zearalenone), in the literature also called ZEN or ZEA

Contents

Background

Cereals can become infected by fungi that then contaminate the grain with toxic secondary metabolites, called mycotoxins. Several species in the genus *Fusarium* infect cereal grain in the field during the period from flowering to harvest (field fungi), while fungi in the genera *Aspergillus* and *Penicillium* infect the grain and produce mycotoxins during storage (storage fungi).

Cereal grain is one of the main components of food and feed, and mycotoxins in cereal grain can pose a risk to both human and animal health. Long-term intake of mycotoxins found in Norwegian cereal grains can, among other effects, cause reduced immune responses, reduced growth and affect reproduction. In the European Economic Area (EEA), maximum limits (MLs) have been determined for mycotoxins in food and recommended for mycotoxins in feed. The Legislation is continuously under revision as new knowledge can lead to new or revised MLs.

In Norway, cereals are grown up to their northern limit. About 80 % of the cereal production is concentrated in the south-eastern counties, while of the remainder the majority is located to the Trøndelag counties. Most growers practise cereal monoculture; barley is grown on about half of the 300 000 ha of cereals, the rest is equally divided between wheat and oats. During most of the last 10 years, domestic production has provided 70-80 % of the bread wheat. Barley and oats are mainly used for animal feed, and only a minor part of the production is for human consumption.

During the last few years an increased occurrence of mycotoxin-producing fungi, especially from the genera *Fusarium*, in which the most important mycotoxin-producing field fungi in Norway belong, has been found in cereals. The cereal harvest during 2008-2011 gave particular rise to concern, as the conditions were particularly favourable for fungal infection and epidemic development. The increase in occurrence of mycotoxin-producing fungi seem to be caused by increased precipitation during the growing season and/or changes in agricultural practices, like reduced tillage.

In the light of constantly new knowledge on toxicity and the rise in the level of mycotoxin contamination during recent years, in a letter dated September 21st 2010 the Norwegian Food Safety Authority requested the Norwegian Scientific Committee for Food Safety (VKM) to undertake a risk assessment on mycotoxins in cereal grain. The risk assessment should cover the fungi and toxins present in grain and grain-related products available on the Norwegian market (both domestic and imported, food and feed, including fish feed). In addition to an evaluation concerning human and animal health, the risk assessment should consider the different factors affecting fungal infection and mycotoxin production in cereal grain, both in the field and during storage.

The risk assessment will be used as a scientific background for risk management related to agricultural practices, food and feed production and to modify MLs in food and feed. In addition to contributing to the constant development of regulations in the EU, the Norwegian Food Safety Authority will communicate the risk to producers in agriculture and industry.

Several significant international assessments regarding toxicity and risks from consumption of mycotoxin-contaminated products have been performed, among others by the Joint WHO/FAO Expert Committee on Food Additives (JECFA) and by the European Food Safety Authority (EFSA). In Norway, both the Norwegian Institute of Agricultural and Environmental Research and the Norwegian Veterinary Institute have performed several studies on mycotoxins. The Norwegian Food Safety Authority carries out an annual

surveillance and control programme in which the levels of mycotoxins in food and feed are monitored, and several reports on their occurrence in domestic and imported products are available. Reports of EU data are also available. All documents provided by the Norwegian Food Safety Authority are listed in the reference chapter.

In order to answer the request from the Norwegian Food Safety Authority, VKM appointed three project groups consisting of both VKM members and external experts, covering the different topics of food and human health, feed and animal health, and plant health. The report from the project group on food and human health has been evaluated and approved by the Panel on contaminants. The report from the project group on feed and animal health has been evaluated and approved by the Panel on animal feed. The report from the project group on plant health has been evaluated and approved by the Panel on plant health. The final opinion, the current document, has been approved by the Scientific Steering Committee of VKM.

Terms of reference

The Norwegian Food Safety Authority requests VKM to undertake a risk assessment of mycotoxins in grain. The risk assessment should cover the fungi and toxins present in grain and grain related products that are available on the Norwegian market (both domestic and imported, food and feed, including fish feed).

The assignment will include several disciplines, and we would suggest that the terms of reference are divided into plant health, feed/animal health and food/human health.

Plant health:

- What are the mycotoxin-producing fungi in Norwegian cereals? Describe the occurrence of the different fungi and mycotoxins.
- Which cereal crops are most prone to mycotoxin contamination? Are there varietal differences in mycotoxin contamination?
- Which mycotoxins are produced in the field and which are produced during storage? Have there been variations year by year or are there regional differences in mycotoxin occurrence? If so, describe the variation.
- What resistant varieties are available for Norwegian farmers? Can growing of resistant varieties reduce mycotoxin contamination of domestic cereals?
- Can use of pesticides be a way to reduce mycotoxin contamination of domestic cereals?
- What has been the effect of changes in agricultural practices (such as reduced autumn ploughing) and other changes in soil tillage on occurrence of mycotoxin-producing fungi? Does irrigation influence the probability of infection?
- How do climatic factors influence the probability of fungal infection and mycotoxin contamination of cereal grain? Will expected climatic changes in Norway affect occurrence of mycotoxins?
- Are there differences in probability of mycotoxin contamination between conventional and organic cereal production? If yes, what are the differences, and what may be the explanation?

- How do storage conditions influence fungal growth and postharvest mycotoxin contamination?

Feed and animal health:

- Assess whether the current exposure for mycotoxins in feed represent a potential risk for animals (including fish).
- Feed decontamination. Current EEA legislation allows the use of simple physical treatment such as cleaning and dehulling to reduce mycotoxin content in feed. There are however research ongoing on additives in feed to detoxify mycotoxins, such as mycotoxin binders. What are the potential risks related to these products?

Food and human health:

- Assess whether the current exposure from mycotoxins in grain represent a potential risk for consumers (in general and potential vulnerable groups) in Norway.
- Which risk could carry-over of mycotoxins from animals (including fish) represent for human health?

Assessment

1 Biology of mycotoxin-producing fungi in cereals

The most important mycotoxin-producing field fungi on cereals in Norway belong to the genus *Fusarium*. This genus name is used for the asexual reproductive stage (anamorph stage) when the fungi produce conidia spores. *Fusarium graminearum*, one of the most important species, commonly produces both conidia and sexual spores (teleomorph stage with ascospores), while for others the sexual stage is rare. The number of *Fusarium* species is currently around 90 and many of them are only known in the asexual stage.

These pathogens can cause *Fusarium* seedling blight, root rot and foot rot, as well as *Fusarium* head blight, also known as ear blight and scab. Seedling blight and infection on roots and lower parts of the straw may reduce the plant density and cause yield losses.

Fusarium infection of the head, and fungal growth towards harvest, may result in production of mycotoxins in the grain. Cereals are most susceptible to *Fusarium* infection at flowering. Humid weather during that critical period favours the development of *Fusarium* head blight . If the grain humidity is too high during storage, fungal growth and mycotoxin accumulation will continue.

We have chosen *F. graminearum* as an example of a fungus with both sexual and asexual spore stages to describe the life cycle and biology of fungal infection and colonisation in cereals in the field. The disease cycle is illustrated in Figure 1, and is further described in the following Sections 1.1 and 1.2.

1.1 Inoculum sources and survival

Between growing seasons, *Fusarium* head blight pathogens survive as saprophytes on host residue or other plant debris on the soil surface and in the upper soil layer (Figure 1). Non-decomposed residues from the previous crop are considered the most important inoculum sources. *Fusarium* head blight severity and mycotoxin contamination have been shown to increase with increased amounts of residues left by the preceding crop. *Fusarium* spp. are also seed-borne.

The ability to survive as saprophytes differs among *Fusarium* spp. The cereal pathogens *F. graminearum* and *F. culmorum* are aggressive plant pathogens and survive in dead leaves and straw. When plant residues have been broken down after two or three years, they are unable to live as saprophytes in soil. *Fusarium avenaceum* has a wide host range, and can cause disease in cereals, potato and legumes. The fungus is predominantly soil-borne (Leslie and Summerell 2006). The saprophytic abilities of the weak plant pathogens *F. langsethiae, F. sporotrichioides* and *F. poae* are not well known.

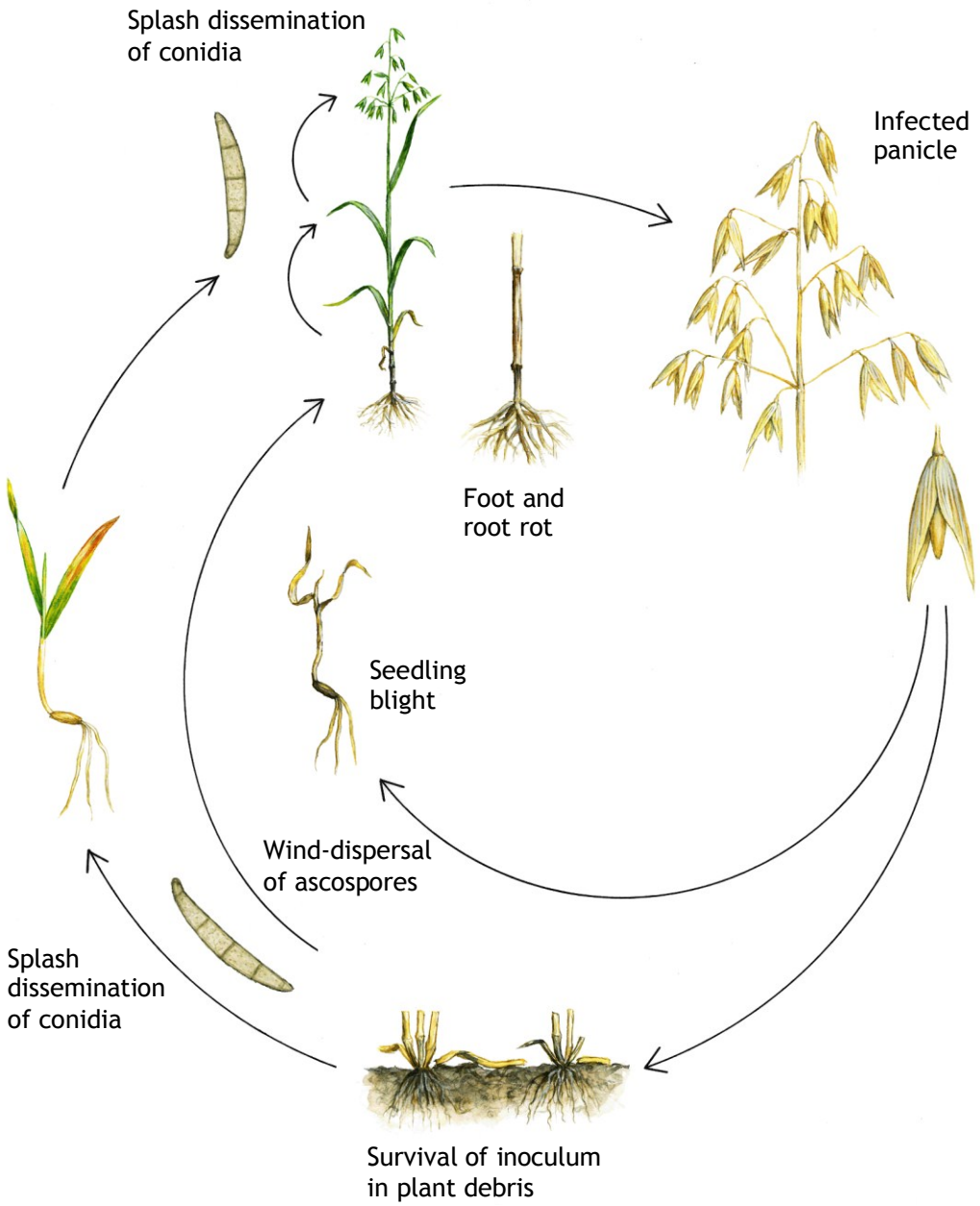


Figure 1: Disease cycle of *Fusarium* head blight in oats caused by *Fusarium graminearum* (drawing by Hermod Karlsen, from Brodal et al. 2009).

1.2 Dispersal and disease development

Under favourable conditions, sporulation occurs on infected plant debris. Conidia are mainly disseminated by water splash (Figure 1). For *F. graminearum*, the main *Fusarium* head blight pathogen in North America, and, in recent years, also in Europe, the sexual stage seems to be important for dispersal. Ascospores are discharged from fruiting bodies in spring and during the summer, and carried by air over long distances. Their importance in spreading the disease in Norway has not yet been documented.

Infected seeds introduce the pathogens to *Fusarium* head blight-free fields. Seed infection is also a pathway for dispersal over long distances, i.e., between countries and continents. Infected seed may result in reduced germination, poor plant establishment and blighted

seedlings in the field. Infected plant parts developed from infected seed can become a source of *Fusarium* head blight inoculum in future years. Seedlings that survive infection may develop foot rot at a later stage, weakening the straw and causing lodging in the field. Sporulation may occur at the stem bases and nodes. Conidia may then be splashed by rain to reach the ears/head of the plant (Figure 1).

Infection of cereal grain (kernels) by water-splashed conidia or air-borne ascospores starts at the flowering stage (anthesis) and continues towards harvest. During anthesis, conidia or ascospores germinate in free water on the glumes and other floral parts. In the initial stage, anthers and pollen may serve as a food-base for the pathogen. In oats, the infection starts at the apices of the florets and progresses down to the basal floral parts. It penetrates the internal surface of palea, lemma and caryopsis (Tekle et al. 2012). Secondary infection from airborne conidia may be possible from early disease outbreaks, but *Fusarium* head blight seems to be largely monocyclic (Dill-Macky 2010).

1.3 Symptoms of *Fusarium* head blight

In wheat, *Fusarium* head blight can be seen as bleaching of one or more spikelets on emerged heads after flowering. In barley, small brown or water-soaked spots on the glumes and the rachis can be symptoms of *Fusarium* infection. If the rachis is infected, all tissues above that point may be killed. Under wet conditions the fungus develops and the whole head may become infected. Bleaching and discoloration gives an early-ripened appearance of infected heads. Towards grain maturity, orange or pink masses of conidia (sporodochia) may be seen on or at the base of diseased spikelets.

Fusarium infection may results in shriveled and discolored kernels, sometimes with a chalky white appearance or sterile florets. Barley kernels may appear grayish brown toward the base. Many of the shriveled, light seeds are lost during combining, although some may remain in the harvested grain. Those seeds remaining are referred to as *Fusarium* damaged kernels (FDK). Kernels can be infected later in their development, but without visible symptoms. However, the kernels may still be contaminated with mycotoxins.

In oats, the symptoms are often more insignificant, however, severely infected fields may get a prematurely bleached appearance, and during moist weather towards grain maturity orange or pink masses of conidia (sporodochia) may be seen on or at the base of diseased spikelets in the peduncle. Despite the lack of visual symptoms high levels of mycotoxins have been detected in oats grain.

2 An overview of the mycotoxins and mycotoxin-producing fungi present in cereal grain in Norway

Cereal grain may become contaminated by mycotoxins both in the field and during storage. A short overview of the different mycotoxins found in cereals is provided in Section 2.1. Occurrence of the different mycotoxins in food and feed is presented in Chapter 6. Regional and temporal variations in levels of mycotoxins and mycotoxin-producing fungi are presented in Chapter 3. Hazard characterisations of the mycotoxins are given in Chapter 9.

The most important toxigenic fungi infecting cereal grain during the growing season belong to the genus *Fusarium*. These *Fusarium* species, and some other relevant field fungi, are listed in Section 2.2, along with a list of the toxins that they produce.

Fungi in the genera *Aspergillus* and *Penicillium* may develop in grain stored at humidities above 14-15 %. Some species in these two genera produce mycotoxins; see Section 2.3 on storage fungi and their toxins.

2.1 Mycotoxins present in cereal grain

2.1.1 Trichothecenes

Deoxynivalenol (DON) and its derivatives are the most prevalent trichothecenes with a worldwide occurrence in most cereals (JECFA 2001). In Norway, DON is frequently found in oats, wheat and barley (Tables 8-10) and is mainly produced by *F. culmorum and F. graminearum* (Table 1). Data on Norwegian rye is very limited. DON is also found in almost all samples of processed cereals intended for human consumption such as wheat flour, wholemeal wheat flour, bran and oat flakes.

T-2 and HT-2 toxins are produced by several *Fusarium* species (Table 1). The toxins are widespread and have been found in all major wheat- barley- and oats-producing parts of the world. T-2 and HT-2 toxins normally occur together, and are found most frequently and in highest concentrations in oats, both in Norway and in other parts of the world. Nevertheless, wheat is probably the main contributor to the daily intake of T-2 and HT-2 in Norway due to the high wheat consumption. In warmer climates, maize frequently contains T-2 and HT-2 toxins (EFSA 2011a).

Some studies have shown no relation between HT-2 and DON content in cereal grain. In oats from UK, mutual exclusion between the different *Fusarium* species producing these mycotoxins was observed (Edwards 2009b). In France, an inverse relationship between DON and T-2 and HT-2 content was registered in barley (Orlando et al. 2010). In Norway, a non-linear negative association has also been registered between DON and T-2/HT-2 content in Norwegian oats (Hofgaard et al. unpublished). Consequently, the highest concentrations of the sum of T-2 and HT-2 toxins are often found in oat grain lots with low DON content, and vice versa.

T-2 toxin is rapidly metabolised to HT-2 toxin in animals, and it is difficult to distinguish the toxic effects of T-2 toxin from those of HT-2 toxin. The tolerable daily intake (TDI) is given as a group TDI, applied to the sum of T-2 and HT-2 toxins. The sum of the two toxins is therefore calculated in each sample.
Nivalenol (NIV) has a low incidence in Norwegian grain, and its concentrations are low compared with the levels of DON (Appendix B).

2.1.2 Zearalenone

Zearalenone (ZON) is produced by several species of the genus *Fusarium* (Table 1) and occurs worldwide in all types of grains. Maize and wheat bran contain the highest concentrations of zearalenone, but grains and grain-based food such as breakfast cereals, bread and bakery make the largest contribution to the estimated dietary intake in Europe due to the high consumption (EFSA 2011b). Even vegetable oils make a significant contribution to the overall dietary intake of zearalenone.

2.1.3 Fumonisins

Fumonisins (FUM) are toxins produced by several fungi of the genus *Fusarium*, but the main producers are *F. verticillioides* and the related *F. proliferatum*. Fumonisin occurrence in maize has been reported in many areas of the world. The occurrence in other grains is very limited. Fumonisins have not been detected in Norwegian grain. Consumption of contaminated maize is the only major source for dietary intake. Fumonisins are divided into the four subgroups, A, B, C and G, of which the B-type occurs most commonly (Marasas 1996).

2.1.4 Enniatins

Enniatins (ENN) are secondary fungal metabolites that are mainly produced by *Fusarium* species (Uhlig et al. 2007; Jestoi 2008). The main producer of enniatins in Norway is probably *F. avenaceum*, but enniatins are also produced by *F. langsethiae*, *F. poae, F. sporotrichoides* and *F. tricinctum* (Table 1). The most prevalent *Fusarium* species in the Nordic countries, *F. avenaceum*, does not produce "traditional" mycotoxins like trichothecenes but emerging mycotoxins like enniatins, beauvericin, and moniliformin (Jestoi 2008). The data on occurrence is sparse, and currently no clear conclusions on differences between cereal species or annual variations in Norway can be drawn.

2.1.5 Beauvericin

Beauvericin (BEA) was first isolated from the fungus *Beauveria bassiana* (Hamill et al. 1969; Gupta et al. 1995) but has later been identified as a secondary metabolite in a number of different *Fusarium* strains (Logrieco et al. 1998; Moretti et al. 2007). The main producers in Norwegain grain are *F. avenaceum*, *F.equiseti* and *F. poae* (Table 1). Beauvericin has been found in grains and maize throughout the world (Ritieni et al. 1997) and has been shown to bioaccumulate and might enter the food chain. The Norwegian occurrence data are sparse, and currently no clear conclusions on differences between cereal species or annual variations in Norway can be drawn.

2.1.6 Moniliformin

Moniliformin (MON) is produced by a large range of *Fusarium* species, but the main moniliformin-producing species in small grains in Norway are probably *F. avenaceum* and *F. tricinctum* (Jestoi 2008; Table 1).

Moniliformin is found most frequently and in the highest concentrations in maize. Lower levels are also commonly detected in maize products such as flour and maize tortillas as well as in small grains such as wheat (Jestoi 2008; Peltonen et al. 2010).

2.1.7 Ochratoxin A

Ochratoxin A (OTA) is produced by storage fungi in the genera *Aspergillus* and *Penicillium*, in both tropical and temperate regions. It is mainly produced during transport and storage under humid conditions. These fungal species are able to grow on a large variety of organic materials, both prior to and after processing. Ochratoxin A is consequently found in a large variety of foods and feeds, including cereal products, dried fruits, in particular raisins, coffee, wine, grape juice, cocoa and spices. In addition, Ochratoxin A can be found in meat products (reviewed in JECFA 2001, 2007; EFSA 2006). This risk assessment only considers the occurrence of Ochratoxin A in cereals, including maize.

2.1.8 Aflatoxin

Aflatoxins are a group of toxic secondary metabolites produced primarily by two species of the ubiquitous fungal genus *Aspergillus*: *A. parasiticus* and *A. flavus*. Aflatoxin B1 (AFB1) is the most important compound with respect to prevalence and toxicity for both man and animals. Aflatoxins are produced pre- and post-harvest under certain storage conditions of temperature, water activity and availability of nutrients, and are mainly found in tree nuts, groundnuts, figs and other dried fruits, spices, crude vegetable oils, cocoa beans and maize from tropical and subtropical regions. In this assessment, aflatoxin is considered only in imported maize for feed.

2.2 Field fungi and their toxins

2.2.1 Mycotoxin-producing field fungi of the genus *Fusarium* in Norway

There are differing reports in the literature regarding the abilities of individual *Fusarium* species to produce mycotoxins. The possibility that mycotoxin producion by a specific *Fusarium* species may be influenced by environmental conditions, plant development stage, and method used for culturing fungi in the laboratory may explain some of the conflicting results. Differences in mycotoxin production between strains within the same *Fusarium* species are also well documented.

The relative importance of different *Fusarium* species has changed over the years. The import of cereal seeds might be an important pathway for the introduction of new strains of previously known species and also of new species. Epidemics caused by virulent, mycotoxigenic *F. graminearum* strains have occurred in both North America and Europe during the last 20 years. Following the recent description of *F. langsethiae*, several countries have detected high levels of its mycotoxins T-2 and HT-2 especially in oats.

The most important *Fusarium* spp. in cereals grown in Norway are presented below. A summary of the species and the mycotoxins they produce is provided in Table 1.

Table 1: Mycotoxins produced by the most important *Fusarium* spp. in cereals grown in Norway.

	T-2/HT-2	DAS	DON*	NIV	ZON	FUSC	ENN	MON	BEA
F. avenaceum						+	+	+	+
F. crookwellense		+		+	+	+			
F. culmorum		+	+	+	+	+			
F. equiseti	+	+		+	+				+
F. graminearum		+	+	+	+	+			
F. langsethiae	+	+					+		
F. poae	+	+		+	+	+	+		+
F. sporotrichioides	+	+			+	+	+		
F. tricinctum						+	+	+	

T-2 = (2α,3α,4β,8α)-4,15-bis(acetyloxy)-3-hydroxy-12,13-epoxytrichothec-9-en-8-yl 3-methylbutanoate; HT-2 = 15-Acetoxy-3α,4β-dihydroxy-8α-(3-methylbutyryloxy)-12,13-epoxytrichothec-9-ene; DAS = diacetoxyscirpenol; DON = deoxynivalenol; NIV = nivalenol; ZON = zearalenone; FUSC = fusarin C; ENN = enniatins; MON = moniliformin; BEA = beauvericin.

*Includes DON-derivatives (e.g. 3-acetyl DON and 15-acetyl DON).

2.2.1.1 *Fusarium avenaceum*

Fusarium avenaceum (Fr.) Sacc. is the most commonly detected species of *Fusarium* in most studies of cereal grain, other cereal plant parts, and settled grain dust in Norway (Appendix A, Tables A1, 2, 3, 4, 5, 6).

Fusarium avenaceum is widely distributed in temperate regions and is commonly soil borne. It can be isolated from cereal seeds and foot rot, and causes head blight. The fungus is also pathogenic on legumes, oilseed rape, potato, and numerous other plants. The teleomorph stage, *Gibberella avenacea* R. J. Cooke, was described in the state of Washington, USA. *F. avenaceum* produces long and slender, slightly curved, 3-5 septate macroconidia and 1-2 septate microconidia, but no chlamydospores.

Fusarium avenaceum lacks the tri5 gene that encodes the enzyme catalysing the first step in trichothecene biosynthesis. The fungus is known to produce enniatins, moniliformin and fusarin C (FUSC). Langseth et al. (1999) found that Norwegian strains of *F. avenaceum* produce moniliformin and enniatins.

2.2.1.2 *Fusarium crookwellense*

The frequency of *Fusarium crookwellense* L. W. Burgess, P. E. Nelson & Toussoun (common synonym: *Fusarium cerealis*) is low in Norway (Appendix A, Table A2).

Fusarium crookwellense is common on cereals, potato, soil, and plant debris in temperate regions, and causes seedling death and head blight of cereals. The teleomorph stage has not been found to date, but molecular data indicate that it belongs to the genus *Gibberella*. The name *F. cerealis* has been proposed, but since no type material exists, *F. crookwellense* is the valid name (Leslie and Summerell 2006). *Fusarium crookwellense* produces 5-septate thick-walled macroconidia, chlamydospores, but no microconidia.

Fusarium crookwellense strains produce fusarin C, zearalenone and nivalenol, but not DON. In a review by Schollenberger et al. (2007), *F. crookwellense* is listed among the fungi producing type A trichothecenes in the diacetoxyscirpenol group.

2.2.1.3 *Fusarium culmorum*

Fusarium culmorum (W. G. Sm.) Sacc. has been one of the *Fusarium* spp. most frequently isolated from cereal grain and other plant parts in Norway (Appendix A, Tables A1, 2, 3, 4, 5, 6). During the last ten years the frequencies of this species have decreased.

In temperate regions, *F. culmorum* causes seedling blight, foot rot and head blight of cereals and is common on cereal plant debris. The unknown teleomorph stage belongs to the genus *Gibberella*. *Fusarium culmorum* produces thick-walled, 3-4 septate macroconidia, chlamydospores, but no microconidia.

The fungus reproduces asexually and is phylogenetically related to *F. graminearum* and *F. crookwellense*. *Fusarium culmorum* contains the tri5 and tri6 genes for trichothecene biosynthesis. In a review, Schollenberger et al. (2007) list *F. culmorum* among those fungi producing diacetoxyscirpenol (DAS). Fusarin C is also produced by *F. culmorum*. Langseth et al. (1999) reported that Norwegian strains of *F. culmorum* produced nivalenol, DON, the acetylated DON-derivative 3-acetyldeoxynivalenol (3-acetyl DON) and zearalenone.

Bakan et al. (2001) divided *F. culmorum* into two chemotypes, the nivalenolchemotype and the DON chemotype, and reported that these chemotypes are distributed independently of wheat host variety and geographical origin. Zearalenone production was significantly higher in the DON-producing strains of *F. culmorum* than in the nivalenol-producing strains.

DON appears to play a role in the pathogenicity of *F. culmorum*. In Denmark, Hestbjerg et al. (2002) found strains of the DON chemotype to be most aggressive on barley seedlings.

2.2.1.4 *Fusarium equiseti*

The frequency of *Fusarium equiseti* (Corda) Sacc. is low to medium in Norway (Appendix A, Tables A1, 3, 4, 5).

In both temperate and tropical climates *F. equiseti* is a cosmopolitan soil inhabitant, and is especially common in dry areas (Leslie and Summerell 2006). The teleomorph stage is *Gibberella intricans* Wollenw. *Fusarium equiseti* has long and slender macroconidia, chlamydospores, but no microconidia. *F. equiseti* produces zearalenone and the trichothecenes nivalenol and T-2 toxin. Schollenberger et al. (2007) include *F. equiseti* among fungi that produce diacetoxyscirpenol.

Three Norwegian strains of *F. equiseti* produced diacetoxyscirpenol, nivalenol, and zaralenone (Langseth et al. 1999). In rice cultures of 24 Norwegian *F. equiseti* isolates, Morrison et al. (2001) found that all isolates produced nivalenol and diacetoxyscirpenol.

2.2.1.5 *Fusarium graminearum*

Fusarium graminearum Schwabe has been known as an important and rather common cereal pathogen in Norway for 70 years (Appendix A, Tables A1, 2, 3, 4, 5). Its frequency has increased during the last ten years (Hofgaard et al. 2010a).

Fusarium graminearum has a world-wide distribution on small grain cereals, maize, and many other plants. The fungus is homothallic and commonly produces the teleomorph stage *Gibberella zeae* (Schwein.) Petch on plant tissue. Released ascospores effectively disseminate the pathogen via air currents during the growing season. The anamorphic stage produces 3-4 septate macroconidia, chlamydospores, but no microconidia.

After harvest, *F. graminearum* survives saprophytically on plant debris, which is a reservoir of inoculum for later crops. In Germany, Oerke et al. (2010) concluded that *F. graminearum* is the only *Fusarium* species in which airborne sexual spores contribute significantly to dissemination and development of epidemics in cereals. At a smaller scale, dispersal of ascospores from a concentrated source will result in aggregation of inoculum.

Fusarium graminearum strains can produce relatively large amounts of the type B trichothecenes, nivalenol and DON, and also high levels of zearalenone. Schollenberger et al. (2007) reported that *F. graminearum* produced type A trichothecenes in the diacetoxyscirpenol group. Norwegian strains of *F. graminearum* produced DON, the acetylated DON-derivative 3-acetyldeoxynivalenol (3-acetyl DON) and zearalenone (Langseth et al. 1999). In recent years also some 15-acetyl DON-producing strains have been detected in Norway. Fusarin C is also produced by *F. graminearum*.

Trichothecene production is regulated by the tri genes. Ward et al. (2002) described three chemotypes of *F. graminearum*: (i) nivalenol chemotype that produces nivalenol and its acetylated derivatives, (ii) 3-acetyl DON chemotype that produces DON and 3-acetyl DON and (iii) 15-acetyl DON chemotype that produces DON and the acetylated DON-derivative 15-acetyldeoxynivalenol (15-acetyl DON). PCR assays have been developed to distinguish between the toxin-producing genotypes using primers based on the sequences of the alleles at Tri3, Tri5, Tri7, and Tri13. In Northern Europe, the 3-acetyl DON genotype is dominant, while in South and Central Europe the 15-acetyl DON genotype is the most prevalent (Yli-Mattila et al. 2009).

DON is a pathogenicity factor for *F. graminearum* causing necrosis in wheat leaves, which allows the fungus to spread into the rachis from florets (Desjardins et al. 1996). There is some evidence that DON also functions as a pathogenicity factor in maize.

A genetic map of *G. zeae* has been published. The species is composed of phylogenetic lineages that can be identified based on genes in the genome. Eleven different lineages have been proposed as new species (Trail 2009).

Waalwijk et al. (2003) employed multiplex PCR and found that *F. graminearum* was the most abundant species in Dutch wheat fields during the years 2000-2001. This contrasts with results fromearlier studies in 1980s and 1990s, when *F. culmorum* was most common in the Netherlands. The authors suggest that increased cultivation of maize in the Netherlands and warmer summers were driving factors behind the change. In Germany, the production of large quantities of ascospores by the homothallic species *F. graminearum* was proposed to favour its rapid dissemination (Obst et al. 2002). Similar increases in the relative prevalence of *F. graminearum* have been reported from other European countries (Xu et al. 2008). Increased occurrence of *F. graminearum* has in recent years also been observed in Norway (Aamot et al. 2008).

2.2.1.6 *Fusarium langsethiae*

The frequency of *Fusarium langsethiae* Torp & Nirenberg has been found to be medium to high in recent studies from Norway (Appendix A, Tables A4, A5 and A6). This species has become one of the most common *Fusarium* spp in oats.

Strains, previously known as “powdery” *F. poae*, were described as *F. langsethiae* by Torp and Nirenberg (2004). Earlier reports on *F. poae* may include both *F. poae* sensu stricto and *F. langsethiae*. The unknown teleomorph stage belongs to the genus *Gibberella*. Sequences of the tri5 gene of *F. langsethiae* are identical to those of *F. sporotrichioides*. Cluster analysis

based on morphology, phenotype data and genotype data indicate that there is a close relationship between *F. langsethiae* and *F. sporotrichioides* (Schmidt et al. 2004). Phylogenetic analyses confirmed that *F. langsethiae* is a distinct new species, and is more closely related to *F. sporotrichioides* than to *F. poae* (Knutsen et al. 2004; Yli-Mattila et al. 2004). Yli-Mattila (2010) concluded that *F. langsethiae* can be divided into two lineages based on IGS sequences. The European lineage has only been found in Europe, and the Asian *F. langsethiae* lineage is distributed in Asia and Eastern Russia.

Imathiu et al. (2009) reported that *F. langsethiae* could be isolated from symptomless oat and wheat grain, but the authors were unable to demonstrate any evidence of pathogenicity in inoculation experiments on wheat leaves. When different inoculation methods for *F. langsethiae* in oats were compared, Divon et al. (2011) concluded that the fungus has a strong preference for panicle infection. Re-isolation and qPCR were used to verify the presence of *F. langsethiae* in infected tissue. Low temperature and high humidity following inoculation with a spore suspension ensured establishment of *F. langsethiae* both at flowering and at the early dough stage. Apical tips of palea and lemma developed a dark brown colour towards maturation. Vacuum inoculation without wounding at the boot stage resulted in even grain infection. Inoculated panicles failed to emerge from the leaf sheath, and brown discoloration of the straw was common.

Thrane et al. (2004) reported that *F. langsethiae* produced enniatins and the type A trichothecenes T-2, HT-2 and diacetoxyscirpenol. In Finland, the level of T-2 production in *F. langsethiae* was higher than in *F. sporotrichioides* (Yli-Mattila et al. 2009).

2.2.1.7 *Fusarium poae*

The frequency of *Fusarium poae* (Peck) Wollenw. has been medium to high in most studies of cereal grain in Norway (Appendix A, Tables A1, 2, 3, 4, 5). However, isolates of *F. langsethiae* were probably included in these frequency data until *F. langsethiae* was identified and described as a separate species (Torp and Nirenberg 2004).

Fusarium poae is most common in temperate regions and cereal seeds and heads are commonly contaminated by this fungus. Mating types have been identified in *F. poae*, but no sexual stage has been found. The unknown teleomorph belongs to the genus *Gibberella*. Not all strains of *F. poae* produce macroconidia. The microconidia are globose to napiform, with a distinct papilla, and usually without septa, but no true chlamydospores are produced. Liu and Sundheim (1996) identified 13 vegetative compatibility groups (VCG) in 22 Norwegian and 2 Polish isolates of *F. poae*. The relatively large number of VCG groups indicates that sexual recombination occurs infrequently in this species (Leslie and Summerell 2006).

Fusarium poae produces the type A trichothecenes HT-2, T-2, and diacetoxyscirpenol and the type B trichothecene nivalenol (Thrane et al. 2004; van der Fels-Klerx and Stratkou 2010). Other mycotoxins, like zearalenone, enniatins and fusarin C, are also produced by the fungus. Liu et al. (1998) found that 20 of the 24 *F. poae* isolates from Norway and Poland produced diacetoxyscirpenol, and half of the isolates produced nivalenol. None of the isolates produced T-2 or HT-2 toxins. Langseth et al. (1999) reported that of five Norwegian *F. poae* strains, one produced nivalenol and diacetoxyscirpenol and four strains produced zearalenone.

2.2.1.8 *Fusarium sporotrichioides*

The frequency of *Fusarium sporotrichioides* Sherb. is low in Norway (Appendix A, Tables A3, 4, 5, 6).

Fusarium sporotrichioides is widely distributed in temperate regions. It grows at low temperatures and can be isolated from grain that has overwintered in the field. The unknown teleomorph stage belongs to the genus *Gibberella*. Morphologically, *F. sporotrichioides* is similar to *F. poae*, *F. tricinctum* and *F. chlamydosporum*. *F. sporotrichioides* produces 3-5 septate falcate macroconidia, pyriform microconidia without septa and abundant microconidia. Phylogenetic analyses have confirmed that *F. sporotrichioides* and *F. langsethiae* are sister taxons (Knutsen et al. 2004).

Strains of *F. sporotrichioides* produce a variety of mycotoxins. The sequences of its tri5 gene are the same as in *F. langsethiae*. *F. sporotrichioides* produces HT-2, T-2, diacetoxyscirpenol, fusarin C and enniatins (Thrane et al. 2004). Three Norwegian strains of *F. sporotrichioides* all produced T2/HT-2, diacetoxyscirpenol and zearalenone (Langseth et al. 1999).

2.2.1.9 *Fusarium tricinctum*

The frequency of *Fusarium tricinctum* (Corda) Sacc. is medium to high in Norway (Appendix A, Tables A1, 2, 3, 4, 5).

Fusarium tricinctum is most common in temperate regions, and it usually occurs as a saprophyte or weak parasite. It produces 3-4 septate macroconidia, napiform or pyriform microconidia and some strains develop chlamydospores. The teleomorph stage, *Gibberella tricincta* El-Gholl, McRitchie, Schoult. & Ridings can be produced in culture.

Fusarium tricinctum produces enniatins, fusarin C, and moniliformin. *F. tricinctum* lacks the tri5 gene and is not known to produce trichothecenes (Leslie and Summerell 2006). Langseth et al. (1999) found that two of three Norwegian strains of *F. tricinctum* produced moniliformin, and all three produced traces of enniatins.

2.2.1.10 *Fusarium verticillioides*

Fusarium verticillioides (Sacc.) Nirenberg (common synonym: *Fusarium moniliforme*) has not been detected on cereals grown in Norway. However, it is sometimes identified in animal feed based on imported maize.

Fusarium verticillioides is an important pathogen on maize worldwide, and causes stalk rot and cob rot, both of which reduce yield and grain quality. The teleomorph stage of *F. verticillioides* is *Gibberella fujikuroi* (Sawada) Wollenw. var. *fujikuroi* (synonym: *Gibberella moniliformis*),, but the name *F. verticillioides* should only be used for strains of the *G. fujikuroi*, mating population A (Leslie and Summerell 2006). The macroconidia of *F. verticillioides* are long and slender, microconidia are oval to club shaped with no septa, and chlamydospores are not produced.

The most important mycotoxins produced by *F. verticillioides* are the fumonisins (FUM). Some strains produce high levels of fumonisins. In Zambian field experiments on maize inoculated with *F. verticillioides*, Schjøth et al. (2009) determined levels of FUMB1-2 in the range 0-13050 µg/kg (average 666 µg/kg). FAO reported that the average daily intake of maize is 418 g/person in Zambia, which is among the highest in Africa. Mouldy maize is commonly used for beer production, and since fumonisin is water-soluble, beer is often contaminated with high levels of fumonisins.

2.2.2 Mycotoxin-producing field fungi other than *Fusarium* spp. in Norway

2.2.2.1 *Claviceps purpurea*

Cereals can be infected by *Claviceps purpurea* (Fr.) Tul. Clavicipitiaceae, Hypocreales. Infection of the inflorescence leads to production of sclerotia, called ergot, which replace the grain in cereal heads. They are of different sizes related to the dimension of healthy seeds. At harvest the sclerotia fall to the ground, or they are harvested with the crop. Sclerotia sown with the seed or overwintered in the field germinate and produce ascospores that may infect the inflorescence of the next crop. Conidia are produced in sticky, sugary exudates on the florets, and insects that are attracted by the exudates disseminate the pathogen. Spores from sclerotia produced on grasses can also infect cereals.

All cereals can become infected with ergot, but outcrossing species with open flowers, such as rye, and several grasses are most the susceptible. Infected grasses may constitute a reservoir for infection of cereal crops. The sclerotia contain up to 40 different toxic alkaloids that can cause ergotism in mammals and humans who consume contaminated grain.

Data from the Kimen Seed Testing Laboratory, Norway, show that around 0.8 - 1.0 % of seed lots of barley, oats and wheat produced in the years 2007-2010 contained *Claviceps* sclerotia (Appendix A, Table A8). In this period, between 2100 and 2500 seed lots from growers of certified cereal seed were analysed. Winter wheat had the lowest contamination, with 0.3 % of the seed lots containing sclerotia. Data from spring wheat (1.23 % of the seed lots), oats and barley did not indicate any differences between the species. Rye cultivation is very limited in Norway, and data on ergots in rye have not been provided by the laboratory. Contaminated seed lots usually contained only one sclerotium per kg seed. However, up to six sclerotia per kg seed were recorded, which is the maximum number permitted for certified seed. A general sowing rate of 200 kg seeds per ha could then give rise to from 200 to 1200 sclerotia, evenly distributed. Under favourable conditions for development of the fungus and infection (e.g. cereal varieties with open flowers, cool weather around anthesis resulting in prolonged flowering), seed containing sclerotia in high numbers close to the upper limit, can represent a risk for contamination of the crop. The data from Kimen do not include information about the proportion of seed lots that failed to meet the certification requirement owing to their high content of *Claviceps* sclerotia, and no information has been made available about the general content of ergot in cereal grain produced for food and feed. Only low frequencies of ergot are generally thought to occur in harvested cereals in Norway because of the effective removal of ergots from the grain during the harvest and cleaning of the crop. However, ergot has been a sporadic issue in the cereal grain industry. Domestic and wild animals may suffer from ergotism from feeding on infected grasses.

2.2.2.2 *Alternaria*

The cosmopolitan genus *Alternaria* Nees contains about 300 species commonly isolated from plants, soil, food, and the indoor air environment. The production of black, melanin-like pigmented spores is one of its major characteristics. The teleomorphic stages of *Alternaria* spp. belong to the genera *Clathrospora* Pleosporaceae, Pleosporales and *Leptosphaeria,* Leptosphaeriaceae, Pleosporales. The genus *Alternaria* contains important plant pathogens such as *A. brassicae* and *A. brassiciola* on crucifers, *A. porri* on onions, *A. dauci* on carrots and *A. solani* on potato. On cereals, saprophytic *Alternaria* spp. commonly colonise heads of plants that are weakened by the take-all fungus (*Gäumannomyces graminis*) or other pathogens of root and straw. Some *Alternaria* spp. are known to produce mycotoxins. The

common contaminant of cereal grain and grain products, *A. alternata* (Fr.) Keissl., produces the mycotoxin tenuazonic acid and other toxic metabolites.

Kosiak et al. (2004) determined *A. infectoria* E.G. Simmons to be the most common *Alternaria* species in Norwegian grain of reduced quality. The authors found a negative interaction between *F. graminearum* and *Alternaria* spp. on poor quality grain.

Mycotoxin production by *Alternaria* spp. in Norway has not been investigated.

2.2.3 *Microdochium nivale* and *Microdochium majus (*formerly *Fusarium nivale)*

Microdochium majus (Wollenw.) Glynn & S.G. Edwards and *Microdochium nivale* (Wollenw.) Glynn & S.G. Edwards (Glynn et al. 2005) (syn. *Microdochium nivale* var. *nivale* and *Microdochium nivale* var. *majus*, formerly *Fusarium nivale*) are commonly found on cereals and grasses in Norway. Although these fungi do not produce mycotoxins, they are mentioned here as they are part of the fungal complex causing *Fusarium* head blight disease in cereals. Earlier reports on mycotoxin production by these fungi are thought to be the result of misidentification (Marasas et al. 1985; Nakajima and Naito 1995). *Microdochium* spp. survive on crop residues and seeds, and cause seedling blight, straw and leaf lesions, and head blight. Snow mould caused by this fungus is a serious disease on winter cereals and perennial grasses. The teleomorph stages are in the genus *Monographella*.

2.3 Storage fungi and their toxins

A diversity of fungi colonises the surfaces of grain in the field during plant development and grain maturation. Freshly harvested grain contains fungi of the genera *Alternaria*, *Cladosporium*, *Epicoccum*, *Fusarium*, *Pyrenophora*, *Septoria* and *Stemphylium*. *Alternaria* occurs especially at high frequencies. However, these field fungi usually do not continue to grow in the grain after harvest, provided that the grain is dried to and stored at a humidity of less than 23-25 %. The associated mycobiota of different grains is given in Table 2. The mycobiota is defined as the total fungal inventory of the area/product under consideration. However, only a restricted number of fungal species, normally less than 10, is actually able to spoil a particular product. We refer to this as “the associated mycobiota” of a product.

Storage fungi mainly comprise several species of *Aspergillus*, with minimum humidity growth requirements of around 13.5-14 %, and species of *Penicillium*, usually found in grain lots stored at low temperatures and with moisture content above 16 %. *Fusarium* toxins formed before harvest are stable and persist during storage. However, *Fusarium* spp. can also continue mycotoxin production during storage if humidity levels exceed 19-20 % (Birzele et al. 2000).

Table 2: Associated mycobiota of cereals and cereal products (Samson et al. 2010).

Product type	Crop/product	Spoilage fungal species
Cereals in the field	Barley, oat, rye, wheat	*Alternaria infectoria species group, Alternaria tennuissima species group, Cladosporium spp., Fusarium avenaceum, F. culmorum, F. graminearum, F. langsethiae, F. poae, Penicillium hordei, P. verrucosum, Ulocladium cucurbitae.*
	Maize	*Alternaria infectoria species group, Alternaria tennuissima species group, Fusarium graminearum, F. proliferatum, F. subglutinans, F. verticilloides, Penicillium citrinum.*
	Millets, sorghum	*Aspergillus flavus, Alternaria infectoria species group, Alternaria tennuissima species group, Curvularia spp., Fusarium proliferatum, F. semitectum, F. verticilloides, Penicillium citrinum.*
	Rice	*Alternaria tennuissima species group, Fusarium fujikuroi.*
Cereals in storage	Barley, oat, rye, wheat	*Aspergillus flavus, Aspergillus parasiticus, Alternaria infectoria species group, Fusarium avenaceum, F. culmorum, F. graminearum, F. tricinctum, Penicillium aurantiogriseum, P. brevicompactum, P. viridicatum, P. cyclopium, P. freii, P. polonicum, P. verrucosum.*
	Maize	*Aspergillus flavus, Aspergillus niger, Aspergillus ochraceus, Fusarium graminearum, F. proliferatum, F. verticilloides, Penicillium chrysogenum, P. citrinum.*
	Millets, sorghum	*Aspergillus flavus, Fusarium proliferatum, F. semitectum, F. verticilloides, Penicillium citrinum, Ulocladium atrum.*
	Rice	*Aspergillus flavus, Aspergillus niger, Penicillium citrinum.*
Cereals in airtight storage	Barley, oat, rye, wheat	*Byssoclamus fulva, B. nivea, Candida spp., Penicillium roqueforti, Paecilomyces variotii, Scopulariopsis candida.*
Cereals preserved with acid	Barley, oat, rye, wheat	*Aspergillus candidus, Aspergillus flavus, Aspergillus terreus, Monascus ruber, Penicillium glandicola, P. roqueforti, Paecilomyces variotii.*
Cereal products	Rye bread	*Eurotium repens, E. rubrum, Penicillium carneum, P. paneum, P. roqueforti.*
	Wheat bread	*Eurotium repens, E. rubrum.*
	Feed	*Penicillium aurantiogriseum, P. solitum, P. viridicatum, Aspergillus candidus, Aspergillus flavus, Fusarium verticilloides, F. poae, F. langsethiae.*

In Norway, about one third of the domestic grain is stored on the farms for a period following harvest. Most years, and especially in wet autums, the grain has to be dried to prevent fungal growth during storage. Cooling will also slow down the growth of storage fungi. Commercial grain is commonly traded at moisture content of 15 %. For long term storage grain should be dried to 14.5% or less (Boys 2011).

Stenwig and Liven (1988a) identified fungi in stored samples obtained from the Norwegian Grain Corporation during the years 1985, 1986 and 1987. The average storage time for the barley, oats and wheat samples was 14, 19 and 18 weeks, respectively. *Penicillium* spp. were found in 99 % of the samples, *Fusarium* spp. in 75 %, and *Aspergillus* spp. in 29 %. Of the 16 species belonging to the genus *Penicillium*, the highest amounts were isolated of *P. aurantiogriseum* Dierckx (syn. *P. puberulum* Bainier), *P. brevicompactum* Dierckx, and *P. viridicatum* Westling (syn. *P. aurantiogriseum* var. *viridicatum* (Westling) Frisvad & Filt). Of the 12 *Fusarium* spp. isolated, *Fusarium avenaceum*, *F. culmorum*, and *F. tricinctum* were the

most common. *Aspergillus* spp. were identified in relatively few samples, with *A. flavus* Link occurring in relatively low numbers.

In a microbiological survey of Norwegian animal feed, 72 lots of standard swine feed and 75 samples of hen feed were sampled (Stenwig and Liven 1988b). From these samples, 29 *Penicillium* spp. were isolated, with *P. aurantiogriseum*, *P. solitum* Westling var. *solitum*, (syn. *P. melanochlorum* (Samson, Stolk & Hadlok) Frisvad), and *P. viridicatum* occurring most commonly. Of six *Fusarium* spp. isolated, *F. verticillioides* and *F. poae* were most frequent.*Aspergillus candidus* Link and *A. flavus* were most often detected of the *Aspergillus* spp. in the survey by Stenwig and Liven (1988b).

It should be noted that studies before 2004 are ignorant to the existence of *F. langsethiae*, formerly called "Powdery poae". *Fusarium langsethiae* was only defined as a separate species in 2004 (Torp and Nirenberg 2004). It is therefore likely that the presence of *F. poae* also might include *F. langsethiae*.

Mycotoxins produced by the different species constituting the associated mycobiota of cereals and cereal products are listed in Table 3.

2.4 In summary

The most important mycotoxin-producing fungi infecting cereals during the growing season in Norway belong to the genus *Fusarium*. The most important storage fungi are species of *Aspergillus* and *Penicillium*. All three genera contain several species with different potential for mycotoxin production.

Of the *Fusarium* species, *Fusarium avenaceum* is the most common, while *F. graminearum* has increased in prevalence during the last 10 years. *Fusarium langsethiae, F. crookwellense, F. culmorum, F. equiseti, F. poae, F. sporotrichoides and F. tricinctum* are other mycotoxin-producing species often identified in Norwegian cereals.

The mycotoxin most commonly produced in the field is DON, and *F. graminearum* and *F. culmorum* are the most important DON producers. Enniatins, produced *by F. avenaceum,* are also common in Norwegian cereals. *Fusarium langsethiae* is the most important producer of the mycotoxins T-2 and HT-2 in the field. Other *Fusarium* spp. produce lower levels of the same mycotoxins. Several *Fusarium* spp. produce low levels of nivalenol, zearalenone, fusarin C and moniliformin in the field. DON is found in practically all samples of flour, bran and oats flakes. T-2 and HT-2 toxins are also widespread, particularly in unprocessed oats, but also in other grains such as wheat and barley. A non-linear negative association has been registered between DON and T-2/HT-2 content in grain, especially in oats. Consequently, the highest T-2 and HT-2 concentrations are often found in oat grain lots with low DON content, and vice versa.

Penicillium and *Aspergillus* spp. produce mycotoxins, including ochratoxin A, during storage. Aflatoxins are a group of toxic, secondary metabolites produced primarily by the two species *A. parasiticus* and *A. flavus*.

While ochratoxin A is a storage mycotoxin of concern in Norwegian-produced grain, aflatoxins may be present in grains imported from tropical climates, mainly in maize. Non-grain food items are important sources of human exposure to aflatoxins and ochratoxin A in the Norwegian population. However, the assessment of the exposure from these sources is not within the terms of reference of the present risk assessment.

Table 3. Toxic secondary metabolites produced by fungi in the associated mycobiota of cereals and cereal products (Samson and Frisvad 2004; Samson et al. 2010)

Species	Important toxic secondary metabolites produced
Aspergillus	
A. flavus	Aflatoxin B_1, B_2, aspergillic acid, cyclopiazonic acid, 3-nitropropionic acid
A. niger	Ochratoxin A, fumonisin B_2, B_4
A. ochraceus	Ochratoxin A, penicillic acid, xanthomegnin, viomellein, vioxanthin
A. parasiticus	Aflatoxins B_1, B_2, G_1, G_2, aspergillic acid, parasiticolide
A. terreus	Citreoviridin, occationally territrem
Byssoclamus	
B. fulva	Patulin (?)
B. nivea	Patulin
Fusarium	
F. avenaceum	Enniatins, fusarin C, moniliformin
F. culmorum	Deoxynivalenol (DON) and other trichothecenes, fusarin C, zearalenone
F. graminearum	Fusarin C, deoxynivalenol (DON) and other trichothecenes, zearalenone
F. langsethiae	Trichothecenes (HT-2, T-2, DAS)
F. poae	Beauvericins, fusarin C, trichothecenes (HT-2, T-2, DAS)
F. proliferatum	Fumonisins, fusaric acid
F. semitectum	Zearalenone
F. subglutinans	Fumonisins, fusaric acid
F. tricinctum	Fusarin C
F. verticilloides	Fumonisins, fusaric acid, moniliformin
Monascus	
M. ruber	Citrinin
Paecilomyces	
P. variotii	Viriditoxin
Penicillium	
P. aurantiogriseum	Penicillic acid, verrucosidin, terrestric acid, nephrotoxic glycopeptides
P. brevicompactum	Botryodiploidin
P. carneum	Patulin, penitrem A
P. chrysogenum	PR toxin, secalonic acids D, F,
P. citrinum	Citrinin
P. freii	Viomellein
P. glandicola	Patulin, penitrem A, roquefortine C
P. hordei	Roquefortine C
P. paneum	Botryodiploidin, patulin
P. polonicum	Penicillic acid, verrucosidin, terrestric acid, nephrotoxic glycopeptides
P. roqueforti	PR-toxin
P. verrucosum	Ochratoxin A, citrinin
P. viridicatum	Penicillic acid, viomellein, xanthoemegnins, viomellein

3 Regional and temporal variation in levels of mycotoxins and mycotoxin-producing fungi in cereals in Norway

3.1 Differences among cereal species with regard to the contamination with mycotoxins and mycotoxin-producing fungi

Fusarium graminearum is now regarded as the most important DON producer in Norwegian oats and spring wheat (Hofgaard et al. 2010a; Hofgaard et al. unpublished; Bernhoft et al. 2010). Generally higher concentrations of DON are observed in Norwegian grown spring wheatthan in winter wheat (Elen et al. 2003). In field trials with Norwegian cereal species during 1992-1995, higher concentrations of DON were recorded in spring wheat, than in barley and oats (Elen et al. 1997). It was concluded that the relative resistance to *Fusarium* head blight in cereal species may vary with the cereal varieties within the cereal species. In another Norwegian field trial, the mean DON content in *F. culmorum*-inoculated spring wheat and barley was higher than in *F. culmorum*-inoculated winter wheat and oats, but only small differences in DON content were recorded among different varieties within the same cereal species (Elen et al. 2003). When grain samples collected from farmers' fields are analysed for mycotoxins a different picture emerges, and higher DON-concentrations are often recorded in oats than in other cereal species (Langseth and Elen 1996, 1997; Langseth 2000; Hofgaard et al. 2010b). This discrepancy between the rankings of cereal species with respect to DON content in field trials compared with naturally infected samples could be partly due to agrotechnical measures as Norwegian oats are often grown in regions with soil types and climatic conditions that are sub-optimal for cereal production. DON in wheat and maize, and partly barley, is the main focus in most temperate cereal growing regions of the world. However, oats are the most severely contaminated cereal crop in Norway. In Norway, there is no maize grain production and hardly any maize production for ensilage.

Fusarium langsethiae has been identified as the most important T-2 and HT-2 producer in Norwegian oats (Hofgaard et al. unpublished; Bernhoft et al. 2010). T-2 and HT-2 toxins are mainly recorded in Norwegian grown oats and barley, but are only rarely detected in spring wheat (Langseth 2000; Bernhoft et al. 2010; Hofgaard et al. unpublished). A mutual exclusion between DON- and T-2- and HT-2-producing *Fusarium* species has been suggested (Edwards 2009b). An inverse correlation between DON and T-2/HT-2 content has been found in Norwegian grown oats (Hofgaard et al. unpublished). Bernhoft et al. (2010) showed that there was no significant correlation between T-2/HT-2 and DON in Norwegian cereals.

3.2 Regional variations in mycotoxins and mycotoxin-producing fungi

The different fungal species in the *Fusarium* head blight complex require slightly different environmental conditions. Therefore, geographical differences in the prevalence of *Fusarium* spp. and their mycotoxins are commonly reported from many countires (Suga et al. 2008; Qu et al. 2008; Edwards 2009b; Xu et al. 2008). *Fusarium poae* has been associated with relatively dry and warm conditions, whereas *F. graminearum* is generally associated with relatively warm/humid conditions. *F. avenaceum* and *F. culmorum* are both associated with cool/humid conditions. *Microdochium nivale* and *M. majus* are associated with cool/moderate temperatures and frequent rainfall of short duration (Xu et al. 2008). *Fusarium langsethiae*

seems to have rather different environmental requirements to those of *F. graminearum* and *F. culmorum* (Edwards 2009b).

Regional differences in the mycotoxin content of harvested grain have been reported in Norway. Langseth and Elen (1997) determined the DON contamination in 5000 samples of Norwegian barley, oats, and wheat, and reported that oats contained higher DON levels than the other cereals. When the results were grouped regionally, significant differences between districts were observed. While DON contamination of barley was lower in Trøndelag than in other regions, oats from Trøndelag contained rather high DON levels. Oats from the districts around Lake Mjøsa contained low DON levels, but relatively high DON contents were often detected in oats from the southeastern part of Hedmark County. The DON content of wheat was generally low, with little variation between the wheat growing districts.

When Langseth and Rundberget (1999) analysed the mycotoxin content of 449 Norwegian grain samples, a south-north gradient became apparent with respect to the T-2 and HT-2 data. Significantly higher levels of T-2 and HT-2 were found in the counties along the Oslo fjord than in the inland counties, Hedmark and Oppland. Negligible contamination with T-2 and HT-2 in oats from Trøndelag was confirmed by Bernhoft et al. (2010), despite similar levels of *F. langsethiae*, the producer of T-2 and HT-2, being measured in Trøndelag as recorded for regions further south.

Figure 2 demonstrates the median DON concentration in oat grain lots from the 2011 harvest in municipalities within Norway. Samples from the different grain lots were taken at delivery, and DON concentration was further analysed at Labnett and Eurofins by enzyme-linked immunosorbent assay (ELISA) delivered by Romer Labs. The data on median DON concentration in altogether 18 447 oats grain lots were kindly provided by Felleskjøpet Agri, Strand Unikorn, Norgesfôr, and their associated partners. In addition to different local weather conditions, agronomic practice (fungicide treatment etc), and field characteristics, such as topography, soil type, moisture, and distance to water sources are expected to cause the local and regional variations observed in mycotoxin concentrations of grain in Norway (Hofgaard et al. unpublished).

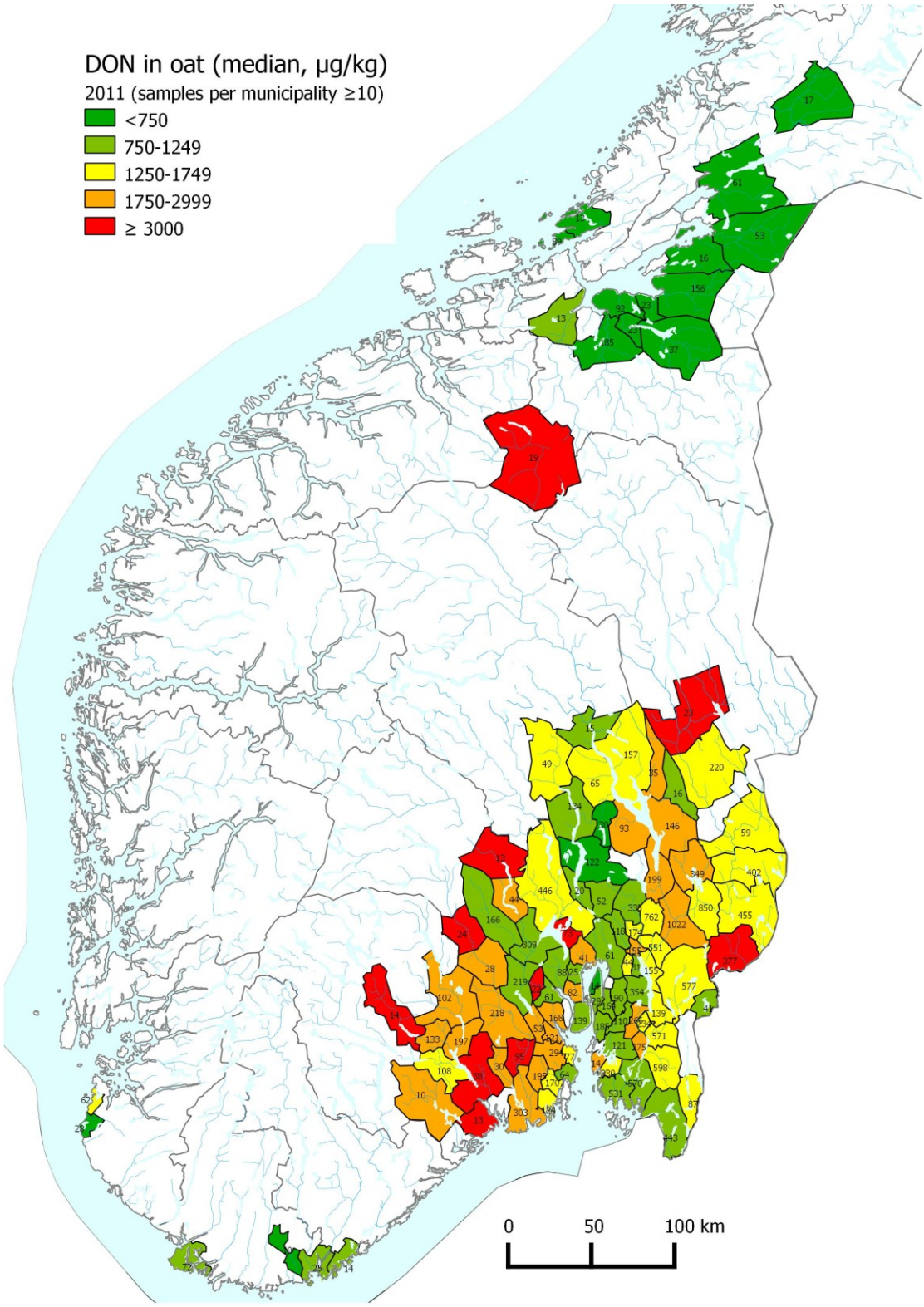


Figure 2: Median concentration (µg/kg) of DON (deoxynivalenol) in oat grain lots from the 2011 harvest, aggregated to municipality level. The data, in altogether 18 447 oat grain lots, were kindly provided by Felleskjøpet Agri, Strand Unikorn, Norgesfôr, and their associated partners. Precautions have to be taken when reading the map as the data on DON concentrations are aggregated according to the farmers addresses (municipalities) and not according to the exact locations of the oat grain fields. The numbers of grain samples analysed within each municipality are indicated. Only data from municipalities with 10 samples or more are included. The map is deveopled by Berit Nordskog, Bioforsk.

As apparent from the map (Figure 2), there can be large variations in contamination values between regions within one year (with median DON concentrations from below 750 µg/kg to more than 3000 µg/kg).). However, there is also a high uncertainty in the calculations of median values because of large variation in the number of grain lots delivered within each municipality. Precautions also have to be taken when reading the map, as the data on DON concentrations of the grain lots are aggregated according to the farmers address (municipality) and not according to the exact location of the oat grain field. Appropriate sampling, as well as precise information about the geographical location of the samples, is key to improving our understanding of the process of disease development and mycotoxin contamination of cereal grain in the field.

In addition to geographical or regional differences in the occurrence of mycotoxin-producing fungi, it is important to notice the within-field spatial variation of mycotoxin-producing fungi. Oerke et al. (2010) investigated the spatial variability of *Fusarium* head blight pathogens and their mycotoxins in wheat fields in Germany. *Fusarium* spp. with high incidence (*F. avenaceum*, *F. crookwellense* (syn. *F. cerealis*), *F. culmorum*, *F. poae*, *F. graminearum*) and also rare species (*F. equiseti*, *F. sporotrichioides*) showed high spatial variability within and among fields. Therefore, sampling of wheat fields for representative assessment of mycotoxins is complicated, especially when the frequency of infection is low (Oerke et al. 2010).

3.3 Temporal variations in mycotoxins and mycotoxin-producing fungi

Fusarium species were recognised as cereal pathogens for many years before their production of mycotoxins was discovered (Roll-Hansen 1940). Research on diseases caused by members of this genus started more than seventy years ago in Norway. During the last twenty years, greater awareness of the potential health problems caused by mycotoxins in humans and domestic animals have led to an almost exponential growth in research on mycotoxins and mycotoxin-producing fungi in Norway.

Roll-Hansen (1940) identified *F. graminearum* as the causal agent of a serious foot rot and head blight epidemic of oats in South-East Norway during 1939. In a report on diseases of field crops Jørstad (1945) referred to *F. avenaceum* as one of the most common *Fusarium* spp. on cereals. He also diagnosed foot rot and head blight caused by *F. graminearum* in herbarium material of oats collected as early as 1911; the teleomorph stage, *G. zeae*, was identified on barley and wheat obtained from Ullensaker, Akershus County in 1941, and on wheat from Øyestad, Aust-Agder County in 1939. Jørstad (1945) concluded that *F. avenaceum*, *F. graminearum*, *F. culmorum* and *F. sporotrichioides* were the most common *Fusarium* species on Norwegian cereals.

Fusarium spp. were isolated from 416 plant samples of barley, oats and wheat during the years 1980-83 (Haave 1985) (Appendix A, Table A1). Symptoms on the plants included discoloration of the lower straw internodes, mycelium growth within the straw, whitehead or shrivelled grain and occasionally pink spore layers in the culm. The most common *Fusarium* spp. were *F. culmorum*, followed by *F. avenaceum* and *F. graminearum*. During the dry summer of 1983, *F. culmorum* was isolated from 41 % of the plant samples, while *F. avenaceum* was obtained from 22 % of the samples. *F. culmorum* and *F. avenaceum* were more common on oats than on barley and wheat. Isolations from kernels during 1982-83 yielded 19.2 % *F. poae*, 5.8 % *F. avenaceum*, 5.3 % *F. culmorum*, and 2.3 % *F. graminearum*.

Abbas et al. (1987) isolated *F. avenaceum*, *F. acuminatum*, *F. culmorum*, *F. sambucinum* and *F. oxysporum* from soil samples collected in Finnmark county, Troms county and the cereal growing counties in Central and southeastern Norway. *Fusarium acuminatum* was most common in the north, while *F. avenaceum* was more uniformly distributed. The toxicity of the isolates was determined in rodent feeding tests. Abbas et al. (1989) determined mycotoxin production in the same group of isolates and found that most of them did not produce trichothecenes. One *F. culmorum* isolate and one *F. sambucinum* isolate produced zearalenone. All but one of the *F. avenaceum* isolates produced fusarin C. The mycotoxin MON was produced by most of the *F. acuminatum* isolates and half of the *F. avenaceum* isolates. Rat feeding tests indicated that other toxic factors were also produced by the Norwegian isolates.

In a comparison of different tillage systems, Henriksen (1999) isolated *Fusarium* spp. from cereal kernels harvested in field trials at five locations over four years (Appendix A, Table A2). At Kvithamar Research Centre in Nord-Trøndelag, *F. avenaceum* was found to be the dominant species in all four years. *F. poae* was the second most common species during 1997, while *F. graminearum* and *F. crookwellense* were the second most common species during 1995. At Apelsvoll Research Centre in Oppland County, *F. tricinctum*, *F. avenaceum* and *F. poae* were identified as the most common species. *F. avenaceum* was found to be the dominant *Fusarium* species at Brandval, Hedmark County. At Norderås Research Farm in Akershus County, the former Agricultural University of Norway, now the Norwegian University of Life Sciences (UMB), *F. poae* and *F. avenaceum* were determined to be the most prevalent *Fusarium* species in the field during two of the four experimental years. At the Hauer farm, Frogn, Akershus County, *F. tricinctum* was identified as the dominant *Fusarium* spp. during two of the four years of field experiments.

Langseth et al. (1999) considered *F. avenaceum*, *F. culmorum*, *F. equiseti*, *F. graminearum*, *F. poae*, *F. sporotrichioides*, *F. torulosum* and *F. tricinctum* to be the most common *Fusarium* species in Norwegian cereals, and mycotoxin production and cytotoxicity were determined in 34 isolates of these 8 species.

In a post-harvest survey of wheat, barley and oats kernels from the major cereal-growing regions of Norway during 1994-96, Kosiak et al. (2003) identified *F. avenaceum*, *F. poae*, *F. tricinctum* and *F. culmorum* as the most common field fungi in all four regions where samples were taken (Appendix A, Tables A3-A5). *F. graminearum*, *F. equiseti*, *F. sporotrichioides* and "powdery *F. poae*", which was later described as *F. langsethiae*, were also common in all regions. Appendix A, Table A5 shows the occurrence of *Fusarium* spp. in oat samples from four regions. Similar data on the prevalence of *Fusarium* in barley and wheat were also presented (Kosiak et al. 2003). The relatively high occurrence of *F. graminearum* in Mid-Norway was unexpected since the temperature during most of the growing season is lower than in the cereal districts of South-East Norway. Samples from South-East Norway had a significantly lower total *Fusarium* prevalence than samples from the other regions. Wheat had lower *Fusarium* levels than barley and oats.

Henriksen and Elen (2005) investigated *Fusarium* infections in wheat, barley and oat grain from field trials. *F. avenaceum* and *F. tricinctum* were the most common of the *Fusarium* spp. isolated. In oats, *F. tricinctum* was the dominant species during both the experimental years. In barley and wheat field experiments, *F. avenaceum* was the dominant species, while *F. culmorum*, *F. graminearum* and *F. tricinctum* were less frequently isolated from the harvested grain.

Halstensen et al. (2006) used a PCR assay to identify *Fusarium* spp. in settled grain dust (Appendix A, Table A6). The dominant species was *F. avenaceum*, which was identified in

94 % of the grain dust samples from wheat, oats and barley harvested in 11 of the most important cereal-producing municipalities in Norway in 1999 and 2000. *Fusarium poae*, *F. langsethiae* and *F. culmorum* were also frequently detected in the same grain dust samples. Less than 10 % of the samples contained *F. graminearum* or *F. sporotrichioides*.

Based on the microbiological and chemical analyses of cereals grown in Norway, Uhlig et al. (2007) identified *F. avenaceum* as the dominant species. They determined concentrations of up to 5.8 ppm enniatins in Norwegian grain harvested during the years 2000, 2001, and 2002, and concluded that conditions in Norway do not favour the production of moniliformin by *F. avenaceum*. The levels of moniliformin in Norwegian grain were determined to be low.

In parallel samples collected at organic and conventional farms during the years 2002-2004, Bernhoft et al. (2010) identified *Fusarium* spp. The mean percentages of kernels infected with the most common species, *F. avenaceum*, were 58 % in organically grown barley and 56 % in conventionally grown barley; in oats, the corresponding figures were 44 % and 43 %, and in wheat the percentages were 50 % and 54 %, respectively. The mean percentages of infected kernels of *F. graminearum* were 8 % in organically grown barley and 10 % in conventionally grown barley; in oats, the corresponding figures were 11 % and 19 %, and in wheat the percentages were 7 % and 10 %, respectively. *Fusarium poae* in oats was at mean proportions of 18 and 13 %, respectively, while in barley and wheat this species was less common. The mean percentages of *F. culmorum*, *F. equiseti*, *F. langsethiae*, *F. sporotrichioides* and *F. tricinctum* were less than 10 % in samples of barley, oats and wheat from the nine counties sampled.

Hofgaard et al. (2010b) used qPCR to identify *Fusarium*-DNA and multi-toxin LC-MS/MS for mycotoxin analyses in about 500 spring wheat and oat samples that were collected from farmers' fields during 2004-2009. The results indicated that *F. avenaceum* and *F. graminearum* and the mycotoxins produced by these fungal species were commonly present in both spring wheat and oats. The DON concentration was above 100 µg/kg in 70 % of the spring wheat samples, and in 66 % of the oat samples. Concentrations of enniatin-B above 100 µg/kg were recorded in 69 % of the spring wheat samples, and in 44% of the oat samples. Both *F. avenaceum* and *F. tricinctum* are enniatins producers. However, analyses for the enniatins producer *F. tricinctum* were not conducted in the study. *Fusarium langsethiae* is regarded as the most important producer of T-2 and HT-2 in oats. Over 0.1 pg *F. langsethiae* DNA per ng plant DNA was found in 72 % of the oat samples, and 38 % of the samples had concentrations of HT-2 + T-2 at more than 100 µg/kg. Very few of the spring wheat samples contained *F. langsethiae* or T-2 and HT-2. *Fusarium poae*, another T-2 and HT-2 producer, was detected in 30 % of the oat samples, but only in 3 % of the spring wheat samples. Moniliformin was detected in approximately half of the grain samples.

The occurrence of mycotoxins in cereal grain and cereal products has been analysed by the Norwegian Veterinary Institute for many years. These data are presented in Chapter 6. The yearly mean concentrations of mycotoxins in crude grain are presented in Table 8 (wheat) and Table 9 (oats) in this chapter. As can bee seen from these two tables, there are no clear trends in occurrence of the T-2 and HT-2 toxins during the last decade. Since *Fusarium langsethiae* was identified as *F. poae* until *F. langsethiae* was described in 2004, it is not possible to determine any long-term trend in its prevalence. Recent studies indicate a shift in the relative species prevalence towards more *F. graminearum* and less *F. culmorum* in Norwegian grain (Hofgaard et al. unpublished; Hofgaard et al. 2010b; Bernhoft et al. 2010). Together with an increased occurrence of *F. graminearum*, there is a trend towards a yearly increase in the median DON concentration in cereal grain, especially in oats (Table 9). The increase in

prevalence of *F. graminearum* reported in Norway is similar to the pattern observed in other European countries.

In the period from 2004-2009, grain samples of oats were sampled from farmers fields in South-East Norway to study the association between agricultural practise and development of *Fusarium* and mycotoxins. All grain samples (about 300) were analysed for mycotoxins. In Figures 3 and 4, the temporal variations in the contamination levels for DON and T-2/HT-2, respectively, in these samples are provided as annual maps with municipality polygons as the spatial resolution. Precautions has to be taken when reading the maps as the sample numbers were quite low, in some cases only one sample per municipality.

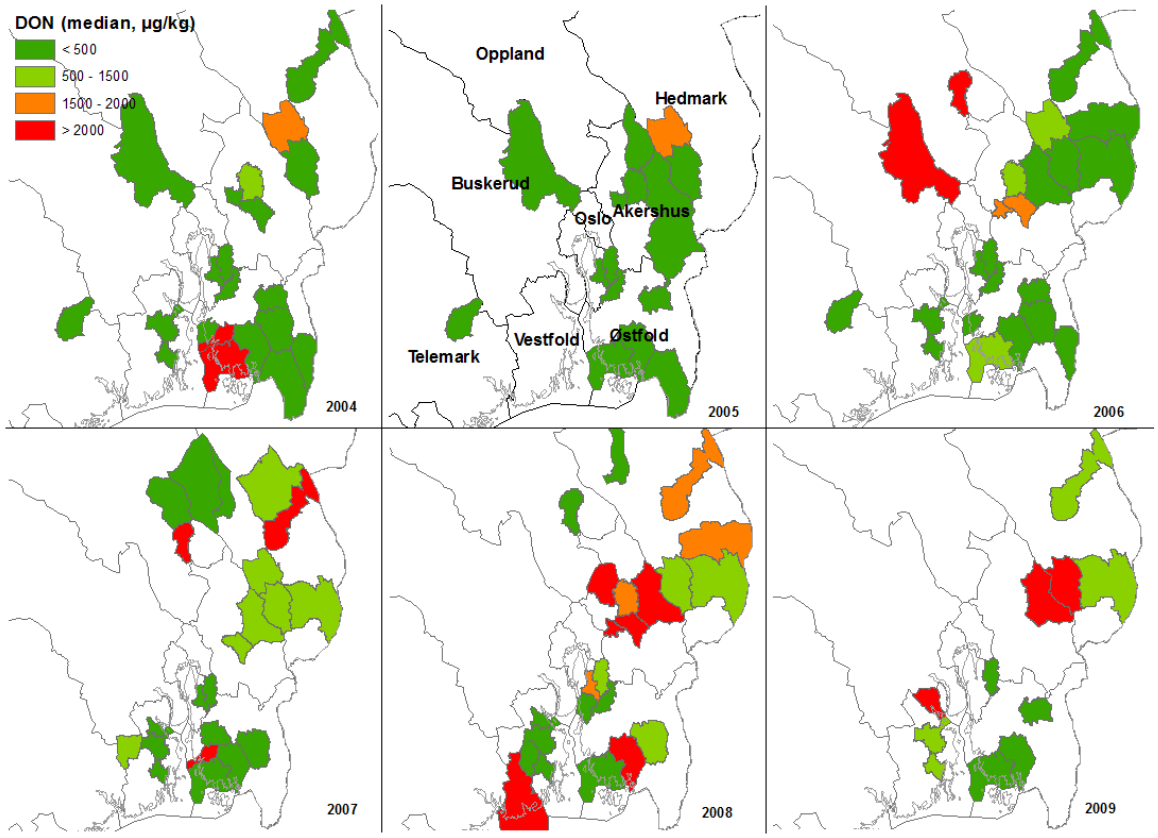


Figure 3: DON (deoxynivalenol) concentration (µg/kg) in oat samples taken during 2004-2009, expressed as median value per municipality (The total number of samples was N = 297 (2004: N=37; 2005: N=23; 2006: N=85; 2007: N = 56; 2008: N = 72; 2009: N = 24). Unpublished data from Bioforsk, from a project financed by the Norwegian Research Counil, the Foundation for Research Levy on Agricultural Products in Norway, and industry partners.

These maps indicate neither a clear spatial pattern nor temporal trends at municipal level. Although this is in line with observations in other countries, where both the incidence and severity of *Fusarium* head blight are known to vary greatly between years and locations (Rossi et al. 2003b), high uncertainty remains about the actual spatio-temporal distribution of mycotoxin contamination in the Norwegian survey, because the sample numbers were low.

The DON data for 2011 shown in Figure 2 apparently provide more detailed information about spatial patterns in DON contamination of oat grain lots. The data indicate higher levels of DON in oats cultivated in south-eastern Norway compared to districts in mid-Norway (Trøndelag). Within south-eastern Norway, the highest median DON values were registered

in municipalities within Hedmark, Vestfold and Telemark counties. Whereas lower median DON values were registered around the eastern side of inner Oslofjord (Østfold, Oslo and parts of Akershus county) and in most municipalities in southern Norway (Agder counties).

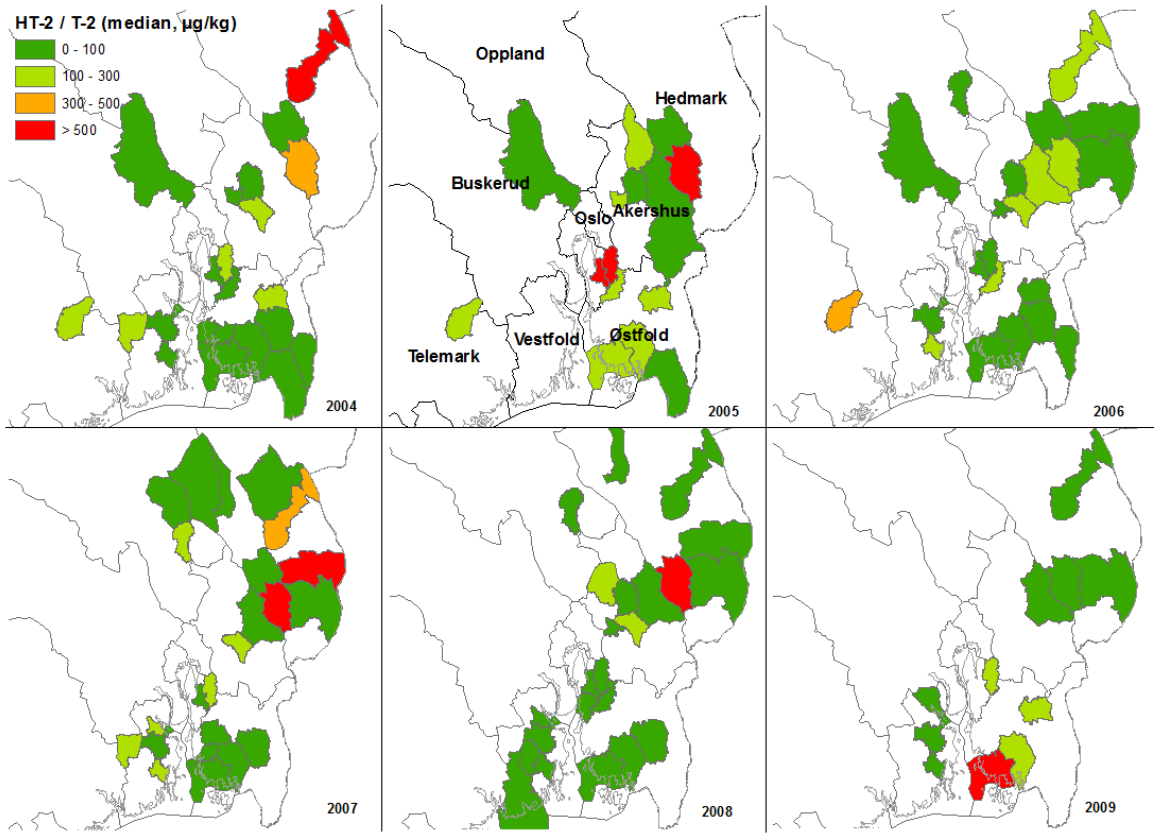


Figure 4: T-2 and HT-2 concentrations (µg/kg) in oat samples taken during 2004-2009 expressed as median value per municipality (The total number of samples was N = 297 (2004: N=37; 2005: N=23; 2006: N=85; 2007: N = 56; 2008: N = 72; 2009: N = 24). Unpublished data from Bioforsk.

3.3.1 *Fusarium* infection levels in cereal seed production are autocorrelated

Cereal seed is grown by contracted seed growers. In order to produce high quality seed these growers' agricultural practices generally follow high standards. However, they also use some specific practices that might actually increase growth opportunities for *Fusarium* spp. One such practice is growing cereal crops on land in which the previous crop was also cereals, i.e. less crop rotation. Furthermore, the same varieties of the same cereal species are often grown in succession in order to maintain pure seed. Although we do not have data on these practices, the fact that these practices are applied is well known, and it is particularly common for some specific cereal species, such as oats. This practise limits the extent to which information from cereal seed production can be extrapolated to ordinary cereal cropping. Nevertheless, effects resulting from cereal seed cropping procedures can provide valuable information about similar processes in ordinary cereal cropping.

Seed infections with *Fusarium* spp. and *Microdochium* spp. have been monitored in Norwegian cereal seed lots since 1970. From 1990, practically all cereal seed lots used for planting in Norway, including all certified and most of the farm-saved seed, have been tested for seedborne pathogens. This is part of the implementation of the governmental policy for

reducing the risk of pesticides use, including "seed treatment only according to need" (Brodal 1991). As the cereal seeds have been produced throughout the main cereal production areas in Norway (southeastern region), it can be assumed that these data reflect the general *Fusarium*/*Microdochium* infection prevalence in cereals intended for food and feed also produced in this area during this period.

The annual average infection percentages recorded during the years 1970-2010 in cereal seeds of barley, oats and spring wheat are plotted in Figure 5, with autocorrelation plots from autocorrelation analyses. Positive autocorrelations indicate that years with high levels of seed infection tend to follow after previous years with high levels; likewise, years with low levels tend to follow years with low levels. Statistically significant autocorrelations are indicated by bars extending beyond the confidence bands (dashed blue lines). The overall trend in the occurrence of *Fusarium*/*Microdochium* in cereal seeds during this period was that significantly higher infection levels were recorded for all three cereal species during the last decade than in the previous 20-30 years.

All statistically significant autocorrelations were positive. Whilst barley displayed significant autocorrelations reaching two years back and spring wheat three years back, the oat data showed dependency that reached as much as five years back.

From this analysis it can be concluded that seed infection levels from previous years contain information that is relevant to the infection level of the current year. Weather can be excluded as an underlying explanation, because the weather would not follow the systematic patterns of autocorrelation. Another possible underlying relationship is that the infected seed can itself serve as a source of inoculum. However, one requirement for seed to be infected in subsequent years is the occurrence of new epidemics during the production of new seed lots. This requirement is not a prerequisite for other sources of inoculum, such as plant debris, because there the inoculum can survive for several years infecting not only the next year's crop, but also for the following two or three years subsequent to the epidemic. This seems to be the most likely explanation for the observed autocorrelations.

The differences among the cereal species can be attributed either to differences in factors that modulate the severity of the epidemics, or differences in practices affecting inoculum depletion. Oats are known to have both a longer period of susceptibility and to be subject to less crop rotation in Norway. The latter is partially due to the fact that oats are generally less susceptible to several plant pathogens, and it is therefore accepted practice to grow oats after oats, i.e. less crop rotation. The relative importance of susceptibility and weather versus inoculum presence/level remains to be determined, but will also depend on locally predominant weather patterns.

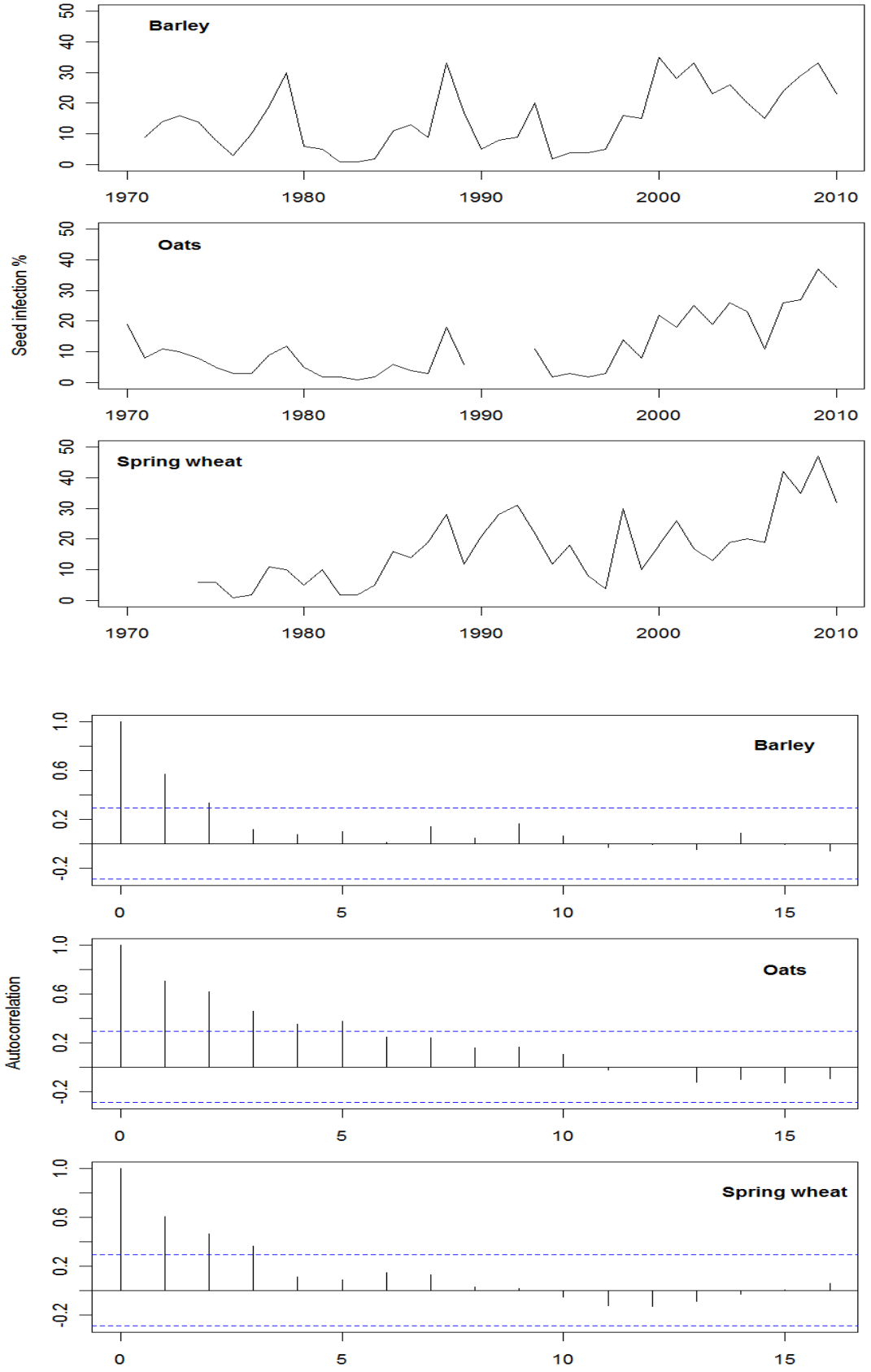


Figure 5: Time series plots of cereal seed infection with *Fusarium* spp. and *Microdochium* spp., and autocorrelation plots indicating time dependencies (years along the abscissa axis) of current year infection levels with previous year's seed infection levels in barley, oats, and spring wheat (Data source: Brodal et al., manuscript in preparation).

3.4 In summary

From 1980-1990s the most frequently isolated *Fusarium* species reported from Norwegian cereals were *F. avenaceum*, *F. poae*, *F. tricinctum* and *F. culmorum*. Recent studies indicate a shift in the relative prevalence of the different *Fusarium* species. *F. avenaceum* is still regarded as one of the most common species in Norway, while *F. culmorum* has decreased in importance. *Fusarium graminearum* has been detected at much higher levels during the last ten years than previously, and recent data shows a trend towards a yearly increased median concentration of DON in cereal grain. *Fusarium langsethiae* appears to be the most important producer of T-2 and HT-2 in oats. This fungal species, often described as a 'powdery poae', was identified as *F. poae* until *F. langsethiae* was described in 2004. Therefore, it is not possible to determine the any long-term trend in its prevalence. There are no clear trends in occurrence of the T-2 and HT-2 toxins during the last decade.

During the last ten years, *Fusarium* infections of cereal seed have increased by more than 100 % in oats, barley and spring wheat, compared with the three previous decades.

T-2 and HT-2 toxins are mainly recorded in Norwegian-grown oats and barley, but are rarely registered above detection limits in spring wheat. DON has been detected in all cereal species in Norway, but ranking cereal species according to DON content reveals a discrepancy in the results from field trials compared with naturally infected samples. Generally, higher concentrations of DON are observed in Norwegian-grown spring wheat than in winter wheat. In studies of naturally infected samples, higher DON concentrations are often recorded in oats than in other cereal species. In field trials, however, oats have been among the species with the lowest DON concentration.

The different fungal species in the *Fusarium* head blight complex require slightly different environmental conditions. Regional differences in the mycotoxin contents of harvested grain have been reported in Norway, but have not been consistent over time. The recent use of fungicides in districts with historically severe *Fusarium* head blight epidemics may mask the differences between districts. In addition, variations in local weather conditions, and in field characteristics, such as topography, soil type, humidity, and distance to water sources, are suspected to influence the local and regional variations in *Fusarium* head blight epidemics. In addition to geographical or regional differences, it is important to be aware of within-field spatial variation of mycotoxin-producing fungi.

4 Factors affecting infection and the production of mycotoxins

Different factors may affect the level of mycotoxin-producing fungi and their mycotoxins in cereals, both in the field and during storage. These factors are climatic and weather conditions (Section 4.1), disease resistance (Section 4.2), cultural practices like tillage, crop rotation and use of chemicals (Section 4.3), and storage conditions (Section 4.4).

4.1 Climate

Many factors are involved in mycotoxin contamination of cereals. However, both food and feed researchers as well as plant pathologists, consider, on the assumption that inoculum is present, climate to be the single most important one (Paterson and Lima 2010; Kriss et al. 2010; Rossi et al. 2003b). For plant diseases, such as *Fusarium* head blight causing mycotoxin contamination of cereals, both incidence and severity vary greatly between years and locations (Rossi et al. 2003b). While it is clear that the weather during the growing season plays a key role in explaining variation between years, it has also been postulated that climate conditions interact with geographical (regional) variations as well, e.g. more humid local climate can be caused by geographical characteristics such as proximity to lakes and/or rivers, in combination with terrain shape and/or vegetation/forest that can affect ventilation of crop fields.

4.1.1 Climate responses of mycotoxin-producing fungi

The main effect of climate is related to the requirements (climate responses) of the fungal organism at its various stages of development (Figure 1), with particular importance during periods when the host plants are susceptible to infection. If an inoculum is available, weather conditions occurring during the growing season determine whether plant disease will develop and whether subsequent production of mycotoxins will occur. When weather conditions are conducive to fungal development, spore production and dissemination, and infection, only one additional requirement for disease development remains: the availability of susceptible plant tissue. The severity of disease depends on the degree to which the above requirements are fulfilled. Temperature and humidity/wetness are the main climatic factors influencing the development of *Fusarium* diseases of cereals. However, the effects of these factors also depend on other environmental and host factors.

The role of climate factors in determining disease development and mycotoxin contamination can be studied under various spatial and temporal aspects. A simple plot of the annual average seed infection for *Fusarium* spp. and *Microdochium* spp. from Norway (see chapter 4.3.3 for details), covering a 41-year period (1970 – 2010) along with representative July rainfall measurements (Figure 6), indicates that years with dry July months are associated with low seed infection levels, whereas years with wet July months are associated with high levels of seed infection. The anthesis/flowering/past-flowering stages of Norwegian spring cereal crops, when cereal plants are most susceptible to infection, usually occur in July. However, this relatively clear pattern is a result of highly culminated data and can be difficult to observe if a predictive model linked to a weather monitoring system operating in real-time is used, because the critical time periods for infection is shorter than a month when considered at the farm level.

It is also interesting to observe (Figure 6) that there is a tendency for a simultaneous increase over the last decade in both infection levels and July precipitation.

Climate change is proposed as one explanation for the increased abundance of *F. graminearum* in Europe. However, relatively high occurrence of *F. graminearum* in post-harvest of cereal grain from Mid-Norway reported from a survey conducted during 1994-96 by Kosiak et al. (2003) suggests that *F. graminearum* can occur further to the north and in colder climates than previously assumed. In a survey studying the the influences of agronomic and climatic factors on *Fusarium* infection and mycotoxin contamination of cereals in Norway, Bernhoft et al. (2012) found *F. graminearum* occurrence to be negatively correlated with July precipitation but positively correlated with high air humidity before harvest, while the prevalence of *F. langsethiae* was positively correlated with July temperature.

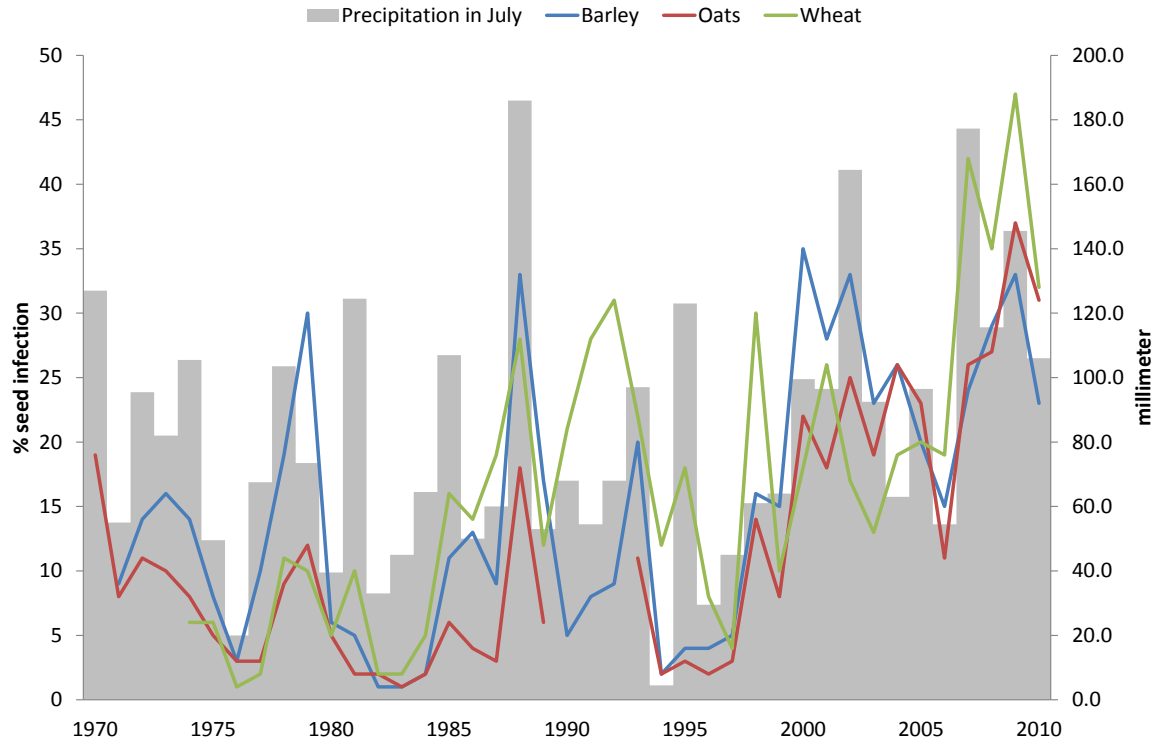


Figure 6. Average *Fusarium* spp and *Microdochium* spp. infection percentages in Norwegian seed lots of oats (red line), barley (blue line) and spring wheat (green line), and July rainfall as bars (1970 – 2010). No data for barley 1970, no data for oats 1990-1992, no data for spring wheat 1970-1973 (Data source: Brodal et al., manuscript in preparation).

4.1.2 Climate effects on host susceptibility

Although climate factors (e.g. extreme weather) are frequently mentioned in the scientific literature as predisposing plants to mycotoxin contamination in the field (Prandini et al. 2009; Whitlow and Hagler 2009), original research reporting such effects on cereal plant susceptibility to mycotoxin-producing fungi is sparse. Papendick and Cook (1974) found that drought stress (plant water stress indicated by lowered leaf osmotic potentials) resulted in wheat plants becoming more susceptible to *Fusarium* foot rot caused by *F. culmorum*.

While the relationship between weather and infection is considered to be most relevant during the phenological stage of anthesis, which is considered to be the most susceptible stage for

infection of cereals, most models also include pre-anthesis weather conditions, without describing in detail the rationale for including this period. Some reseearchers have proposed that weather-induced physiological stress can result in the plants being more susceptible to infection, while others assume that pre-anthesis weather affects the ability of the fungus to produce infective spores (inoculum). The relative importance of pre-anthesis weather conditions on host susceptibility versus the fungus characteristics remains unclear. Simple correlation studies cannot differentiate between these two possible explanations, and this is one of several areas where uncertainty remains regarding climate in mycotoxin contamination of cereals.

4.1.3 Predictive modelling of plant disease and forecasting mycotoxin risk

Research on climate responses of mycotoxin-producing fungi on cereals has been focuseed on host-pathogen-climate interactions for the main cereal species (e.g. wheat), and on life cycle stages (Figure 1), where weather-dependent measures (e.g. fungicide application) can be applied to reduce development of mycotoxins in the crop. The two main objectives of such research have been: (1) to develop predictive models for the determining the need and timing of fungicide application at the cereal's susceptible stage of infection, and (2) to predict the final level of contamination in order to optimise post-harvest management. In Norway, this research has been concentrated on *Fusarium* head blight disease and DON contamination of oats as well as wheat.

The relationships between weather, disease incidence, and severity of mycotoxin-producing fungi, have prompted extensive modeling efforts in the last decade. A model with a good predictive performance would namely provide opportunities for mycotoxin risk management in cereal cropping. When linked to a real-time weather monitoring system, model predictions could provide valuable decision-support information about the need for, and the timing of, fungicide spraying/application.

Various modelling approaches are used, ranging from purely empirical approaches based on the correlations between weather factors and disease incidence/severity data or DON content data (often guided by the knowledge on susceptibility of the cereal variety), to more explanatory models that attempt to integrate causal relationships by modelling the underlying processes. A typical difference between these approaches is that the empirical models may have limited utility beyond their empirical basics (in time and space), i.e. extrapolation is difficult, while process-based models will be more robust in this aspect. However, to produce and process the necessary data, both for model development and for validation, and to produce precise and reliable outputs, is a lengthy, time-consuming procedure.

A disease-forecasting system is principally based on the combined effects of host susceptibility, inoculum strength, and meteorological conditions on the disease development (Xu 2003). Several mathematical models have been developed to describe the relationship between climate and mycotoxin production. Some models describe the relationship between weather factors and development of *Fusarium* head blight disease, while others target the prediction of DON contamination levels directly.

An overview of models developed developed to predict *Fusarium* head blight disease and/or DON contamination of wheat is given in Table 4. The compilation is partly based on a review by Prandini et al. (2009) that summarises the predictive models for *Fusarium* head blight and mycotoxin contamination in wheat, and that has been partially further elaborated by Madgwick et al. (2011). Further details about the models can be found in Appendix A, Tables A7a and A7b.

Table 4. Models for predicting *Fusarium* head blight disease development or toxin contamination in cereals caused by mycotoxin-producing fungi. This overview is partly based on Prandini et al. (2009) and Madgwick et al. (2011).

Predictive models	Disease/ Mycotoxin	Crop	Weather inputs	Performance	Limits	References	Year
Argentina	FHB	Wheat	RH*, daily rainfall, daily temperature	NA	Site- and year-specific	Moschini and Fortugno Fernandes et al.	1996 2004
Belgium	FHB	Winter wheat	Rainfall	NA	Instrumental (radar) availability	Detrixhe et al.	2003
Canada	DON	Cereal grain		$R^2 = 0.55$ $R^2 = 0.79$ $R^2 = 0.56$	Do not consider: crop rotation, crop variety, tillage, fertilisation, etc.	Hooker et al. Hooker and Schaafsma Schaafsma and Hooker	2002 2003, 2004 2007
Italy	FHB, DON, ZON	Wheat		"Good"	Low accuracy for high levels of predicted mycotoxin contents	Rossi et al.	2003a, 2003b
USA	FHB	Spring and winter wheat		NA	Low accuracy	De Wolf et al. van Maanen and Xu Xu Madden et al.	2004 2003 2003 2004
Italy	*F. verticillioides*	Maize		NA	Aspects of the dynamic cycle of fungi are needed	Rossi et al.	2003a, 2003b, 2006
Europe	*P. verrucosum*	Stored cereal grain		NA	Lack of field and storage management effects	Pardo et al.	2006
Norway	DON risk	Wheat	RH*	NA		Elen	Unpublished
Norway	DON in mature crop	Oats	RH*, daily rainfall, daily temperature	$R^2 = 0.44$; R^2(pred) = 0.28 $R^2 = 0.65$; R^2(pred) = 0.60	Site and variety specific. Crop observation	Elen	Unpublished

DON = deoxynivalenol; FHB = *Fusarium* head blight; NA = Not available; RH = Relative humidity; ZON = zearalenone

4.1.4 Comparison between models from Norway and from other countries

While the primary focus in the development of models related to the epidemiology of mycotoxin-producing fungi has been on predictive models, some climate response insights have also been incorporated, at least for guiding the selection of time windows for the aggregation of explanatory weather variables. The models compared in Table 4 are selected on the basis of being relevant for the Norwegian cereal production; e.g. models applied to wheat, and DON-producing fungi. In general, none of the published models included in Table 4 can document a very strong performance, neither for predicting disease development nor for predicting mycotoxin content. The publications presenting the models are generally weak regarding model validation and documentation of operational performance. For some of the models, statistics documenting prediction performance are reported, while for others only statistics on how well the models fit the data are reported, or qualitative statements characterising the model performance. Attempts have been made in Norway to apply some of the internationally developed models, but these have been unsuccessful to date (O. Elen, personal comm.).

4.1.5 Climate change and mycotoxins

In a review, Coakley et al. (1999) concluded that research covering impacts of climate change on plant diseases has been limited, with most work concentrating on the effects of a single atmospheric constituent or meteorological variable on the host, pathogen, or the interaction of the two under controlled conditions. Their results indicate that climate change can alter the stages and rates of the development of the pathogen, modify host resistance, and change the physiology of host-pathogen interactions. The most likely consequences are shifts in the geographical distribution of both hosts and pathogens, and altered crop losses, caused in part by changes in the efficacy of control strategies. A decade later, Paterson and Lima (2010) reported that few studies specifically address the impact of climate change on mycotoxins in food (only a single paper and one abstract). However, more studies have been published since then. The impact of climate change on mycotoxin-producing fungi in cereals (wheat), from a plant pathological point of view, was recently studied for the UK by Madgwick et al. (2011). By combining crop and disease models for different climate change scenarios, Madgwick et al. (2011) hypothesised that with predicted climate change, wheat anthesis will become earlier and *Fusarium* head blight epidemics will be more severe, especially in southern England. Another study by West et al. (2012) elaborates that climate change will have direct impact on *Fusarium* head blight in wheat crops since weather factors greatly affect epidemics, the relative proportions of *Fusarium* head blight pathogens responsible, and the production of DON toxin by *F. graminearum* and *F. culmorum*. Moreover, West et al. (2012) suggest that the severity of the disease is likely to increase further due to indirect effects of climate change such as increased cropping of grain maize, since maize debris is a potent source of inoculum of *F. graminearum*.

Madgwick et al. (2011) concluded further that these projections suggested that improved control of *Fusarium* head blight should be a high priority in industry and government strategies for the adaptation to climate change with the aim to ensure food security. According to Madgwick et al. (2011), some climate-related changes are already influencing the occurrence and composition of mycotoxin-producing fungi (confirming postulates made by Coakley et al. (1999)). *Fusarium culmorum* and *M. nivale* have been the most prevalent species in the cooler temperate climates of Europe, but in the last decade *F. graminearum* has become the dominant species causing *Fusarium* head blight in the Netherlands (Waalwijk et al. 2003), England and Wales (Jennings et al. 2004), and northern Germany (Miedaner et al.

2008), because its higher temperature optimum favours its dominance in the disease complex. Since *M. nivale* is non-toxigenic and *F. culmorum* generally produces less mycotoxin than *F. graminearum*, mycotoxin concentrations may consequently increase. In Canada, a 3-acetyl DON chemotype of *F. graminearum* with increased toxigenic and ecological fitness has replaced the 15-acetyl DON chemotype, indicating genetic differentiation along environmental gradients (Ward et al. 2008). Two recent reviews have considered these and other changes in *Fusarium* head blight pathogen populations, with potential concomitant changes in mycotoxin contamination (Xu and Nicholson 2009; Paterson and Lima 2010). Discussion about climate change and mycotoxins appears in Paterson and Lima (2010) and Magan et al. (2011).

The weather-based models developed to predict the severity of *Fusarium* head blight epidemics, and to advise on the use of fungicides for disease control in Europe, North America, and South America (Prandini et al. 2009) all use relative humidity as an input (Table 4, Appendix A Table A7a). However, because relative humidity is not easily included in projected weather for different climate changes scenarios, Madgwick et al. (2011) chose to develop a new and simple temperature/rainfall-based model for predicting *Fusarium* head blight. These results do not consider direct impacts of changes in CO_2 concentrations or indirect impacts of predicted climate change (e.g. through changes in cropping practices, such as increasing maize production) on the severity of *Fusarium* head blight epidemics.

Changes in temperature and rainfall patterns are likely to affect host-pathogen interactions. Although both groups of organisms are sensitive to climate, their responses differ, and consequently the host-pathogen interactions are likely to be affected.

Whilst it is projected that the UK will experience drier summer weather, which would not favour *Fusarium* head blight, use of the wheat growth model suggests that climate change will result in wheat anthesis occuring earlier in the UK. This means that the critical time period, when rainfall favours *Fusarium* head blight infection, will occur about 2 weeks earlier in the year, when the weather is projected to be warmer but of similar wetness to baseline.

According to Magan et al. 2011, the clearest examples of climate change influencing mycotoxins in preharvest crops are the changes in profile and infection of small grain cereals by *Fusarium* head blight pathogens and DON contamination. In these cases, *F. culmorum* has been gradually outcompeted by *F. graminearum* because of changes in crop rotation and the increasing use of varieties of maize for animal feed that can be grown at more northern latitudes. This has been reviewed as a case study by Chakraborty and Newton (2011).

In Norwegian surveys from a decade ago, *F. culmorum* was the dominant source of DON in cereals (Langseth and Elen 1997; Kosiak et al. 2003). During the last ten years there has been an increase in July precipitation and *F. graminearum* has become the most important DON-producer in Norwegian cereals (Hofgaard et al. 2010a; Hofgaard et al. 2010b; Bernhoft et al. 2012).

Hanssen-Bauer et al. (2009) predict both short-term and longer-term climate changes for Norway. In comparing the two time periods 1961-1990 and 1979-2008, considerable climate differences appear, although such changes are probably due to natural climate variability. However, it is also considered likely that anthropogenic climate change has contributed to this picture, and that the latter will be steadily more strongly expressed in climate variables in the coming years. Towards 2100, the yearly average temperature in Norway is expected to rise by between 2.3 and 4.6 °C. The temperature increase is expected to be greatest in the winter months and least during the summer. Rainfall is projected to increase by 5 – 30 %, with the largest increase during winter, with a 40 % increase over the current level. Thus, this indicates that summer rainfall will be reduced, resulting int the Norwegian climate situation being

similar to that of Great Britain, as assessed by Madgwick et al. (2011). Considered in isolation, the reduced rainfall during the summer suggests less favourable environmental conditions for *Fusarium* infection. However, the increased temperature will result in anthesis occurring earlier. A shift in the timing of the anthesis period might result in the flowering period occurring during the period of increased rainfall, thereby generating a scenario similar to that projected for *Fusarium* head blight in UK.

4.2 Differences in disease resistance among cereal varieties

When available, host resistance is the most practical, efficient and sustainable method for disease control. Complete resistance to *Fusarium* species in cereals, involving hypersensitive resistance responses, has never been documented. However, variations in susceptibility to *Fusarium* head blight among different varieties of wheat have been recognised for more than 100 years (Arthur 1891, in Bai and Shaner 2004).

Although none of the commercial varieties of wheat, oats, or barley demonstrate complete resistance to *Fusarium* infection, some differences in *Fusarium* head blight resistance among varieties have been found (Liu et al. 1997; Hilton et al. 1999; Elen et al. 2003; Kiecana et al. 2002).

4.2.1 Breeding resistant cereal varieties

Breeding for resistance to *Fusarium* is an important goal worldwide. The sources of resistance seem limited, and putative sources often carry undesirable agronomic traits. However, the Chinese wheat variety "Sumai 3" has been widely and successfully exploited, and its resistance is regarded as heritable and stable across environments. Resistance to *Fusarium* is a complex and quantitative trait (dependent on many genes). Several quantitative trait loci (QTLs) have been identified for "resistance", or more correctly, lower disease severity (Bai and Shaner 2004) and considered for introducing into breeding lines. All current sources are for partial resistance. Selection for traits that permit plants to avoid the pathogen may also be a promising approach. Yan et al. (2011) reported that height *per se* contributes towards improved resistance to initial infection, while narrow flower openings are associated with low *Fusarium* head blight in wheat (Gilsinger et al. 2005). Breeding cereal varieties with narrower flower openings should therefore reduce the risk of *Fusarium* infection.

4.2.2 Types of resistance

Two different types of resistance to *Fusarium* head blight in wheat were originally proposed: 1) resistance to initial infection, and 2) resistance to spread of symptoms within the spike (Schroeder and Christensen 1963). These two types can be differentiated between by using different methods for inoculation. It is also common to classify resistance as either morphological or physiological (Rudd et al. 2001). Mesterházy (1995) described five types of physiological resistance (Table 5).

Table 5. Components of resistance to *Fusarium* (Mesterházy 1995).

Type of resistance	Mechanisms
Type I	Resistance to initial infection
Type II	Resistance to spread within the host (spike)
Type III	Kernel size and number retention
Type IV	Yield tolerance
Type V	Decomposition or non-accumulation of mycotoxins

Five different components of resistance, not identical to the five types described by Mesterházy (1995) in Table 5, have been described and tested in the EuroWheat programme (http://www.eurowheat.org/EuroWheat.asp):

1. Resistance to initial infection. Assessed using spray inoculation of heads with *Fusarium* spores, or by spreading *Fusarium* infected debris (or grain) on the soil and evaluating number of infected spikes.

2. Resistance to spread of *Fusarium* fungus within the spike. Assessed by point inoculation of a middle spikelet in the head and evaluating the extent of symptoms spread from inoculation point. Inoculation methods for type I are also widely applied.

3. Resistance to mycotoxins (DON, NIV) i.e., non-accumulation or ability to degrade (or inactivate) mycotoxins. Evaluated by analysis of mycotoxin amounts in grain using ELISA tests or chromatographic techniques.

4. Resistance to kernel infection. Assessed by counting the proportion of kernels visibly damaged by *Fusarium* or by analysis of the amounts of ergosterol or *Fusarium* DNA in grain.

5. Tolerance to *Fusarium* (i.e., tolerant cereal varieties have lower yield losses than intolerant varieties with the same *Fusarium* head blight severity level).

The ability of a cereal variety to escape disease (avoidance) is also an important type of "resistance". Variety traits, such as: 1) plant height, 2) short grain filling period, 3) early maturity, 4) anthers extrusion, and 5) duration and width of flower openings, may all affect *Fusarium* infection and severity.

4.2.3 Will host resistance vary for different *Fusarium* species and isolates?

Host resistance to *Fusarium* seems to be non-specific, at least in wheat, regarding both different *Fusarium* species and different isolates (Mesterházy et al. 1999, 2005; Rudd et al. 2001). A five-year study showed that disease resistance in wheat genotypes (measured as *Fusarium* head blight, *Fusarium* damaged kernels and yield loss) was similar for isolates of all *Fusarium* species tested. Resistance to *Fusarium*, therefore, relates not only to one particular isolate, but the resistance is similar for isolates of several *Fusarium* species (Mesterházy et al. 2005).

Different *Fusarium* species and isolates vary in pathogenicity, and disease-causing capacity is also dependent on environmental conditions (Snijders 1987; Mesterházy et al. 2005). Mesterházy et al. (1999) found a clear connection between the isolate aggressiveness and DON production.

4.2.4 *Fusarium* head blight resistance and DON/mycotoxin accumulation

Generally, there seem to be a close relationship between *Fusarium* head blight values in wheat genotypes and DON content. However, in a study of 53 double haploid progeny lines, the ratio between DON and *Fusarium* DNA was found to decrease with increasing susceptibility (Gosman et al. 2005). This indicates that wheat genotypes that are highly susceptible to *Fusarium* head blight may have only a relatively moderate accumulation of DON.

Mesterházy et al. (1999, 2005) have also reported genotypes with lower infection severity and high DON, and highly susceptible genotypes with moderate or low accumulation of DON. The most resistant genotypes, however, with no or very low infection, had no or very low DON accumulation.

Miller et al. (2001) suggest that resistant cereal varieties may have factors that prevent synthesis and/or promote degradation of DON, since the ergosterol:DON ratio is much higher in resistant varieties than susceptible ones. Ergosterol is a component of fungal cell membranes and often used as quantitative indicator of fungal growth in plant tissue, since it is not part of plant cells.

The fact that Mesterházy et al. (1999, 2005) found additional genotypes with significantly higher DON content than would be expected from their *Fusarium* head blight rating is very important. Cereal varieties that can accumulate extremely high DON levels pose a significant problem to food and feed safety. This phenomenon has major implications for practical breeding, as selection should incorporate both for resistance to fungal damage and also minimal mycotoxin accumulation.

Although DON production is proportional to aggressiveness, DON production is not a prerequisite for pathogenicity, and DON does not play a crucial role in the initial infection of wheat florets (Proctor et al. 1997; Horevaj et al. 2011; Walter et al. 2010).

4.2.5 Variation among cereal grain varieties available in Europe

In 2009, a project was started in Sweeden for assessment of *Fusarium* head blight severity in varieties of winter wheat, spring wheat, and triticale (http://www.jordbruksverket.se/). *Fusarium*-infected grain was spread out as inoculum before flowering, in May 2009 and 2010, and moisture was supplied through irrigation. Disease severity (*Fusarium* head blight) among winter wheat varieties ranged from 5 to 50 %, from 5 to 35 % in spring wheat, and from 10 to 25 % in triticale.

In Denmark, the Knowledge Centre for Agriculture divides the wheat varieties into three susceptibility groups: low (1), moderate (2), and very susceptible (3). Winter wheat varieties have been screened for *Fusarium* head blight severity by inoculating four times during flowering, with irrigation twice daily. In 2010, the disease severity varied among the varieties from 5 % *Fusarium* head blight (CV Petrus) to almost 70 % *Fusarium* head blight (CV Oakley). Based on results from 2008 and 2009, a correlation between DON content and disease severity of 0.66 was identified.

Denmark is also a key partner in the European consortium ENDURE. Through ENDURE, eight European countries present a ranking of wheat varieties regarding susceptibility to *Fusarium* (http://www.eurowheat.org/EuroWheat.asp).

The ranking is not always consistent from country to country, probably due to interactions between the varieties and the different environments. Screening methods also vary among the countries: this factor, as well as different fungal strains, cannot be excluded as a reason for inconsistent ranking.

4.2.6 Variation among cereal varieties available in Norway

Testing and selection for resistance to *Fusarium* resistance in cereals has become an important objective in Norwegian cereal-breeding programmes. However, the official Norwegian variety testing has, to date, not included screening for this trait.

As part of projects at UMB and Graminor, varieties and breeding lines of barley, oats, and spring wheat have been artificially inoculated in field and screened for *Fusarium* head blight severity and DON content during the years 2007-2011 (Lillemo et al. 2013). In this period, DON content in barley has varied, om average, from 5.5 mg/kg in the most resistant variety, to 18.4 mg/kg in the most susceptible variety (Figure 7). In spring wheat, DON content has varied from 5.0 mg/kg in the most resistant breeding lines, to 17.4 mg/kg in the most susceptible one (Figure 8). On average, the most resistant oat variety, Odal, contained 7.7 mg/kg DON, whereas the most susceptible oat varieties, Bessin and Steinar, had 15.4 and 16.9 mg/kg DON, respectively (Figure 9). Among current cereal varieties, the barley varieties Gustav, KWS Olof, Iver and Tyra, and the oat varieties Oda and Skarnes, seem to represent the lowest risk for DON contamination. There are also promising breeding lines. In spring wheat, the best current varieties must be regarded as only moderately resistant in comparison to the promising breeding lines.

Norwegian varieties of winter wheat seem to be less prone to *Fusarium* head blight than more southern varieties (Skinnes pers. comm.). One reason for this may be that these varieties have been selected (by natural infection) for resistance to snow mould, a disease that is most commonly caused by *Microdochium nivale*. This conforms with the non-specific nature of resistance to *Fusarium*.

4.2.7 Conclusion on differences between cereal varieties

European resistance-breeding in wheat has yielded cereal varieties with moderate levels of resistance and that are adapted to the Central European climate. Resistance breeding in barley and oats has been initiated in several countries. The Norwegian cereal-breeding programmes have prioritised *Fusarium*-resistance in cereals. Cereal varieties developed in other Nordic countries may also be considered in Norway. Among current cereal varieties, some barley and oat varieties seem to represent lower risk for DON contamination than others. There are also promising breeding lines.

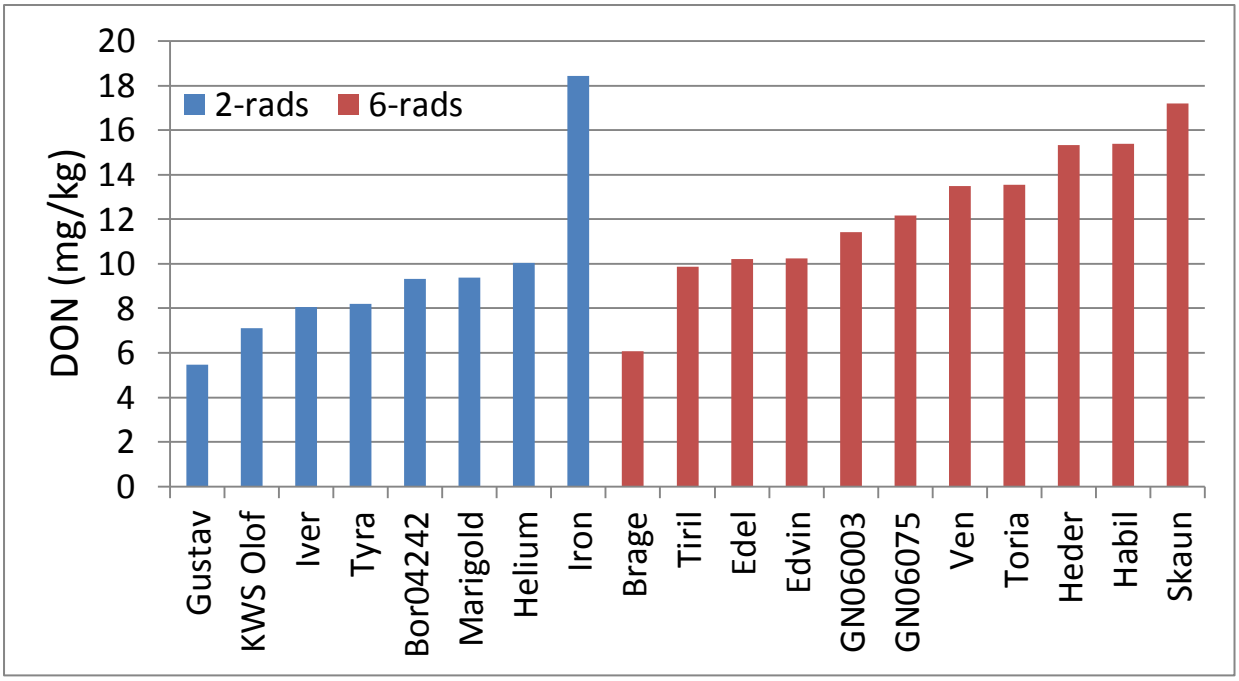


Figure 7. DON (deoxynivalenol) concentrations (mg/kg) in barley varieties (x-axis) inoculated with *Fusarium graminearum* in field trials in the period 2007-2011 (data from Graminor/UMB).

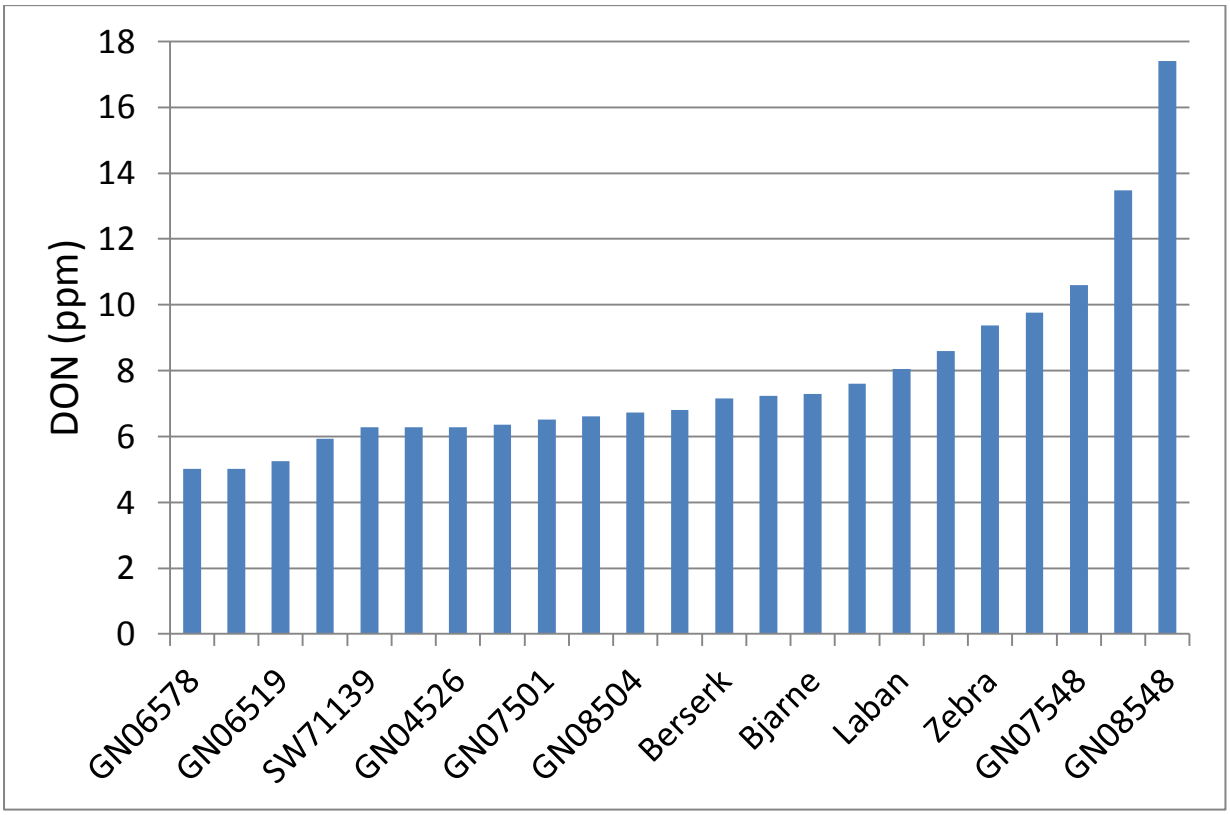


Figure 8. DON (deoxynivalenol) concentration (mg/kg) in spring wheat varieties (x-axis) inoculated with *Fusarium graminearum* in field trials in the period 2007-2011 (data from Graminor/UMB).

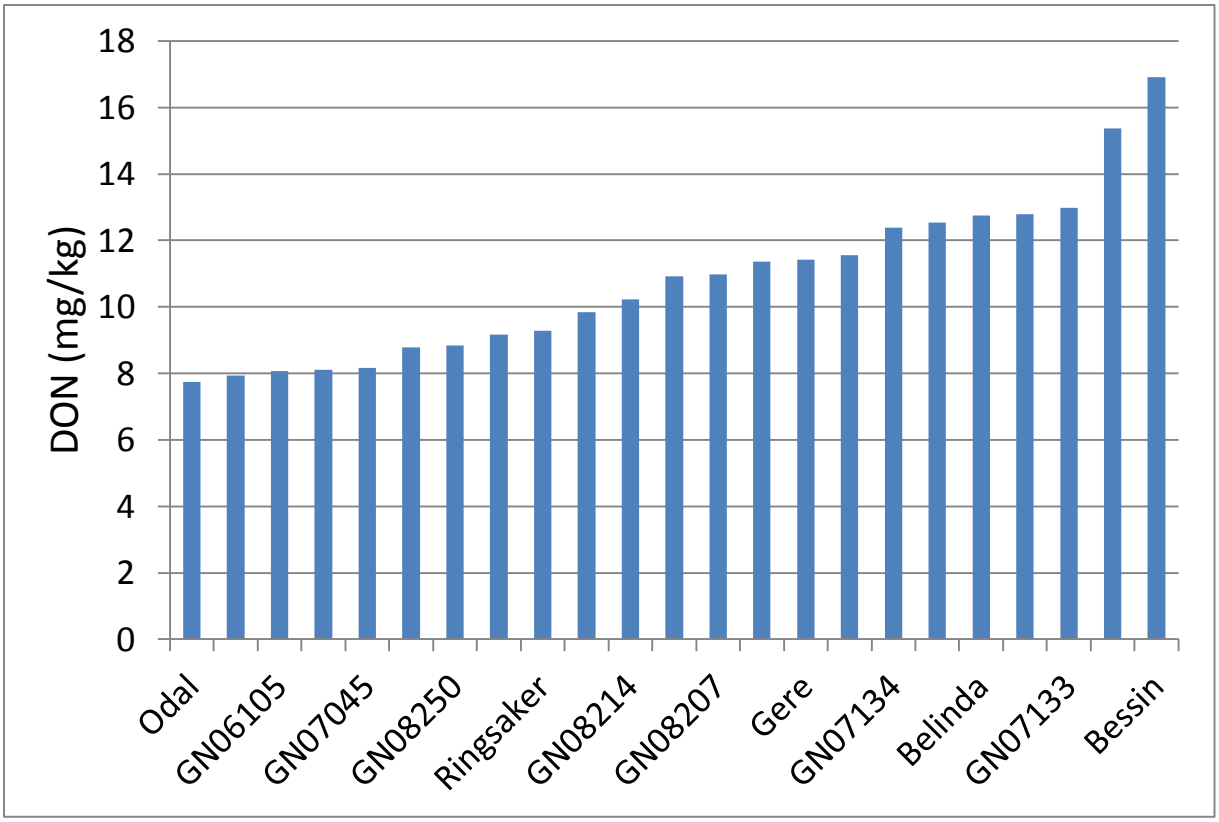


Figure 9. DON (deoxynivalenol) concentration (mg/kg) in oat varieties (x-axis) inoculated with *Fusarium graminearum* in field trials in the period 2007-2011 (data from Graminor/UMB).

4.3 Agronomy

4.3.1 Tillage

Deep tillage and crop rotation are among the cultural practices that have been considered to be of importance for reducing *Fusarium* infection and the production of mycotoxins in cereals. Generally, DON accumulation correlates well with the number of *Fusarium* propagules in the field (Mesterházy 2003), emphasising the importance of appropriate management of previous crop residus (Maiorano et al. 2008).

Reduced tillage or no tillage after wheat or maize can result in increased *Fusarium* head blight and DON contamination of the following wheat crop (Dill-Macky and Jones 2000; Champeil et al. 2004; Teich and Hamilton 1985; Eiblmeier and Lepschy von Gleissenthall 2007). When quantifying the average effect of tillage on DON development in wheat from several separate studies (pre-crop maize), mouldboard ploughing lowered DON concentrations in wheat by 66 % in comparison with minimal tillage or no tillage (Beyer et al. 2006). Under Norwegian conditions, significantly increased average levels of *Fusarium*-contaminated grain were recorded in cereals (oats, wheat and barley) after reduced tillage (harrowing, spring-ploughing or no tillage) compared with autumn ploughing in two out of four field trials conducted between 1994-1997 (Henriksen 1999).

Cereal residues tend to support *F. graminearum* colonisation of plant debris longer in no-tillage/zero tillage systems than in reduced tillage production systems (Pereyra and Dill-Macky 2008), but the opposite situation, in which zero tillage was found to reduce the level of *F. graminearum* inoculum levels compared with those recorded in residues from conventional tillage systems, has also been reported (Fernandez et al. 2008). It seems that the total amount

of plant residues remaining in the top layer of the soil, and not necessarily the tillage practice itself, is the factor influencing disease development. In an Italian study, the total amount of maize residues in the first layer of the soil (10 cm) and on its surface were positively correlated with DON contamination in the following wheat crop, but the effect of tillage was not significant (Maiorano et al. 2008). Within tillage treatments, use of shallow-mixing conservation tillage was associated with only a slight increase in DON concentration in winter wheat compared with conventional mouldboard ploughing, provided that the previous wheat straw soil cover was reduced to 30 % (Koch et al. 2006). The effect of tillage on DON levels was found to be much less important than the effects of year, preceding crop, and cereal variety. The lack of effect of tillage practices on *Fusarium* head blight has also been reported by many others (Bérubé et al. 2012; Schaafsma et al. 2001; Miller et al. 1998; Teich and Nelson 1984).

While most *Fusarium* spp. are spread only over short distances by water splash during rain, *F. graminearum* is known from other countries to produce airborne ascospores (Figure 1). This method of long-distance dissemination will reduce the effect of crop rotation, ploughing, and other cultural measures to control *Fusarium* head blight in cereals.

In a currently on-going project (2010-2014), Bioforsk is comparing the influence of straw treatment and tillage on straw coverage and overwintering of *Fusarium* spp. in crop debris, and the further development of mycotoxins in oats and spring wheat under Norwegian conditions. Preliminary results show that shallow harrowing results in 17-64 % residue coverage, whereas only 1-5 % of the soil is covered with residues in deep tillage plots (Seehusen et al. 2011). Tillage and straw treatment did not seem to affect the relative occurrence of *Fusarium* species in plant debris to the same extent, but due to a reduction in residue coverage, the total amount of *Fusarium*-inoculum was reduced in the tilled plots. Little is known about the epidemiology of the T-2- and HT-2-producer *F. langsethiae* (Imathiu 2012). In UK, significant reductions in T-2 and HT-2 levels were recorded in oats from ploughed fields compared with non-ploughed fields when the previous crop was not a cereal, but no effects of ploughing were found in fields when cereals had been the previous crop (Edwards et al. 2009). In barley grown in France, tillage practices were found not to influence T-2 and HT-2 levels (Orlando et al. 2010). In Norway, field trials conducted in 1997 demonstrated that consistently higher levels of HT-2 toxin were found in continuously grown cereals (oats and barley) after reduced tillage (harrowing, spring-ploughing or no tillage) than in autumn-ploughed fields (Henriksen 1999). In field trials performed in Norway in 2011, a clear reduction in T-2 and HT-2 levels was observed in oats where previous year's straw residues were removed, indicating that the risk of T-2 and HT-2 contamination increases with the amount of cereal residues present in the field (Hofgaard et al. 2012).

Management of previous crop residues may have an important impact on the development of *Fusarium* species within a field. In addition, *Fusarium* spores dispersed from surrounding fields might also have an impact on disease development when weather conditions favour dispersal and development of the disease (Lori et al. 2009). For *Fusarium* species with significant long-distance transport of spores, such as *F. graminearum*/*G. zeae*, management of inoculum in individual fields may have little or no impact unless performed over extensive production areas (Maldonado-Ramirez et al. 2005). Reduced tillage is encouraged in Norway, in order to minimise soil erosion and water pollution. Reduced soil tillage increases the amounts of plant residue on the soil surface, and infected seeds or plant residues may serve as sources for *Fusarium* inoculum in the following season, thus increasing the risk of *Fusarium* infection and mycotoxin contamination of cereal grain.

There are studies showing that soil type influences the *Fusarium* reservoir in soil. The association is not straight foreward, but there are indications of small particle-sized soils as those rich of clay or humus contain less *Fusarium* than soil with larger particles. Huang and Wong (1998) in Australia, Knudsen et al. (1999) in Denmark, Alabouvette (1999) in France, and Kurek and Jaroszuk-Scisel (2003) in Poland have worked on this field. Improved environment for microbial diversity and antagonism in small particle-sized soil is proposed as an explanation. Correspondingly, in a Norwegian survey of *Fusarium* mycotoxins in cereals less DON were shown in cereals grown on clay soil and partly also in samples from silty soil compared to sandy soil (Bernhoft et al. 2012).

4.3.2 Crop rotation

The effect of crop rotation depends on whether the previous crop is a potential host for the pathogens causing *Fusarium* head blight (Champeil et al. 2004).

Residues lying on the surface of the soil play a major role in *Fusarium* infection of cereals (Maiorano et al. 2008). *Fusarium graminearum* survives longer in residues on the soil surface, than in buried crop residues (Pereyra et al. 2004; Khonga and Sutton 1988) and survives for extended periods in wheat residues (Pereyra and Dill-Macky 2008; Inch and Gilbert 2003). Ascospores of *F. graminearum*/*G. zeae*, fully capable of inducing disease, can be produced on residue pieces of wheat after two years in the field (Pereyra et al. 2004). Rotations of at least two years therefore seem necessary in order to avoid infection of new crops by *F. graminearum* originating from *Fusarium*-infected crop debris.

A high incidence of *Fusarium* head blight and elevated DON concentrations in harvested grain are often observed if cereals are planted after corn/maize (Teich and Hamilton 1985; Eiblmeier and Lepschy von Gleissenthall 2007; Beyer et al. 2006; Dill-Macky and Jones 2000). Planting wheat after crops other than maize was found to lower the DON content to, on average, 33 % (Beyer et al. 2006).

A high *Fusarium* head blight index and elevated DON levels are also often recorded in cereals that have been preceded by cereals, compared to cereals that have been preceded by non-cereals such as oilseed, pulses, or soybeans (McMullen et al. 2008; Fernandez et al. 2005; Dill-Macky and Jones 2000). A similar tendency has also been observed for T-2 and HT-2 mycotoxin contamination in barley and oats grown after small-grain cereals, compared with being grown after crops (Orlando et al. 2010; Edwards et al. 2009). In Norway, an increased risk of T-2 and HT-2 contamination has been recorded in continuously grown oats (Hofgaard 2012), and fields with non-cereal crops preceding cereals were found to have lower *Fusarium* infection and less mycotoxin contamination than fields with cereal monoculture (Bernhoft et al. 2012).

Fusarium graminearum has been isolated from residues of several plant species, including wheat, barley, corn, sunflower and grasses (Pereyra and Dill-Macky 2008), whereas a higher colonisation rate with *Fusarium* spp. is often found on cereal residues compared with residues of oilseed, soybean/pea, flax or fallow (Fernandez et al. 2008; Guo et al. 2010; Golkari et al. 2008). Residues of forage legumes seems to have minimal colonisation by *F. graminearum* (Pereyra and Dill-Macky 2008). *Fusarium equiseti* and *F. acuminatum* are considered to be saprophytes or weak pathogens, and these species seem to play a role in the survival of *Fusarium* spp. in crop residues (Fernandez et al. 2008).

In 2005, *F. graminearum* was isolated from potato (dry rot) for the first time (Ali et al. 2005). This finding can have epidemiological implications, as potato is often used in rotation with wheat, oats, and barley (Ali et al. 2005). In a Slovakian study, the highest mean DON content

in wheat was reporded from regions where potatoes were mainly grown (Vanco et al. 2007). In Norway, a high DON contamination of wheat has been recorded in some fields with potatoes as previous crop (Brodal unpublished).

Although plant residues play an important role in the production of *Fusarium* inoculum, a consistent pattern of *Fusarium* colonisation or disease development is not necessarily recorded in relation to the previous crop (cereals compared with non-cereals), suggesting that other factors, such as airborne inoculum, are also important factors in epidemics (Golkari et al. 2008; Davis et al. 2009).

Crop rotation, with less cereal-intense rotations, is generally an effective way of reducing the risk of *Fusarium* and mycotoxin contamination. The planting of consecutive crops of small grain cereals, such as oats in Norway, should only be considered after an assessment of the risks of *Fusarium* infection has been conducted. One of the main challenges to cereal cultivation in Norway is an extensive practice of cereal monoculture in large areas. This is mainly due to limited use of rotation with non-cereal crops on farms in south-eastern and central Norway. A Government policy, initiated 60 years ago, transferred most of the domestic animals out of these two regions. Because of the short growing season in Norway, and shortage of suitable cultivars, the production of alternative crops, such as oil seed crops, field pea and field beans, is limited to the southern regions of the country (Abrahamsen et al. 2005). Growing these crops is economically more risky than growing cereals, because of yield instability related to cultivation techniques and pest and disease management.

4.3.3 Irrigation

Rainfall during the critical period around heading and flowering has a major impact on the development of *Fusarium* head blight and DON in cereals (Hooker et al. 2002). Rainfall probably triggers the final ascospore and perithecia maturity before release. High inoculum levels of *Gibberella zeae* have been positively related to rainfall (de Luna et al. 2002). Rain may contribute to increased inoculum levels on the heads by spores being splashed from nearby inoculum sources (Paul et al. 2003). Similarly, irrigation in the period around heading and flowering is likely to increase the risk of *Fusarium* infection and DON contamination of cereals. In field studies of wheat, *Fusarium* head blight disease severity and DON levels increased with the number of days (0-3) of irrigation after *Fusarium*-inoculation at mid-flowering (Lacey et al. 1999). In contrast, extended periods of irrigation from anthesis until harvest reduced the DON levels in *Fusarium*-inoculated wheat (Culler et al. 2007; Lemmens et al. 2004a). Gautam and Dill-Macky (2012) demonstrated that DON can be leached out from the host tissues of wheat after a single wetting event (irrigation, rainfall etc).

4.3.4 Seed infection

In areas with sufficient *Fusarium* inoculum in the debris from the previous crop, there is no evidence that seed infection contributes significantly to the development of head blight and mycotoxin production. However, infected seed represents a considerable risk for introducing the pathogen into an area where it was previously absent.

In addition to infected plant debris from the previous crop and airborne spores, seeds infected by *Fusarium* are the other major source of inoculum (Parry et al. 1995; Champeil et al. 2004). Seed infection allows the disease to develop during emergence of seedlings, and seedling blight is reported mainly to be a result of seedborne infection (Duthie and Hall 1987; Colhoun et al. 1968; Jones 1999). Severe crown rot can result in DON translocation to the head and

consequently DON contamination of grains at harwest (Covarelli et al. 2012). *Fusarium*, including *Microdochium*, has frequently been detected in routine testing of Norwegian cereal seeds for many years (Brodal et al, manuscript in preparation). Since 2004, more aggressive and fast growing strain(s) of *F. graminearum* have been observed in germination analyses, causing severe germination damage, especially in oats (Håkon Tangerås, pers. comm.). Prior to 2004, such severe germination damage had never been observed. It has been speculated that the strain(s) might have been introduced by imported oat seeds. The occurrence of germination damage coincides with the increased occurrences of DON in Norwegian cereals in recent years. Due to germination problems, it has been difficult to provide enough oats seed in Norway, and this has resulted in a reduction in oat production of approximately 20 % in recent years.

In Alberta, Canada, serious problems with *Fusarium* head blight and mycotoxin contaminations have been avoided to date. In order to prevent the establishment of *F. graminearum* and to prevent its increase and spread should it occur, the pathogen has been declared as a pest under Alberta's *Agricultural Pests Act* from 1999. Alberta's *Agricultural Pests Act* is the legislative authority for enforcement of control measures for named pests in Alberta. A management plan has been established that includes restrictions on cereal grain intended for use as seed (Government of Alberta 2002). All cereal grain intended for use as seed in Alberta must be accompanied by a laboratory certificate showing that the relevant seed lot has been tested and found to have non-detectable levels of *F. graminearum*. Prior to planting, all cereal grain intended for use as seed in Alberta must also be treated with a registered fungicide that includes the genus *Fusarium* on the label list of fungi that are controlled. Cereal grain intended for use as seed that tests positive for *F. graminearum* must be disposed of properly and effectively. These lots may be sold for food or feed, but treated (fungicide or insecticide) lots must be destroyed.

4.3.5 Fungicides

The possibilities for controlling *Fusarium* and reducing mycotoxin contamination of grain by application of fungicides have, so far, been limited. The timing of fungicide application is critical. The best effects for *Fusarium* head blight control are achieved when fungicides are applied at full flowering (Mesterhazy 2003). In addition, methods for fungicide application differs from that used to control foliar diseases (McMullen et al. 2012). Data from recent publications have shown that using a triazole fungicide (except fenbuconazole) lowers DON content in wheat by about 50 % (Beyer et al. 2006). The triazole fungicide tebuconazole reduces both *F. culmorum* and *F. avenaceum* infection and DON content of wheat grain, but provides little control of *M. nivale* (Simpson et al. 2001). In contrast, the strobilurin fungicide azoxystrobin is active against *M. nivale*, and its use can result in reduced *Fusarium* head blight, but not in a decrease in DON contamination (Edwards 2004). The triazole fungicide prothioconazole has shown to be superior in *Fusarium* head blight control in wheat (Mesterházy et al. 2004). Control of *F. graminearum* or *F. culmorum* has been the main target when testing the efficacy of fungicides for *Fusarium* head blight control. Recent studies indicate that *F. langsethiae* or T-2 and HT-2 toxins in oats are not reduced by prothioconazole treatment, whereas a clear reduction of DON was recorded from the same plots (Elen et al. 2008). As *F. langsethiae* and T-2 and HT-2 contamination are commonly recorded in Norwegian oats, focus may be directed towards identifying fungicides for effective control of *F. langsethiae* and T-2 and HT-2 toxins.

The effect of fungicides on DON production in *F. graminearum* is influenced by a complex interaction between environmental factors (Ramirez et al. 2004). Application of fungicides at

early developmental stages of the cereal plants may stimulate development of *Fusarium* spp. and mycotoxins (Magan et al. 2002; Chala et al. 2003; Henriksen and Elen 2005; Eiblmeier and Lepschy von Gleissenthall 2007).

Fusarium head blight and DON reduction in cereals would be best achieved by fungicide spraying when used in combination with cereal varieties with high resistance and when the previous crop was non-cereals (McMullen et al. 2008; Beyer et al. 2006; Blandino et al. 2012). As with other fungicides there is a risk that *Fusarium* spp. may develop resistance to triazoles.

4.3.6 Herbicides

Under zero tillage on the Canadian prairies, previous glyphosate applications were positively correlated with the mean percentage isolation of *F. avenaceum* and negatively correlated with *F. equiseti* from cereal crop debris (Fernandez et al. 2008). Positive associations have also been registered between *Fusarium* head blight levels in spring wheat and application of glyphosate within the previous 18 months (Fernandez et al. 2005). A positive association between previous glyphosate use and *Fusarium* head blight levels caused by *F. avenaceum* and *F. graminearum* was also shown by Fernadez et al. (2009), and they suggested that glyphosate might cause changes in fungal communities. Increased frequency of root-colonizing *Fusarium* and shift in the balance between beneficial and detrimental plant-association microorganisms were shown during several seasons of glyphosate application on transgenic, glyphosate-resistant soybean and maize (Kremer and Means 2009). In contrast with with these results, no effect of glyphosate treatment on *Fusarium* seed infection was recorded in field experiments with Norwegian oats and barley in which glyphosate was applied to the stubble in the autumn (Henriksen and Elen 2005). Although field surveys in Saskatchewan, Canada have detected positive associations between incidence of *Fusarium* head blight and application of glyphosate, others claim that there is currently insufficient evidence to prove or disprove a link between glyphosate and *Fusarium* diseases (Powell and Swanton 2008). A recent report from field experiments in Canada concluded that glyphosate had no significant effect on *Fusarium* head blight and DON content in wheat and barley, regardless of either the trial or the site. *Fusarium graminearum* was enhanced in only one of twelve trials (Bérubé et al. 2012). However, the authors discuss the possibilities that their findings were influenced by low levels of application of glyphosate and that precipitation could have caused the herbicide to leach out of the ground. In a survey comparing Norwegian organic farms and conventional farms, Bernhoft et al. (2012) showed a positive correlation between the use of herbicides and the prevalence of *F. graminearum* and *F. langsethiae*. The herbicides used on the conventional farms included glyphosate, MCPA, and various other compounds.

In Norway, the application of glyphosate in cereal fields has recently increased, mainly due to a greater use of reduced tillage. When soil tillage is performed in spring, about 30 % of the area used for cereals is treated with root-weed herbicides, compared with only 20 % of the area in autumn-ploughed fields (Gundersen et al. 2009). More research is needed to clearify whether application of glyphosate influences the risk of *Fusarium* and mycotoxin development in cereals.

4.3.7 Integrated management of *Fusarium* and mycotoxins

Several authors have ranked the effects of different agronomic factors on the development of *Fusarium*, as in Beyer et al. (2006) (Figure 10). A German study provided the following

ranking order of the effects of environmental and management factors on *Fusarium* head blight and DON in winter wheat: Annual variation in rainfall during anthesis = *Fusarium* infection/susceptibility of the previous crop = *Fusarium* susceptibility of the wheat variety being grown > soil tillage applied to cultivated wheat >= fungicide application at anthesis of wheat (Koch et al. 2006). According to McMullen et al. (2012), control of *Fusarium* head blight and DON are best achieved in studies where cultivars with the best *Fusarium* head blight resistance are combined with optimal use of fungicides (type, time point, and method of application), and the effect is even better when combined with a non-host as a previous crop. When summarising the effects of agronomic factors on the content of T-2 and HT-2 toxins in French grown barley, the following ranking order was suggested (starting from the most important factor): sowing date, rotation, tillage, varietal susceptibility and fungicide use (Orlando et al. 2010).

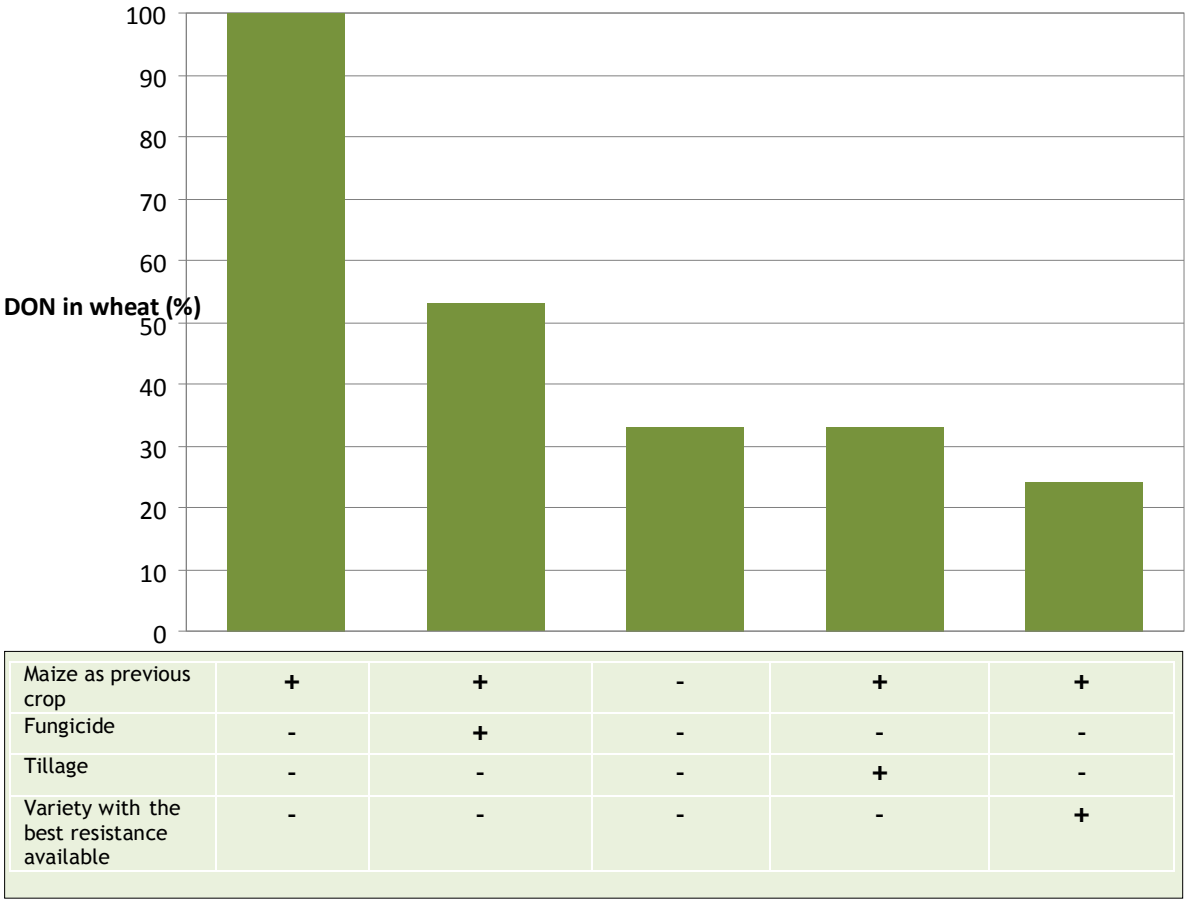


Figure 10: The effects of previous crop, tillage, cereal variety, and triazole fungicides on deoxynivalenol (DON) content of wheat grain, reproduced from Beyer et al. (2006). DON content in wheat with maize as previous crop is given a value of 100% DON. The original data is published in a review of 17 publications from year 1985 to 2005 (Beyer et al. 2006).

Ongoing research is focusing on reducing the inoculum potential of residues by aiding residue decomposition and/or directly targeting *Fusarium* spp. on residues (Dill-Macky 2008).

Beyer and co-workers reviewed literature from 17 publications presenting the effects of previous crop, tillage, cereal variety, and triazole fungicides on DON content of wheat grain

in the period from 1985 to 2005 (Beyer et al. 2006). Based on these data, an average relative effect of each different crop factor on DON content in wheat was presented (Figure 10, data from Beyer et al. 2006). The use of moderately resistant cereal varieties was shown to have the largest impact on DON reduction in these studies.

The data presented by Beyer et al. 2006 should be considered in association with the fact that no resistant cereal varieties have been commercialised, and that there are no available fungicides that give 100 % protection against *Fusarium* mycotoxin contamination of cereals. Thus, an integrated approach, using a combination of crop rotation, deep tillage, cereal varieties with the best resistance available, and appropriate fungicide treatment at flowering, is a strategy to control *Fusarium* and mycotoxins (especially DON) in cereals.

4.3.8 Organic farming

The Council Regulation (EC) No 834/2007 of 28 June 2007 provides the framework for what is defined as organic farming within the European Union and the European Economic Area. The following information is taken from this regulation: "The essential elements of the organic plant production management system are soil fertility management, choice of cereal species and varieties, multiannual crop rotation, recycling organic materials and cultivation techniques. Additional fertilisers, soil conditioners and plant protection products should only be used if they are compatible with the objectives and principles of organic production".

Lower rates of *Fusarium* head blight and mycotoxins in cereals from organic farming systems compared with conventional farming systems have been reported (Birzele et al. 2002; Bernhoft et al. 2010; Benbrook 2006).

Lower content of DON was registered in bread produced from organically grown cereals than bread produced from conventionally grown cereals, with a significant effect in mixed wheat bread (Schollenberger et al. 2005). An organic centre in Germany refers to several reports on reduced DON levels in organically produced cereals or their products as compared with the equivalent conventionally produced items (Benbrook 2006). In a Norwegian study, *Fusarium* infection in oats, wheat and barley, and mycotoxin levels (DON in wheat, T-2 and HT-2 in oats and barley) were significantly lower in organically grown cereals compared with conventionally grown cereals (Bernhoft et al. 2010). In an English study, the predicted T-2 and HT-2 mean for organically grown oats was five times lower than for conventionally grown oats (Edwards 2009b), and the predicted incidence of T-2 and HT-2 in organically grown wheat was about half that of the conventionally produced wheat (Edwards 2009c). Significantly lower contamination levels with HT-2 and other type A trichothecenes were found in oats and oat products from organic farming systems than conventional farming systems (Gottschalk et al. 2007). However, most of the studies presented here have been performed in a period when fungicides against *Fusarium* head blight were not a common practice. In Norway, triazole fungicides for *Fusarium* control were available from 2008 (Personal communication, Erlend Spikkerud, the Norwegian Food Safety Authority). Moreover, in Norway, the *Fusarium* head blight disease pressure and the DON contamination level were, in general, considerably lower at the time period when the Norwegian survey was perfomed (2002-2004) compared with recent years (2009-2012).

In about half the surveys published lower mycotoxin content has been recorded in organically grown cereals than in conventional production, while the rest of the sureveys show no difference in mycotoxin content between the two farming systems. The lower mycotoxin content recorded in organicly grown cereals compared with conventionally grown cereals could be due to more widespread use at organic farms of cultivation practices known to

reduce the risk of mycotoxin contamination in cereals. Less cereal-intense rotations are common at organic farms, whereas monoculture of cereals is frequent in conventional growing systems. Crop rotation reduces the risk of mycotoxin contamination as the use of non-cereals as previous crop can reduce the build-up of *Fusarium* inoculum in the field (Guo et al. 2010). Deep ploughing followed by harrowing is a common cultivation practice prior to sowing of organically grown cereals in Norway (Frøseth 2011). In addition, harrowing once or twice after sowing is used for weed control. Cereal residues tend to support *G. zeae* colonization of plant debris for longer in no-till/zero till systems than in reduced tillage production systems (Pereyra and Dill-Macky 2008). The tillage practices most common in organic farming systems might reduce colonization of crop debris with pathogenic *Fusarium* species, and this will further reduce the inoculum pressure within the field. However, the data from a Norwegian survey by Bernhoft et al. (2012) demonstrated that most of the organic as well as the conventional fields were ploughed. Thus, soil cultivation was not systematically different between organic and conventional practice, and it was not found to be a significant factor explaining the lower *Fusarium* and mycotoxin levels in organic cereals.

Organic farming systems might grow mixed species in the field and a sub-crop of clover is recommended (Frøseth 2011). In organic and low-input systems, a cover crop like red clover or broadleaved weeds might act as a barrier, reducing dispersal of the inoculum from crop debris to wheat ears (Munger et al. 2010).

Cereal varieties with short straw are not recommended for organic farming systems in Norway (Frøseth 2011). Shorter plants might be more exposed to primary inoculum from soil and plant debris, and, in addition, be exposed to a microclimate that is more favourable for *Fusarium* development (Mesterházy 1995; Miedaner 1997; Yan et al. 2011). Cereals with long straw seem to be at lower risk of mycotoxin contamination (Choo et al. 2004). However, the results from a Norwegian survey by Bernhoft et al. (2012) demonstrated that in general the same varieties were used in organic and conventional fields. Furthermore, when comparing the *Fusarium* and mycotoxin concentrations for each variety in organic and conventional fields, the results showed either no difference or reduced concentrations of *Fusarium*/mycotoxins in organic fields.

Avoiding the use of fungicides and herbicides, as occurs in organic cultivation systems, might assist in maintaining a more “natural” balance between fungal species in the field. As previously noted, associations between incidence of *Fusarium* head blight and application of glyphosate have been observed in field surveys (Fernandez et al. 2009), and fungicide application at early developmental stages of the cereal plants may stimulate development of *Fusarium* spp. and mycotoxins (Henriksen and Elen 2005). Knudsen et al. (1995) speculated whether pathogenic *Fusarium* species may be suppressed by antagonistic *Fusarium* spp. to a larger extent in an organically cultivated field than in a conventionally cultivated field. A higher number of antagonistic *Fusarium* spp. and a higher total number of non-pathogenic *Fusarium* spp. have been recorded in organically farmed fields than conventionally cultivated fields (Knudsen et al. 1995). In a review, Lairon (2010) concluded that mycotoxin contamination of cereals is widespread, but at a low level, and that an organic or conventional modes of production do not result in overall noticeable differences. Furthermore, it was concluded that the preventive measures used in organic systems, despite the lack of fungicide use, appears able to maintain mycotoxin contamination at a generally low level.

Fertilizer usage might also influence the occurrence of *Fusarium* and mycotoxins. Development of *Fusarium* head blight in wheat has been found to increase with increasing nitrogen input (Lemmens et al. 2004b). Other studies have also shown a significant association between use of mineral fertilizer and increases in *Fusarium* and DON (Martin et

al. 1991; Elen et al. 2000; Yi et al. 2001; Heier et al. 2005). Based on a literature survey, Champeil et al. (2004) stated that the effects of mineral nutrition on *Fusarium* head blight are unclear, and that different forms of nitrogen may have different effects on *Fusarium* head blight. In a Norwegian survey, by using a multivariate statistical test, Bernhoft et al. (2012) reported that the use of mineral fertilizers was highly significantly associated with increased levels of *Fusarium graminearum* in cereal. A higher humidity in a denser stand on farms applying mineral fertilizer cannot be excluded as a factor that could be conducive to infection and mycotoxin production. However, excess nitrogen supply may also influence the cell wall structure and chemical composition of the plants (van Arendonk et al. 1997), possibly making them more susceptible to fungal attack.

There are studies where no significant difference could be detected in mycotoxin contamination of cereals between organic and conventional production systems (Edwards 2009c; Edwards 2009a; Hoogenboom et al. 2007; Jestoi et al. 2004). However, several studies report that organically produced cereals contain less mycotoxins than conventional cereals (Doll et al. 2002; Schollenberger et al. 2005; Bernhoft et al. 2010; Bernhoft et al. 2012; reviewed by Köpke et al. 2007). A recent systematic review of organic and conventional foods related to safety and healthiness, reported lower levels and lower risk of contamination with DON in organic grains than conventional alternatives (Smith-Spangler et al. 2012).

There are no Norwegian field experiments where mycotoxin contamination in organic and conventional cereals are compared. More research is needed to clarify if there are differences in the risk of mycotoxin contaminations between organically and conventionally produced cereals, and if the use of pesticides and fertilizers in conventional farming systems influence on the risk of *Fusarium* infection and mycotoxin contamination.

4.4 Storage conditions

Besides the presence of nutrients, the most important factors for fungal growth, and subsequent production of mycotoxins, are water activity (a_w), temperature and oxygen.

A_w is a measure of the unbound water in the product, which is available for fungal growth. Fungi are generally able to grow at a lower aw than bacteria (Figure 11). Fungal growth can be prevented by drying a product to a level below 0.65 a_w and keep it at that level. The growth of bacteria becomes less likely at pH levels of less than 5 and a_w levels of below 0.9. However, this is not enough to inhibit the proliferation of food spoilage fungi.

Table 6 shows the minimal water activities of species belonging to the associated mycobiota of cereals and cereal products.

Table 6. The minimal water activities (a_w) of species belonging to the associated mycobiota of cereals and cereal products (Samson et al. 2010).

Minimal a_w	Fungal species
0.85 - 0.89	*Fusarium avenaceum, F. culmorum, F. graminearum, F. poae*
0.80 - 0.84	*Byssoclamus nivea, Penicillium citrinum, P. verrucosum, P. roqueforti,*
0.75 - 0.79	*Aspergillus candidus, A. flavus, A. parasiticus, A. ochraceus, Paecilomyces variotii, Penicillium aurantiogriseum*
0.70 - 0.74	*Eurotium repens, E. rubrum*

However, if the temperature in the product or the storage environment is not uniform, the a_w can rise locally due to condensation, thus leading to a rise in a_w and subsequently fungal

growth. Concerning temperature, *Pencillium* spp. in general have a lower minimal temperature range than *Aspergillus* spp.

Oxygen is usually necessary for the growth of fungi, but certain species can also grow under anaerobic or microaerophilic conditions. Oxygen also influences production of mycotoxins.

The pH has a limited influence of the growth of storage moulds, but may influence on the production of mycotoxins. However, as pH may restrict growth of other microorganisms, it will indirectly affect the growth of moulds (Figure 11).

The presence of other microorganisms may also restrict the fungal growth and mycotoxin production. For instance may *Aspergillus flavus* produce only little aflatoxin in the presence of other moulds.

The presence of grain-infesting insects and mites and the general condition of the grain can influence on the fungal growth and mycotoxin accumulation.

Finally, time is a factor that must be considered.

Cereal grain carries a wide range of microorganisms including fungi, yeast and bacteria when entering stores on farms or in silos. The population structure depends mainly on the climatic conditions during development of the plants in the field prior to maturation and until harvest, but also agronomic practice and cereal variety will influence on the diversity of microbial contaminants. The mycobiota is defined as the total fungal inventory of the area/product under consideration. However, only a restricted number of fungal species, normally less than 10, is actually able to spoil a particular product. We refer to this as "the associated mycobiota" of a product.

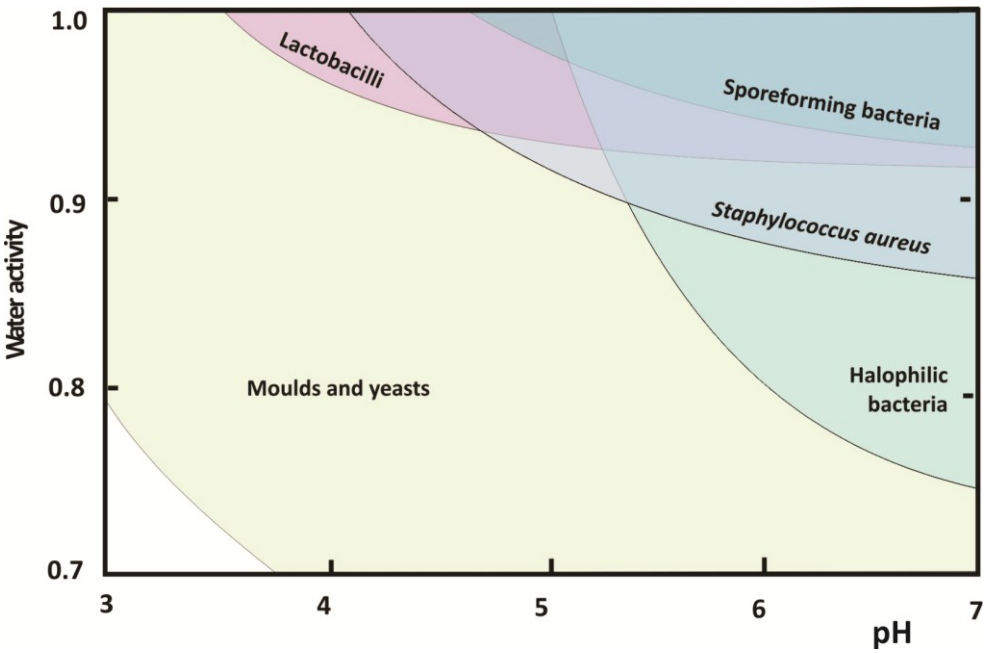


Figure 11. A schematic diagram showing the combined influence of water activity (a_w) and pH on microbial growth (Pitt and Hocking 2009).

During storage with low a_w and low temperature, only minor changes occur in the composition of the microorganisms of the cereal grain. By storing grain at a moisture content of about 14 % and at a temperature of 20°C to 25°C for some months, most field fungi are

eliminated without detectable invasion of the grain by storage fungi (Follstad and Christensen 1965). Under the usual conditions of storage of food and feed grain, *Fusarium* may die within a few months (Christensen and Kaufmann 1965).

However, fungi can grow and produce toxins during transport, storage, and processing of feed and food if the moisture content and prevailing environmental conditions are favourable. Because of their relative stability, no significant reduction in trichothecene levels occurs during storage and drying operations. *Fusarium* toxin production may continue during storage under appropriate conditions. Homdork et al. (2000) studied wheat samples with different levels of *F. culmorum* held in different storage conditions. They found that zearalenone accumulated to higher contents under warm and humid conditions (25°C at 90 % relative humidity (RH)). The mycotoxin was produced only after a long period of storage, especially in the severely infected samples (> 50 % *Fusarium*) which reached up to 5070 µg/kg. DON content of severely infected samples remained unchanged (around 2500 µg/kg) under any of the conditions tested and increased in samples with a slight (4 %) and moderate (15 %) *Fusarium* infection level, from < 500 to 1935 and 3270 µg/kg respectively, when the grain was stored under warm and humid conditions. Nivalenol was not found in any samples immediately following harvest, but was detected after storage in humid conditions (15 °C and 84 % RH, and 25 °C and 90 % RH). Under warm, dry conditions (25°C and 62 % RH), the seed germination rate showed a slight increase or remained nearly constant, and the *Fusarium* infection level of the kernels decreased relatively rapidly. Homdork et al. (2000) concluded that cool, dry storage conditions are required in order to maintain good seed and grain quality, and that even a slight *Fusarium* infection can result in an increase in mycotoxin production under improper storage conditions. Homdork et al. (2000) also observed that *Fusarium* was suppressed by competing storage fungi, mainly *Aspergillus* and *Penicillium*, under warm, humid storage conditions. During storage, the content of *Fusarium* toxins in the samples did not reflect the percentage of *Fusarium*-infected kernels.

Birzele et al. (2000) also concluded that trichothecene concentrations, like DON, can increase significantly in the presence of *Fusarium* inoculum if the humidity level exceeds the safe storage level, e.g. 15 % water at 20°C.

Langseth et al. (1993) demonstrated that drying wheat grain with an initial moisture content of 32 % with different intensities of forced ambient air, followed by storage, resulted in a *Fusarium* population that remained metabolically active until at least 5 weeks of storage.

Naik et al. (1978) found that toxin production may or may not be enhanced at low temperatures. Five strains of *F. graminearum* produced significant amounts of zearalenone at 25°C, but only one strain produced toxin at 10°C.

Interactions between mycotoxigenic fungi and insects occur, but they have not been studied thoroughly. Some insects disseminate mycotoxigenic species, others are known to use moulds as a food source, while others avoid certain fungal species. As noted by Magan et al. (2003), a more holistic ecological view is needed when considering management approaches for safe storage of cereal grain after harvest.

4.5 Future trends

The observed climate trends of increased temperatures lead to a change in the Norwegian mycobiota. New mycotoxin-producing species, combined with the possibility of changed rainfall patterns, can be of vital importance for the risk of mycotoxins in Norwegian cereals. The observations recorded until now, of increased rainfall during critical periods, are still

within the range of natural climate variability. Nevertheless, this pattern is in line with the projected scenarios of climate change. However, the uncertainties related to climate change projections for rainfall are much higher than those for temperature, due to different levels of understanding of these two phenomena in climate physics.

Further improvements in the prediction models can be expected. However, even a perfect model will still struggle with variations in the development stage of the crop, even within single fields. Limitations in what can be achieved regarding the principle of predicting the optimal timing of fungicide treatment cannot be avoided. Practical constraints will remain regarding optimal application of fungicides to prevent disease development. Even within single cereal fields, there will be variations in the stage of development, and hence the timing of maximum susceptibility; it would be practically impossible to divide up work operations at such a degree of resolution. Similarly, models predicting post-harvest contamination levels will be subject to equivalen constraints.

Other factors that affect the balance of inoculum build-up and depletion are likely to be the focus of greater attention, because they are considered to be the primary underlying causes for the rise in inoculum levels of mycotoxin-producing fungi in cereal crops in Norway.

Factors affecting infection and the production of mycotoxins can be separated into controllable and uncontrollable factors. The most important uncontrollable factor is the weather.

Further improvements in the predictive models and their use in mitigating the effects of uncontrollable factors such as weather should be encouraged. Nevertheless, it is likely that the future trend in the management of the mycotoxin problem complex will tend focus to a greater extent on controllable factors.

Various approaches to reducing the saprophytic stage, and the ability of mycotoxin-producing fungi to produce inoculum, e.g. accelerated plant debris decomposition and/or reduced access to plant debris by burial, chemical killing of the fungus at this stage, are likely to be the subjects of greater attention. However, the plant residues act as a soil-binding factor and have environmental implications due to the roles they play in reducing nutrient leakage and soil erosion. Thus, plant residues remaining after harvesting a crop, are a key factor, both in relation to the process and the effects of soil tillage and crop rotation on mycotoxin contamination of cereals in the field.

It is still not fully understood how growth and mycotoxin production of storage fungi could be prevented, and further investigation is required. The interactions between different microorganisms and fungi in the field and in the storage environment, and the possible impact of biofilms in this context, also need more attention.

Novel approaches: future insights on mycotoxin gene expression may provide new possibilities for controlling plant pathogens like *Fusarium*. Ismail et al. (2011) found that a fungal symbiont of plant-roots modulated mycotoxin gene expression in the pathogen *F. sambucinum*. Such an effect, which had not previously been described, may be an important mechanism for biological control and has fascinating implications for advancing our knowledge of plant-microbe interactions and controlling plant pathogens.

4.6 In summary

The scientific information about the epidemiology of mycotoxin-producing fungi indicates that processes aiding inoculum build-up have strong agronomic causality, and these causes are disadvantageous to the inoculum-depleting processes, both at the local and regional scale. It is

likely that an alteration in the balance between inoculum build-up and depletion processes is the primary factor responsible for the trend of increased frequency and severity of *Fusarium* epidemics in Norway.

In additon, weather conditions clearly play an important role as an uncontrollable factor in modulating the temporal severity of the epidemics. The repeated severity of epidemics during the three growing seasons 2008-2010 coincides with the occurrence of weather conditions conducive to infection at the susceptible phenological stages of cereals. Moreover, the potential long-term role of weather patterns in explaining the increased frequency and severity of infection with mycotoxin-producing fungi also seems plausible in the Norwegian situation, and is in line with projected climate change scenarios, although the data have not yet reached beyond the scale of natural climate variability.

One of the main challenges to cereal cultivation in Norway is an extensive cereal monoculture, with limited use of crop rotation in combination with reduced tillage. Both factors may contribute to the increased occurrence of mycotoxin-producing fungi. The influence of pesticides (such as glyphosate) on *Fusarium* and mycotoxin development in cereals remains unclear.

Four different cultural practices are currently considered to be of importance for reducing *Fusarium* head blight and the production of mycotoxins. These are: i) ploughing to reduce inoculum in soil, ii) the choice of the preceding crop in the rotation, iii) the choice of appropriate cereal varieties, and iv) use of fungicides. As the T-2 and HT-2 content in oats is not reduced by fungicide treatment, focus should be directed towards identifying fungicides for effective control of *F. langsethiae* and T-2 and HT-2 toxins. Organic cereal production, with its soil fertility management without synthetic pesticides and fertilizers, and with few cereal-intense crop rotations, seems to be another strategy to reduce *Fusarium* head blight and the production of mycotoxins.

5 Effects of grain handling, processing and mitigation procedures

In the work to reduce the risk of mycotoxins from grain in food and animal feeds, the main focus should always be on diminishing the growth of fungi and the production of mycotoxins in the grain. However, as discussed in Chapter 4, experience has shown that in some years, it is difficult to avoid substantial mycotoxin contamination in Norwegian cereal grain, in particular with DON and other trichothecenes.

This situation necessitates the use and/ or consideration of different measures to reduce the amount and effects of mycotoxins, and of trichothecenes in particular, in the final products. The relevant measures available can be broadly divided into the following categories:

1. Organisational, logistical and technical procedures to reduce or adjust the amount of mycotoxins in cereal-based foods and feeds.
2. Physical or chemical treatment of the cereal in order to destroy the mycotoxin molecules and change them into less toxic substances.
3. Addition of chemical adsorbents ("mycotoxin binders") to mycotoxin-contaminated feeds, in order to reduce the absorbtion of mycotoxin in animals eating the feed.
4. Addition of microbes or microbial products intended to degrade mycotoxins in contaminated feed and convert them into less harmful products.

In EU there are maximum limits for DON in grains for production of food as well as for food products as sold. Dilution of mycotoxin levels by mixing contaminated baches of grain with bachtes containing lower levels for food production is not allowed. Foodstuffs shall not be deliberately detoxified by chemical treatments.

5.1 Organisational, logistical and technical procedures

Measures to reduce mycotoxin contamination of cereals in the field have been discussed in Chapter 4. After harvest the first and most important procedure to avoid serious mycotoxin contamination of grain products is the inspection and control of cereal lots at deliverance to the reception units of the grain companies. In Norway, reception and control of harvested cereals was previously the responsibility of a government-owned company. The task has now been delegated to the private companies; with the farmer-owned company "Felleskjøpet" receiving the greater part of the harvest. Only minor amounts of cereals are kept and used locally, by the producing farmers. About 75 % of the harvest each year is delivered directly to the cereal company, in the harvest season (Røhne, Felleskjøpet, personal statement). The rest is delivered later in autumn or in the winter, after drying and temporary storage on the farms.

At deliverance, each lot of cereal is evaluated by visual inspection. Most lots of oats and a fair percentage of other grain lots are also tested for DON level at deliverance, using a DON-specific quick test (Stokke, Felleskjøpet, personal statement). Seriously damaged grain is rejected. Less damaged, but still visibly affected lots of cereals are stored separately, until results of further screening analysis for mycotoxins are available. DON quick test results are used for the first sorting of delivered grain into quality classes suited for different usage.

After reception and primary storage, screening samples are taken both of suspected batches and as average samples from larger amounts of "normal" grain. In addition to analysis of water content, the samples are screened for total amount of fungi (recorded as "colony forming units" - cfu), amount of *Fusarium spp*. and concentrations of DON. A percentage of

the screening samples are also tested for concentrations of T-2 toxin, nivalenol, zearalenon and other toxins. Most of the DON analyses and some of the other analyses are performed in the company laboratories, by an Elisa technique. A number of representative samples of stored batches are also delivered to the Norwegian Veterinary Institute for more exact and extensive analysis of fungal contamination and mycotoxin concentrations (Røhne, Felleskjøpet, personal communication). Starting in 2011, payment to producers of oats in Norway is adjusted according to DON concentrations measured at deliverance (Røhne, Felleskjøpet, personal communication). The gradual price reduction in payment according to DON content in the grain delivered in 2012 is shown in Appendix A, Table A9.

The results of the different analyses are used to direct the subsequent handling of the cereal. Lots with too high levels of mycotoxins are destroyed or used for bioetanol production. Lots with moderately elevated levels are used in concentrates for ruminants or chicken , while the lots with low DON levels are reserved for human food and pig feeds. Some lots with moderate levels may be blended with low-level lots to achieve an average in the final product that is within the acceptable limits for pig feeds. Dilution of lots of oats containing more than 1750 µg DON/kg with low-level lots is not allowed for food production.

In oats, mycotoxin levels may also be lowered by dehulling. This is done to some extent, and dehulled oats are primarily used in feed for piglets. Dehulling may reduce the DON contamination in the oats groat by 30-60%, depending on the degree of hull removal (Røhne, Felleskjøpet, personal communication). In a pilot study at Norwegian Veterinary Institute (Clasen et al. 2002) of trichothecenes in paired samples of oats hull and kernels, most of the hull samples contained T-2 and HT-2 (mean conentrations 226 and 98 µg/kg, respectivey). No quantifiable concentrations of HT-2 or T-2 were shown in the oats kernels (< 20 µg/kg). DON was present at low concentration in a few hull samples but in none of kernel samples. Dehulling is a normal procedure in the production of food from oat grains, and does not compromise the quality of the final product. In a Finnish study 50-90 % of the DON and 3-acetyl DON were removed by sorting and dehulling grain of barley and oats (Hietaniemi et al. 2009). After dehulling, no T-2 and HT-2 were detected in the samples. For wheat there seem to be less differences of mycotoxin concentration (DON) in hull and kernels.Trichothecenes are not destroyed but unevenly redistributed between fractions during the milling process. The toxins are normally mostly attached to the outer hull of the grain and cleaning, sorting, sieving and de-hulling of grain lead to marked increases in trichothecenes in cereal by-products, e.g. bran, and a decrease in the flour fraction. It has, however, been reported that the distribution of DON into the milled wheat fractions was dependent on the degree of fungal penetration into the endosperm of the wheat kernel, and that this penetration was variety-dependent (Nowicki et al. 1988). It is also noteworthy that higher concentrations of DON in the grain has been reported to result in a greater proportion of DON within the flour fraction after milling (Edwards et al. 2011). This finding indicates that the reduction of DON concentrations during the milling process is lower in years with high DON levels.

By harvesting wheat with higher fan speed and air flow a significantly lower content of *Fusarium* damaged kernels and DON compared to standard harvesting were obtained in a study in Ohio (Salgado et al. 2011). However, the harvesting configurations that resulted in the best grain quality also resulted in significant reductions in the volume or weight of harvested grain. Removing of outer layers (pericarp) of wheat kernels by controlled debranning revealed that most of the microbial contaminations (including *Fusarium*) were located close to the surface (Laca et al. 2006). This work showed that debranning technology can provide cereal of high microbial purity.

5.2 Physical or chemical treatment

Thermal treatment is only moderately efficient in removal of mycotoxins, because most mycotoxins are rather heat-stable (Kabak 2009; Jard et al. 2011) Still, roasting and other kinds of heating are used to lower the amount of aflatoxins, both in food and in feeds (Rustom 1997).

In aquaculture, the feeds used today are based on high-temperature extrusion processes that allow the regulation of the floating/sinking capacity of the pellets while maintaining the nutritional values of the aquafeeds. Production of extruded catfish feed (cooker-extrusion and heat drying) caused an up to 60% reduction in aflatoxins (Manning et al. 2005). The effect of extrusion techniques on other mycotoxins such as fumonisin and trichothecenes is less documented, but seems to be in the same range. Generally, the amount of mycotoxin reduction is highly dependent on extrusion conditions, such as temperature, time, pH and amount of water (Kabak 2009).

Ammoniation has been used for some years to reduce the level of aflatoxins in maize for feed. The method is costly, may reduce feed quality, and is less effective against other mycotoxins (Jard et al. 2011; Huwig et al. 2001). Chemical oxidation with hydrogen peroxide or ozone has been shown to be able to reduce the amount of several mycotoxins (McKenzie et al. 1997), including trichothecenes (Young 1986). Treatment with other reactive chemicals, such as sodium bisulphite, sodium nitrites or strong acids has also been tried. However, all these methods have limitations similar to those of ammoniation (Jard et al. 2011).

The degradation of mycotoxins in normal food processing procedures has been reviewed in several occasions, including in the international risk assessments of each toxin and by Hazel and Patel (2004), Bullerman and Bianchini (2007), and by Castells et al. (2005). The reduction in mycotoxin processes during normal baking of bread is variable, ranging from no significant reduction to > 50 % reduction of DON during the baking process (Hazel and Patel 2004; Bretz et al. 2006; Lancova et al. 2008; Schwake-Anduschus et al. 2010; Voss and Snook 2010). The chemical and biological processes occurring during food processes such as baking are comples and dependent on several parameters including yeast strain, the fermentation process, temperature, moisture and ingredients (Bergamini et al. 2010). Cooking of pasta reduces the concentrations of water-soluble toxins such as deoxynivalenol. The reduction is dependent on the pasta to water ratio. The decomposisition of several mycotoxins including trichothecenes, zearalenone and aflatoxins are significantly increased during alkaline cooking procedures commonly used when preparing tortillas. A significant reduction of mycotoxin concentrations may occur during extrusion cooking, a process which includes high temperatures and pressure. The degradation during the extrusion cooking process is dependent on a range of physiochemical parameters during the process, including pH, temperature, moisture, pressure, residence time in the extruder and protein concentrations (Castells et al. 2005, 2006; Wu et al. 2010) and even increased concentrations of mycotoxins, particularly nivalenol, after extrusion under some conditions have been reported (Scudamore et al. 2008).

5.3 Addition of chemical adsorbants (“mycotoxin binders”) to contaminated feeds

Internationally, a range of adsorbant materials have been tried as feed additives, in attempts to reduce the absorption of mycotoxins. Materials used or tested for this purpose include different kinds of aluminosilicates, natural glucomannans and related products, synthetic

organic polymers, and activated carbon (Boudergue et al. 2009). Most of the aluminosilicates used are derived from natural clays, either used in the natural form (bentonite, zeolite etc.), or as a derived, further processed product (e.g. hydrated sodium calcium aluminosilicates – HSCAs). The latter is also commonly used as an anticaking addition in animal feeds. The glucomannans are complex carbohydrate products made from the cell walls of yeast; especially from *Saccharomyces cerevisiae*. The main synthetic organic polymer tested is polyvinylpolypyrrolidone.

Trichothecenes show generally weak adherence to adsorbants, and addition of other adsorbants than activated charcoal to feed are generally inefficient in counteracting their effects. This is especially true for mineral adsorbants, which are generally agreed to be ineffective (Boudergue et al. 2009). With glucomannans, significant binding of trichothecenes has been shown *in vitro* (Cavret et al. 2010; Freimund et al. 2003), but studies *in vivo* have not been able to confirm an effect on the toxicity of trichothecene-contaminated feed (Diaz-Llano and Smith 2006; Wetscherek 1998)

Activated carbon binds most of the mycotoxins studied *in vitro*. Even DON has been shown to be adsorbed rather efficiently to activated carbon (Döll et al. 2004). However, because of its lack of specificity as an adsorbant activated carbon will bind all kinds of substances, including important nutrients. For these and several other reasons activated carbon is impractical for long-term dietary inclusion in feed (Boudergue et al. 2009), although it is an indispensable remedy in the treatment of acute poisoning, both in man and animals.

Aluminosilicates – especially HSCAs - have been shown to be quite effective in binding aflatoxins (Boudergue et al. 2009; Phillips et al. 1988). The binding is both efficient and specific, and addition of HSCAs to feed has been shown to effectively reduce the toxic effects of aflatoxins, both in poultry and in pigs (Harvey et al. 1989; Kubena et al. 1990). Use of HSCA as a feed additive has also been shown to reduce the aflatoxin content in animal products (Beaver et al. 1990; Kutz et al. 2009; Smith et al. 1994). In contrast to terrestrial farmed animals only a few studies have been performed with aluminosilicates against aflatoxin contamination in farmed finfish species. In Nile tilapia a diet supplemented with Egyptian bentonite and montmorillonite clays gave a significant reduction on aflatoxin B1 induced toxicity (Hassan et al. 2010), and intragastric dosing of montmorillonite clay mineral reduced the absorption and genotoxicity of sterigmatocystin, which is a precursor of aflatoxin (Abdel-Wahhab et al. 2005). In juvenile silver catfish that were fed sodium and calcium aluminium silicate as adsorbent in diets contaminated with aflatoxin, the adsorbent caused reduced feed intake and growth due to loss of palatability (Lopes et al. 2009). In rainbow trout bentonite clay reduced the bioavailability of ^{3}H labelled aflatoxin B1 in contaminated feed (Ellis et al. 2000).

For other mycotoxins, however, the effect of aluminosilicates is more variable (Boudergue et al. 2009). In an *in vitro* study of the binding of *Fusarium* toxins to different adsorbants (Döll et al. 2004), a modified aluminosilicate showed substantial ability to bind zearalenone. However, in a follow-up study *in vivo*, addition of the modified aluminosilicate to the feed had no effect, neither on the general performance of pigs, nor on the specific zearalenone-dependent changes in female sex organs (Döll et al. 2005). Other researchers have observed a positive effect of zeolite added to a zearalenone-contaminated feed on the zearalenone-induced increase in uterus weight in piglets (Coenen and Boyens 2001).

Yeast-derived glucomannans have been shown to be effective in reducing the toxicity of aflatoxins, especially in poultry (Boudergue et al. 2009; Karaman et al. 2005). In addition, glucomannans have shown positive effect against the toxicity of both ochratoxin A (Aravind et al. 2003; Raju and Devegowda 2000) and zearalenone (Aravind et al. 2003) in poultry.

There are indications that glucomannans may also have effect against zearalenone toxicity in pigs, though the documentation is weaker (Sinovec 2006).

Polyvinylpyrrolidone has only shown effect against aflatoxins (Celik et al. 2000).

5.4 Addition of microbes or microbial products intended to degrade mycotoxins in contaminated feed

A large number of microbes have been tested *in vitro* and been confirmed to be able to degrade mycotoxins, in particular aflatoxins (Boudergue et al. 2009). However, only a limited number have also been tested in animals, and even fewer developed into products marketed for detoxifying mycotoxins in feed. The most important microbial agents reported to detoxify mycotoxins *in vivo* are *Eubacterium BBSH 797* and *Trichosporon mycotoxinivorans*.

Eubacterium BBSH 797 is a microbe isolated from rumen fluid (Binder et al. 1997), and patented for use as a mycotoxin detoxifying agent, with specific activity against trichothecenes. *In vitro*, it has been shown to degrade both type A and type B trichothecenes, and break them down to much less toxic de-epoxy metabolites (Binder et al. 1998; Fuchs et al. 2002).

A feed additive containing this microbe as active ingredient did counteract the growth reduction induced by a diet containing naturally contaminated feed with 2.5 mg DON per kg in piglets (Plank et al. 2009). In data delivered to the European Food Safety Authority (EFSA), four other experiments are described with pigs given feed contaminated by *Fusarium* toxins (Anadon et al. 2005). In one of these, also in piglets, the *Eubacterium*-based feed additive did improve growth significantly, compared to a group given the contaminated feed without the feed additive. In another experiment in piglets and two experiments in fattening pigs, the groups given the feed additive had higher mean growth than the control groups, but the differences were not statistically significant.

In chickens, a commercial product with *Eubacterium BBSH 797* and adsorbent minerals did counteract the negative effect of 1 mg/kg feed of purified diacetoxiscirpenol (DAS) in a three week feeding experiment. However, with 2 mg/kg diacetoxyscirpenol, the effect of the *Eubacterium*-containing product was not significant (Diaz 2002).

Trichosporon mycotoxinivorans is a yeast isolated from the hindgut of termites. It is shown to degrade both ochratoxin A and zearalenone *in vitro* into less toxic metabolites (Molnar et al. 2004; Schatzmayr et al. 2006). However, no feed additive with this microbe as the single active ingredient seems to have been tested *in vivo*.

The company owning the patent of both these microbes have combined them in a product (Mycofix® Plus) that also includes adsorbent minerals and some other constituents (Biomin GmbH 2009, 2011). The fact that the brand name Mycofix® has been used on several generations of the company's mycotoxin-detoxifying products, with different composition, may lead to some bewilderment in the evaluation of different published studies. With this precaution in mind, there seem to be inconsistent results concerning the effect of the combined product *in vivo* (Boudergue et al. 2009). In one trial with dietary T-2-toxin in chickens, the combined Mycofix® product did alleviate the adverse effects of T-2-toxin, in contrast to aluminosilicates and esterified glucomannans (Diaz et al. 2005). In two other studies, the combined product had only small effect *in vitro* (Döll et al. 2004), and no effect in pigs fed naturally DON-contaminated wheat *in vivo*, neither on the excretion of DON in pig urine nor on the performance of the pigs (Dänicke et al. 2004).

A third line of development involves the isolation of trichothecene-degrading microbes isolated from chicken intestines (He et al. 1992; Young et al. 2007). However, so far commercially available feed additives based on these microbes have not been marketed.

Addition of *Saccharomyces cerevisiae* has been shown to counteract the effects of aflatoxin feed contamination on Japanese quail (Parlat et al. 2001). This effect may be related to the glucomannan content of the yeast cell walls. Addition of the bacteria *Nocardia corynebacteroides* has also been shown to counteract aflatoxin toxicity in broilers (Tejada-Castaneda et al. 2008). As for the microbes from chicken intestines, further development of a feed additive from *Nocardia spp*. has not been reported (Boudergue et al. 2009).

In conclusion, more studies are clearly needed before the practical usefulness of these mycotoxin-degrading microbial products can be finally established.

5.5 In summary

The most important technical procedures to avoid elevated levels of DON and other mycotoxins in food and feed in Norway is the inspection, analyses and sorting of delivered grain according to quality and mycotoxin concentrations.

In oats, dehulling is also highly effective to reduce trichothecene levels in the final product.

Dilution of mycotoxin levels by mixing contaminated baches of grain with bachtes containing lower levels for food production is not allowed. Foodstuffs shall not be deliberately detoxified by chemical treatments.

Addition of chemical adsorbants, though important in the treatment of aflatoxin-contaminated grain, has so far not been documented to be effective against trichothecenes in feed. There may be a risk for binding of chemical adsorbants to other kind of substances including important nutrients. In particular, adsorbant activated carbon lack specificity and will bind all kind of substances. Among potential mycotoxin-degrading microbes tested, the rumen bacteria BBSH 797 has shown good trichothecene-degrading effect *in vitro*, but the product needs further testing of the trichothecene-reducing and clinical effect *in vivo*. If found to be effective, the risk in using these products in feed is expected to be insignificant.

6 Occurrence of mycotoxins in cereals

The occurrence of mycotoxins in cereal grain and products thereof has been analysed at the Norwegian Veterinary Institute for many years. All methods are accreditated methods and the laboratory is the National Reference Laboratory for mycotoxins. The methods have been slightly modified over the years and the limits of detection and limits of quantification have been in the same range throughout this period and are given in the table below. In addition, samples have been sent to other accredited laboratories, when there have been technical problems or during holiday seasons. For these reasons, the limits of detection (LOD) have varied over the years. Typical values for the LODs are given in Table 7.

Table 7 Typical limits of detection (LOD) for various mycotoxins in grains (µg/kg)

Toxin	DON	T-2	HT-2	NIV	ZON	BEA	ENN	MON	OTA	Aflatoxins				FUM
										B1	B2	G1	G2	
LOD	20-40	2.5-30	2.5-20	5-30	2.0	3.0	3.0 (4.0 for A1)	40	0.015	0.25	0.1	0.2	0.15	10

DON: deoxynivalenol, **NIV**: nivalenol, **ZON**: zearalenone, **BEA**: beauvericin, **ENN**: enniatins, **MON**: moniliformin, **OTA**: ochratoxin A, **FUM**: fumonisin.

6.1 Occurrence in crude grain

The occurrence of selected mycotoxins in Norwegian and imported crude cereal grain are summarised in Tables 8-10 and more detailed data on the occurrence are given in the Appendix B. In addition a limited number of samples of compound feed have been analysed (Table 14). In recent years a number of processed grain products intended for human consumption have been analysed for the most common *Fusarium* mycotoxins in cereals (Table 17).

The occurrence data used in this assessment are mainly from the database at the Norwegian Veterinary Institute. The main part of the samples originates from the national monitoring programs for mycotoxins in feed and food organised by the Norwegian Food Safety Authority. The sampling has been carried out according to the guidelines prepared by the Norwegian Food Safety Authority based on the EU guidelines to be representative of the levels in feed and food. Additional data from samples of cereal grains from specific studies analysed at the Norwegian Veterinary Institute have been included, when the samples are considered to represent the levels in cereals for feed and food production. Potential outliers and samples from non-representative studies have been excluded from the dataset. The analysis are carried out at the Norwegian Veterinary Institute or in some cases at other accredited laboratories.

6.1.1 Wheat

DON is the mycotoxin most frequently found in wheat (Table 8). The other trichothecene mycotoxins nivalenol, T-2 toxin and HT-2 toxin are found at lower frequency (see Appendix B) and in lower mean concentrations. There is a considerable annual variation in mean DON-concentrations ranging from 16 µg/kg in 1999 to 405 µg/kg in 2011. The annual variation is influenced on the weather conditions during the growth season as well as several other factors (see Chapter 4).

Table 8 Mycotoxins in wheat grain in Norway. Mean middle bound concentrations (µg/kg) with sample numbers/ number of samples below limit of detection in parenthesis, are given. All data are from the Norwegian Veterinary Institute.

Year	DON	T-2*	HT-2*	NIV*	OTA	ZON	MON	ENN	BEA
1990	104 (165/29)	n.a.	n.a.	27 (165/164)	0.2 (165/160)	2.7 (165/160)	n.a.	n.a.	n.a.
1991	84 (107/7)	n.a.	n.a.	35 (107/80)	n.a.	n.a.	n.a.	n.a.	n.a.
1992	218 (128/18)	n.a.	n.a.	25 (128/128)	n.a.	n.a.	n.a.	n.a.	n.a.
1993	161 (131/48)	n.a.	n.a.	25 (131/131)	0.7 (18/10)	n.a.	n.a.	n.a.	n.a.
1994	50 (142/87)	n.a.	n.a.	26 (142/138)	0.4 (33/28)	n.a.	n.a.	n.a.	n.a.
1995	71 (39/20)	n.a.	n.a.	25 (39/39)	0.3 (45/39)	3.0 (45/43)	n.a.	n.a.	n.a.
1996	118 (42/35)	15 (42/42)	10 (42/42)	10 (42/42)	0.4 (42/40)	10 (42/42)	n.a.	n.a.	n.a.
1997	108 (38/22)	19 (38/35)	19 (38/35)	21 (38/31)	0.2 (38/12)	2.6 (38/27)	n.a.	n.a.	n.a.
1998	39 (59/31)	15 (59/59)	10 (59/59)	11 (59/51)	0.5 (59/56)	1.3 (59/54)	n.a.	n.a.	n.a.
1999	16 (83/u)	14 (83/u)	14 (83/u)	12 (83/u)	n.a.	n.a.	n.a.	n.a.	n.a.
2000	109 (59/u)	13 (59/u)	13 (59/u)	11 (59/u)	n.a.	n.a.	20 (13/u)	126 (12/u)	1.5 (12/u)
2001	155 (18/u)	5.3 (18/u)	6.1 (18/u)	5.3 (18/u)	n.a.	n.a.	92 (35/u)	730 (34/u)	1.5 (34/u)
2002	62 (74/u)	15 (74/u)	9.9 (74/u)	15 (74/u)	n.a.	n.a.	210 (35/u)	297 (34/u)	1.5 (34/u)
2003	83 (88/u)	15 (88/88)	10 (88/88)	15 (88/88)	n.a.	n.a.	n.a.	n.a.	n.a.
2004	256 (60/u)	15 (60/60)	10 (60/60)	15 (60/60)	n.a.	n.a.	n.a.	n.a.	n.a.
2005	209 (93/u)	15 (93/93)	10 (93/93)	15 (93/93)	n.a.	n.a.	n.a.	n.a.	n.a.
2006	153 (22/u)	14 (22/u)	9.9 (22/u)	15 (22/u)	n.a.	n.a.	n.a.	n.a.	n.a.
2007	130 (6/u)	13 (6/u)	9.2 (6/u)	13 (6/u)	n.a.	n.a.	n.a.	n.a.	n.a.
2011	405 (30/0)	15 (30/30)	10 (30/30)	15 (30/29)	n.a.	n.a.	n.a.	n.a.	n.a.

DON: deoxynivalenol, **NIV**: nivalenol, **OTA**: ochratoxin A, **ZON**: zearalenone, **MON**: moniliformin, **ENN**: enniatins, **BEA**: beauvericin, **n.a.**: not analysed, **u:** unknown number of samples below LOD, but it is known that half LOD is used in the calculation of mean, *****mean concentrations below the LOD.
Samples below LOD are considered with a value of ½ LOD for the calculation of the mean values. LODs (µg/kg); DON: 20, T-2: 3-30, HT-2: 3-20, NIV: 5-30, ZON: 2.0-3.0, Enn: 3.0 (4.0 for Enn A1), Bea: 3.0, MON: 40, OTA: 0.015. Wheat grain not been analysed for aflatoxins or fumonisin.

Both the mean concentrations and the incidence of positive samples are much lower for T-2, HT-2 and nivalenol than for DON. The mean concentrations are respectively 5-19 and 6-19 µg/kg for T-2 and HT-2, and at a similar level for nivalenol (Table 8). The mean calculated concentrations for T-2, HT-2 and nivalenol are below the LOD and are uncertain.

Other *Fusarium* toxins have only been analysed in crude wheat samples in a limited number of years. Zearalenone was analysed in crude wheat in the time periode 1990 – 1998. It was

concluded that the levels were low and probably not of any concern. Consequently the toxin was not included in the monitoring in following years.

Moniliformin, enniatins and beauvericin were analysed in a selection of the crude wheat samples in the years 2000-2002 (Table 8).

In addition, wheat samples were analysed for ochratoxin A for the years 1990 – 1998. The concentrations were very low, demonstrating that the current procedures of drying and storage are sufficient to limit the production of this storage toxin.

6.1.2 Oats

Crude oats are the grain most frequently contaminated by trichothecenes, and also the cereal species where the highest concentrations are normally found (Table 9). DON is the most common mycotoxin found in oats with annual mean levels of 26-2149 µg/kg (Table 9). Data from the last decade show a strong increase in the median concentration of DON in oats (Table 9). An increased median concentration of DON can also be seen in wheat during this period (Table 8). During the same period *Fusarium graminearum*, one of the most important producers of DON, has been detected at a much higher level than previously (Hofgaard et al. unpublished; Hofgaard et al. 2010b; Bernhoft et al. 2010). T-2 and HT-2 toxins are also found relatively frequently in crude oats, and the annual mean levels have been varying from 19 - 50 µg/kg for T-2 toxin and 43-250 µg/kg for HT-2 toxin (Table 9). As opposed to DON, there are no clear trends in occurrence of the T-2 and HT-2 toxins in oats during the last decade (Table 9). This is also true for wheat (Table 8). The highest levels of zearalenone in Norwegian grain are found in oats, with an annual mean concentration in crude oats of 1-179 µg/kg. Zearalenone has, however, not systematically been included in the monitoring program for crude grain. The levels of moniliformin, enniatins and beauvericin are only analysed in Norwegian oats harvested 2000-2002. The toxins were frequently detected but the mean levels in oats were considerably lower than in wheat during these years (Table 9).

Table 9 Mycotoxins in oat grains in Norway. Mean middle bound (µg/kg) with sample numbers/ number of samples below limit of detection in parenthesis, are given. All data from the data base at the Norwegian Veterinary institute.

Year	DON	T-2	HT-2	NIV	OCH	ZON	MON	ENN	BEA
1990	111 (20/0)	n.a.	n.a.	30 (20/20)	0.4 (20/17)	2.5 (20/19)	n.a.	n.a.	n.a.
1991	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
1992	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
1993	470 (3/0)	n.a.	n.a.	40 (3/2)	0.2 (3/2)	n.a.	n.a.	n.a.	n.a.
1994	538 (3/0)	n.a.	n.a.	25 (3/2)	3.5 (3/2)	2.5 (2/2)	n.a.	n.a.	n.a.
1995	61 (25/8)	n.a.	n.a.	25 (25/25)	0.3 (20/19)	4.6 (20/19)	n.a.	n.a.	n.a.
1996	104 (14/5)	50 (14/7)	142 (14/0)	10 (14/14)	0.2 (14/14)	10 (14/12)	n.a.	n.a.	n.a.
1997	61 (11/0)	50 (11/5)	194 (11/0)	14 (11/8)	0.1 (11/4)	3.0 (11/8)	n.a.	n.a.	n.a.
1998	134 (22/4)	20 (22/12)	66 (22/3)	10 (22/22)	0.1 (22/14)	1.0 (22/22)	n.a.	n.a.	n.a.
1999	55 (82/u)	19 (82/u)	53 (82/u)	12 (82/u)	n.a.	n.a.	n.a.	n.a.	n.a.
2000	26 (61/u)	19 (61/u)	43 (61/u)	14 (61/u)	n.a.	n.a.	20 (21/u)	19 (21/u)	1.5 (21/u)
2001	39 (45/u)	21 (45/u)	46 (45/u)	8.2 (45/u)	n.a.	n.a.	20 (26/u)	49 (26/u)	7.8 (26/u)
2002	54 (77/u)	40 (77/u)	102 (77/u)	33 (77/u)	n.a.	n.a.	73 (26/u)	65 (26/u)	19 (26/u)
2003	245 (112/u)	33 (112/u)	103 (112/u)	50 (112/u)	n.a.	n.a.	n.a.	n.a.	n.a.
2004	842 (136/u)	30 (136/u)	83 (136/u)	29 (136/u)	n.a.	n.a.	n.a.	n.a.	n.a.
2005	1623 (355/u)	45 (355/u)	191 (355/u)	21 (355/u)	0.2 (17/8)	3.3 (6/5)	n.a.	n.a.	n.a.
2006	878 (245/u)	46 (245/u)	250 (245/u)	25 (245/u)	0.2 (15/9)	17 (10/2)	n.a.	n.a.	n.a.
2007	858 (138/u)	43 (138/u)	151 (138/u)	22 (138/u)	0.1 (17/10)	159 (6/1)	n.a.	n.a.	n.a.
2008	1668 (77/u)	25 (77/u)	89 (77/u)	24 (77/u)	0.1 (17/15)	84 (14/3)	n.a.	n.a.	n.a.
2009	2149 (30/u)	28 (30/u)	106 (30/u)	29 (30/u)	0.8 (12/9)	179 (10/0)	n.a.	n.a.	n.a.
2010	1877 (24/0)	43 (24/8)	153 (24/2)	<30 (24/17)	n.a	n.a	n.a	n.a	n.a
2011	1755 (28/0)	28 (27/16/)	92 (28/6)	15 (28/28)	n.a	n. a	n.a	n.a	n.a

DON: deoxynivalenol, **NIV**: nivalenol, **OCH**: ochratoxin, **ZON**: zearalenone, **MON**: moniliformin, **ENN**: enniatins, **BEA**: beauvericin,; **n.a**.: not analyzed, **u:** unknown number of samples below LOD. Samples below LOD are considered with a value of ½ LOD for the calculation of the mean values. Oat grain has not been analysed for aflatoxins or fumonisin.

6.1.3 Barley

Samples of barley have been analysed for mycotoxin content for several years as a part of the feed surveillance programme. The levels of particularly DON are considerably lower than for wheat and oats (Table 10). The levels of T-2 and HT-2 are usually low but far more frequently found in barley than in wheat. The levels of moniliformin, enniatins and beauvericin are only analysed in Norwegian barley harvested 2000-2002. The levels were lower than for wheat, but seem to be higher than for oats. The levels of zearalenone were low as in wheat and oats. About 50% of the samples were below the limit of detection.

6.1.4 Spelt, rye and maize

Rye is less important in both feed and food in Norway. A very limited number of samples have therefore been collected for mycotoxin determination. The levels of the analysed mycotoxins in rye are considerably lower than in wheat and oats (Table 10). The production of maize is very limited, but maize is imported, mostly for for use in feed. A few samples have occasionally been analysed for aflatoxins and fumonisins.

Table 10 Mycotoxins in barley, spelt, rye, and maize grains and flours in Norway. Mean middle bound bound concentrations (µg/kg) with sample numbers/ number of samples below limit of detection in parenthesis, are given. Combined results for 1990-2011. All data are from the data base at the Norwegian Veterinary Institute.

Species	DON	T-2	HT-2	NIV	OTA	ZON	MON	FUM	ENN	BEA	AFL
Barley	37 (805/u)	15 (785/u)	14 (785/u)	16 (805/u)	1.01 (82/55)	7.2 (62/28)	40 (75/u)	n.a.	363 (75/u)	1.5 (75/u)	n.a.
Spelt	27 (11/4)	7.7 (11/11)	7.2 (11/11)	11 (11/11)	n.a.	1.2 (7)	n.a.	n.a.	n.a.	n.a.	n.a.
Rye	23 (64/32)	14 (4/4)	10 (4/4)	23 (64/64)	0.3 (36/30)	3.5 (28/25)	n.a.	n.a.	n.a.	n.a.	n.a.
Maize	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	992 (55/6)	n.a.	n.a.	0.6 (127/65)

DON: deoxynivalenol, **NIV**: nivalenol, **OTA**: ochratoxin A, **ZON**: zearalenone, **MON**: moniliformin, **FUM**: fumonisin, **ENN**: enniatins, **BEA**: beauvericin, **AFL**: aflatoxin; **n.a.:** not analysed, **u:** unknown number of samples below LOD, but it is known that half LOD is used in the calculation of mean.
Samples below LOD are considered with a value of ½ LOD for the calculation of the mean values.

The Norwegian Veterinary Insitute has conducted for Norwegian Food Safety Authority a pilot study of ergot and ergot alkaloids (semi-quantitiative metod) in randomly collected samples of rye, barley and wheat of in sum 36 samples in 2012. The results (unpublished) show that rye was most exposed but fungal material and toxins were also found in several of the barley and wheat samples. The highest ergot alkaloid concentration was 1.6 mg/kg rye.

6.2 Occurrence in feed for terrestrial animals

The majority of available data on trichothecene concentrations in cereals are from crude cereal grain. Wheat, barley and oats have been collected yearly during the last one and half decades. Most samples have been collected at cereal grain receivers or mills by Norwegian Food Safety Authority and sent to the Norwegian Veterinary Institute for chemical analysis. Most of the samples have been analysed at the Norwegian Veterinary Institute, and in addition samples have analysed at other accreditated laboratories and the results reported to the Norwegian Veterinary Institute. The results of trichothecenes (DON, T-2 and HT-2) in wheat

(N=548), barley (N=807) and oats (N=1434) are presented in Table 11. The whole dataset of results during the last 15 years are included without weighting the results by year.

DON, HT-2 and T-2 are known to constitute a particular potential animal health problem with their considerable occurrence in cereal grain surveyed during many years in Norway. Analysis results of the mycotoxins in compound feed for most animal species are sparse or available primarily from the last couple of years. Thus, a quantitative estimation of the occurrence of these toxins in animal compound feed is made on the basis of their occurrence in the grain species combined with the knowledge on the content of these grain species in the compound feed.

The mycotoxin concentrations in the cereal grain species show great variation, and are considerably higher in oats than in barley and wheat. Thus, the mycotoxin levels in prepared compound feed will often be related to the content of oats in the recipes.

Table 11: Mean and median DON and HT-2+T-2 concentration and variation (µg/kg) in cereal grain samples from the Norwegian Veterinary Institute during the 15-year periode 1997-2011. The values are based on the same data set as presented yearly in Tables 8-10.

	Wheat (n=548)		Barley (n=807)		Oats (n=1434)	
	DON	**HT-2+T-2**	**DON**	**HT-2+T-2**	**DON**	**HT-2+T-2**
Mean	126	25	73	36	915	181
SD	265	1	186	33	2627	352
Min	10	22	10	25	10	25
Max	2282	42	2032	609	36628	7802
SEM	11	0	7	1	69	9
50 percentile (median)	23	25	10	25	93	93
95 pct	586	25	323	72	3983	564
99 pct	1488	25	1052	177	12487	1356

These data were used as the basis for modelling estimates of the concentrations of the corresponding mycotoxins in typical recipes of feed for pigs, poultries, ruminants, horses and dogs. The composition of cereal grain in different feed recipes may vary considerably for different species and there may be different recipes for feed for the same species as well. Typical examples are given in Table 12. These examples are based on information from the main feed producers in Norway (Felleskjøpet and Norgesfôr).

Table 12: Animal feed content of wheat, barley and oats (% of dry matter), typical recipes of compound feed.

	Wheat	Barley	Oats
Piglet recipe A	45 %	16 %	13 %
Piglet recipe B	35 %	37 %	8 %
Growth pigs recipe A	40 %	38 %	0 %
Growth pigs recipe B	0 %	59 %	10 %
Sow recipe A	50 %	26 %	4 %
Sow recipe B	42 %	37 %	5 %
Broiler chicken	40 %	0 %	10 %
Laying hens	40 %	0 %	30 %
Ruminant	25 %	30 %	10 %
Horse	10 %	30 %	30 %
Dog	35 %	0 %	0 %

The expected mycotoxin concentrations in the typical feed recipes based on these estimations are presented in Table 13. Monte Carlo simulation was used to include the variability of toxin concentration in different batches of grain. The mycotoxin concentration observed in random samples of wheat, barley and oats, was used in each of 10000 iterations, and multiplied by their respective fraction. For pigs, poultry and dogs receiving these feed formulations as their complete diets, the concentrations of DON and T-2/HT-2 are estimates of total exposure. For ruminants and horses, receiving maximum 50% compound feed, the figures in total ration constitute a corresponding ratio – maximum the half of the figures in Table 13.

There are several factors influencing these estimates. The two first are supposed to increase the trichothecene concentrations: Wheat bran, which may contain elevated levels of the mycotoxins compared with the crude wheat, may be used to a certain degree in ruminant and pig compound feed and increase the mycotoxin concentration in the feed. The second factor is that the compound feed recipes may contain other ingredients which also may be a source of mycotoxins from field fungi. In practice, this refers to maize. Maize is normally not used in feed for pigs but some recipes include up to 5% maize. Broiler chicken feed usually contains 10-20% maize, and laying hen feed may contain 0-20% maize. Ruminant compound feed recipes may vary from 0-20% maize. Maize is hardly used in compound feed for horses. In dog feed, 10-20% maize grits is used. The concentration of trichothecenes in maize imported as feed ingredient is not known. However, based on results of international mycotoxin surveys on maize, use of maize in compound feed may most likely rise the trichothecene concentration.

Two other factors (actions) may contribute to reduce the trichothecene concentrations of compound feed: Some of the oats may be dehusked which reduce the trichothecene content to a certain extent. Dehusking has primarily been practiced for piglet feed but also to some extent for feed for other pigs and poultry in Norway. The other important action is the recently adopted quick analysis for DON at the mills to sort out batches with high levels of DON. This tool now increase the possibilities to avoid particularly DON contaminated feed to susceptible species as pig, and may contribute to reduce the risk related to DON in animal feed.

Table 13. Estimated DON and HT-2+T-2 concentrations (µg/kg) in animal feed based on mycotoxin concentrations measured in cereal grains and typical content of cereals (wheat, barley and oats)*.

	Mean		SD		Median		95th percentile		99th percentile	
	DON	HT2+ T2	DON	HT2+ T2	DON	HT2+ T2	DON	HT2+ T2	DON	HT2+ T2
Piglet feed A	182	40	346	46	70	29	671	91	1694	218
Piglet feed B	140	36	230	30	63	29	500	72	1128	145
Pig growth feed type A	75	23	124	12	29	20	307	37	649	71
Pig growth feed type B	132	39	273	40	38	29	548	88	1375	181
Sow feed type A	114	29	169	16	55	25	415	49	937	87
Sow feed type B	122	33	178	21	60	27	462	59	886	109
Broiler chicken feed	138	28	270	35	48	20	533	66	1294	166
Laying hen feed	318	65	756	105	82	39	1253	177	3773	479
Ruminant feed	141	35	264	36	58	26	508	76	1299	173
Horse feed	304	67	752	106	71	42	1219	183	3773	479
Dog feed	42	9	89	0	8	9	199	9	494	9

*For pigs, poultry and dogs these figures represent the total ration. For ruminants and horses, receiving maximum 50% concentrate, the figures in total ration is maximum half of the numbers given in this table.

DON and HT-2+T-2 have also been analysed in a number of samples of prepared compound feed, as shown in Table 14. The samples analysed at the Norwegian Veterinary Institute were obtained between 2004 and 2011. The other swine feed results were kindly provided by the Norwegian feed producers and are results from 2010-11. The analytical method is the same or similar, and comparable as for the cereal grain samples.

The observed levels of DON and sum of T-2 and HT-2 toxins in pig feed samples are slightly higher than those estimated from levels in crude grains and based on typical feed recipes, which could be explained by several variability factors as described above. These include recipes that contain bran or other ingredients as maize which also may be a source of mycotoxins from field fungi. Furthermore, most of the samples of pig feed were collected from the last seasons with considerably higher DON levels than the average level surveyed during several years. The other factors which may contribute to reduce trichothecene concentrations, dehusking of oats, has been done at a relatively low level except for piglet feed. The effect of newly established use of quick analyses at the mills has not yet been documented in the present data. The concentrations estimated from cereal grain concentrations and recipes compared with analysed concentrations in compound feed for pigs remain however within the same order of magnitude. According to the estimated results , pigs receiving 100% of compound feed would be exposed to a mean DON content of 75-182 µg/kg feed (95-percentile 307-671 µg/kg feed), while the observed mean DON concentration in pig feed samples was 435 µg/kg feed (95-percentile 1056 µg/kg feed). For T-2 and HT-2 the estimated and measured concentrations in pig feed are more similar.

Comparison between estimated and measured concentrations of DON and sum T-2 and HT-2 for other species than pigs may not be appropriate as the number of analysis results from compound feed is low.

Table 14. Concentrations of DON and HT-2+T-2 analysed in compound feed for pig, poultry, horse, dog and ruminants*. All pig feed contain samples analysed by the feed industry in Norway (2010-11) in addition to the samples analysed at the Norwegian Veterinary Institute (2004-11).

DON	n	Mean	SD	Median	95th pct	99th pct
All pig feed	309	435	313	370	1056	1459
NVI, pig feed	131	321	279	244	920	1081
NVI, poultry feed	53	360	370	218	1131	1580
NVI, horse feed	16	377	211	371	679	693
NVI, dog feed	28	408	274	364	824	1174
NVI, ruminant feed	3	1207	1709	355	2893	3119
HT2+T2	**n**	**Mean**	**SD**	**Median**	**95th pct**	**99th pct**
All pig feed	175	48	43	35	109	259
NVI, pig feed	131	53	49	36	114	286
NVI, poultry feed	53	80	77	58	230	367
NVI, horse feed	16	43	48	25	103	195
NVI, dog feed	28	29	11	25	54	67
NVI, ruminant feed	3	62	64	25	125	134

*For pigs, poultry and dogs these figures represent the total ration. For ruminants and horses, receiving maximum 50% concentrate, the figures in total ration is maximum half of the numbers given in this table.

From the estimated values, feed for laying hens contain high levels of DON and HT-2+T-2, which are due to the use of considerable amounts of oats. Also horse compound feed may contain elevated concentrations of mycotoxins due to use of oats in the recipes. Horses may also receive crude oats, which may lead to a significant exposure to mycotoxins – first of all trichothecenes. The discussion of the significance of the result on occurrence of DON and HT-2 + T-2 in animal feed is in risk characterisation after the comprehensive characterisation of the hazards. In addition to the large data base of trichothecenes in cereal grain sampled during several years, there are also some available data on concentrations of zearalenone, ochratoxin A, moniliformin, enniatins and beauvericin in cereal grain. The mean results are presented in Tables 8-10 and the complete results in Appendix B .

6.2.1 A summary of occurrence relevant for animal feed

The yearly mean zearalenone concentrations measured during years 2005-09 in barley and oats varied from 1.5-31 and 3.3-179 µg/kg, respectively, with yearly maximum concentrations 3.0-196, and 11.0-855 µg/kg, respectively. Zearalenone has also been measured in wheat (imported and Norwegian) during 1990-1998 with yearly means 1.3-10.0 µg/kg and no difference between imported and Norwegian wheat.

The yearly mean ochratoxin A concentrations measured during years 2005-09 in barley and oats varied from 0.14-4.5 and 0.07-0.21 µg/kg, respectively, with yearly maximum concentrations 0.8-40.0, and 0.5-2.1 µg/kg, respectively. Ochratoxin has also been measured in wheat (imported and Norwegian) during 1990-1998 with yearly means 0.1-0.9 µg/kg and no difference between imported and Norwegian wheat.

Of moniliformin measured in wheat, barley and oats during 2000-2002 the yearly means were <40-210, <40-48 and <40-73 µg/kg, respectively, with yearly maximum concentrations 87-950, 43-380 and 70-210 µg/kg, respectively.

Enniatins were measured in wheat, barley and oats during 2000-2002 and the yearly medians were 126-730, 153-493 and 19-65 µg/kg, respectively, with yearly maximum concentrations 1590-7400, 1213-5100 and 223-440 µg/kg, respectively.

Beauvericin was measured in wheat, barley and oats during 2000-2002 and the yearly medians were all low; <3.0 µg/kg in wheat and barley, and <3.0-19 µg/kg in oats.

Maize is as mentioned above, commonly used in compound feed. Maize may be a source for several mycotoxins. The knowledge on mycotoxins in maize used in Norway is very sparse. From the literature maize is known to be vulnerable for contamination with trichothecenes, zearalenone, fumonisins, ochratoxin as well as aflatoxins. There is no data on trichothecenes, zearalenone and ochratoxin in Norwegian maize samples. Fumonisins have been measured in some maize samples, and aflatoxins more extensively. Fumonisins (B1+B2) were measured in 2-5 samples per year of maize grain during 2005-2007 and the yearly mean values were 1237-2342 µg/kg and yearly maximum concentrations were 2634-6990 µg/kg. In addition, a few samples of maize gluten were also analysed for fumonisins with a similar level. Fumonisins were also measured in 28 samples of compound feed for dogs collected at the Norwegian market in 2010. Fumonisin B1 and B2 were detected in most samples with maximum sum of B1 and B2 at 237µg/kg.

Aflatoxins (B1+B2) were measured in 4-20 samples per year of maize grain during 2005-2009 and the yearly means were 0.2-0.7 µg/kg and corresponding maximum concentrations were 0.4-4.3 µg/kg. Samples of maize gluten (5-20 per year) showed a similar level of aflatoxins (means 0.5-1.5 µg/kg and maximum 1.0-4.8 µg/kg). In addition, Norwegian Food Safety Authority provides each year results on aflatoxins (B1, B2, G1, G2) in ruminant compound feed. The levels use to be below detection limits (0.84 µg/kg for aflatoxins B1 and G1, 0.24 µg/kg for B2 and G2). Aflatoxins were also measured in 28 samples of compound feed for dogs collected at the Norwegian market in 2010. Aflatoxins was only detected as detectable traces in some samples.

6.2.2 Occurrence of mycotoxins in straw

Straw are used as fodder and beddings for various animal species. Similarly as cereal grain also the straw may contain mycotoxins but levels seem to be lower than in the cereal grain. Norwegian data on this field are scarce.

6.2.3 Occurrence of mycotoxins in brewery cereal waste

Cereal waste from beer brewery is used as animal feed. This is delivered warm and humid and is certainly a substrate for mould production. It has to be used without particular storage to avoid moulds and mycotoxin production. However, the mycotoxin content of the cereal grain product itself is probably very satisfactory as it is has been used for beer production. Furthermore, elevated mycotoxin level in this cereal are known to cause production problems in form of foaming and therefore avoided

6.3 Occurrence in aquafeed materials

The main farmed finfish species in Norway are the salmonids Atlantic salmon (*Salmo salar*) and rainbow trout (*Oncorhynchus mykiss*) with a production of 859,056 and 76,008 tons round weight in 2009, respectively. These species account for ~90% and ~8% of the total Norwegian finfish aquaculture, respectively. Other minor cultured species include Atlantic cod (*Gadus morhua*), Atlantic halibut (*Hippoglossus hippoglossus*), and Arctic charr (*Salvelinus alpinus*). In 2011, a total of 1,426,864 tons of fish feed was produced at Norwegian commercial fish feed production sites (Directorate of Fisheries 2011).

Intensive Atlantic salmon farming is a relatively new sector compared to farming of land animals, and fish feed formulations are under constant development. Pressure on wild fish populations stocks and hence limited access to fish meal and fish oil for the rapidly growing aquaculture industry has led to the development of aquafeeds that rely less on fish meal and fish oil (Torstensen et al. 2008; Tacon and Metian 2008).

Norwegian produced salmonid feeds do not include Norwegian grown cereals as feed ingredients, the vegetable feed ingredients used are all imported (pers. communication, EWOS, Biomar and Skretting). A random selection of the main vegetable feed ingredients used at nine Norwegian commercial fish feed production sites as well as two large scale experimental sites were analysed by the Norwegian Veterinary Institute on behalf of the Norwegian Food Safety Authority. Table 15 gives the levels of mycotoxins in the main vegetable feed ingredients used in Norwegian produced salmonid feeds. The main cereals used in salmon feeds are maize and wheat, while soybean products are the main oilseed feed ingredient followed by rapeseed and sunflower. For wheat and maize feed ingredients, ochratoxin A, zearalenone, and DON were the dominant mycotoxines present. Soybean products also had high incidences of ochratoxin A contamination, while zearalenone contamination was low and DON contamination absent. The ochratoxin A, zearalenone and DON contamination in rapeseed and sunflowers was low or non-existing. Typical for maize products was the high occurrence of the fumonisins (FB1, B2 and B3) and aflatoxin B1. Soybean products where the only other feed ingredients were fumonisins were observed, although to a lesser extent than in maize. The mycotoxin HT-2 was found in wheat and maize, but not in the other products. The mycotoxins nivalenol and T-2 were not observed in any of the vegetable feed ingredients.

In addition to plant feed ingredients, marine feed ingredients such as ensilage (3 samples) and especially fish meal (49 samples) were analysed for ochratoxin A, which is a typical storage mycotoxin; of the fish meal only 6 out of the 49 of the analysed samples had values exceeding the LOD, with mean levels of 0.05 µg/kg.

Table 15. Mean values (µg/kg) and sample numbers () of feed ingredients used in Norwegian produced aquafeeds 2005-2009. Data source: Norwegian Food Safety Authorities monitoring programme for aquafeeds.

Feed ingredient	DON	T-2	HT-2	NIV	OTA	ZON	MON	FUM	ENN	BEA	AFL
Ensilage	n.a.	n.a.	n.a.	n.a.	0.05 (3)	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
Fish meal	n.a.	n.a.	n.a.	n.a.	0.05 (49)	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
Horsebeans	10 (2)	15 (2)	10 (2)	15 (2)	0.03 (2)	1 (2)	n.a.	5 (2)	n.a.	n.a.	0.38 (2)
Wheat products	94 (28)	15 (27)	12 (28)	16 (28)	0.3 (28)	5.4 (27)	n.a.	15 (6)	n.a.	n.a.	0.37 (6)
Maize and maize gluten	72 (16)	15 (16)	13 (16)	21 (16)	1.3 (16)	246 (16)	n.a.	1662 (13)	n.a.	n.a.	1.6 (16)
Rapeseed products	19 (6)	15 (6)	12 (6)	18 (6)	0.04 (6)	1.3 (6)	n.a.	6.7 (6)	n.a.	n.a.	0.39 (6)
Sunflower products	12 (7)	15 (7)	15 (7)	15 (7)	3.9 (7)	13 (7)	n.a.	10 (2)	n.a.	n.a.	0.40 (2)
Soya products	10 (17)	15 (17)	11 (17)	18 (17)	0.06 (21)	2.7 (17)	n.a.	22 (30)	n.a.	n.a.	0.32 (30)

DON: deoxynivalenol, **NIV**: nivalenol, **OTA**: ochratoxin A, **ZON**: zearalenone, **MON**: moniliformin, **FUM**: fumonisin, **ENN**: enniatins, **BEA**: beauvericin, **AFL**: aflatoxin
Samples below LOD are considered with a value of ½ LOD for the calculation of the mean values.

For aquafeeds data are only available for protein feed ingredients. No data are available on final aquafeeds or oil feed ingredients. Also, there is a lack of data at the fish farm level. In absence of feed data, a pure theoretical estimate is made on the levels that could be expected in the feeds based on the levels found in the protein feed ingredients. Data on mycotoxin levels in aquafeed ingredients (Table 15) were used as the basis for modelling estimates of the concentrations of the corresponding mycotoxins in typical recipes of feed for farmed Atlantic salmon, similar to the modelling of feed for pigs, poultries, ruminants, horses and dogs (see section 6.2). The current development of aquafeeds, the great variability in feed ingredients, and limited detailed information on feed formulation for the major farmed fish species in Norway (Atlantic salmon) precludes an assessment of actual exposure levels based on the occurrence of mycotoxins in individual feed ingredients. However, an example of a commercially relevant grower salmon feed composition in 2010 had approximately 18% wheat products, 15% soy products and 7% maize products (Berntssen et al. 2010). Based on the levels in feed ingredients (Table 15) and inclusion level of wheat, soy and maize products in a commercial feed composition for grower feed (adult > 1kg salmon) (Berntssen et al. 2010), a hypothetical exposure scenario can be made. Table 16 gives the possible levels of mycotoxins in commercial salmon feeds.

Table 16. Hypothetical mycotoxin levels (µg/kg) in commercial salmon feed with approximately 18% wheat products, 15% soyabean products and 7% maize products. Only for ochratoxin A (OTA) fishmeal (25%) was included as a mycotoxin source. The assumption is made that these feed ingredients are the sources for mycotoxins. Values are given as median, mean and maximum (max) estimated levels, and are based on the levels reported for feed ingredients in Tables 11 and 15.

In feed	DON	HT-2	T-2	NIV	OTA	ZON	FUM	AFL
Median	14	3.7	5.6	5.9	0.042	7.9	127	0.17
Mean	23	4.3	5.5	6.6	0.17	19	135	0.23
Max	128	10	5.6	10	1.2	104	407	0.59

DON: deoxynivalenol, **NIV**: nivalenol, **OTA**: ochratoxin A, **ZON**: zearalenone, **FUM**: fumonisin, **AFL**: aflatoxin

6.4 Occurrence in food

In 2008-2011 selected *Fusarium* toxins have been analysed in commercially available wheat and oat products for human consumption, such as flour, bran, groat, and oat flakes. The samples of milling products were sampled on the mills during the production process. Each sample was collected during one day's production and was collected to represent the flour from one day's production. The sample size was dependent on the total production on the day of sampling and could be up to 10 kg. The samples were well mixed before subsamples were taken out for analysis.

Furthermore, infant porridges and breakfast cereals were analysed for trichothecenes and zearalenone and maize products were analysed for fumonisin. The infant porriges were sampled in the shops.

DON was present above the limit of detection in practically all wheat samples (Table 17). As observed for crude grain, the mean values in wheat products vary from year to year, from 152 µg/kg in 2010 to 224 µg/kg in 2011. The levels in processed wheat products are in the same range as in crude wheat, indicating that there is no systematic reduction in DON levels during processing. The samples from crude grain and processed products were, however, not from the same batches, making it impossible to draw definite conclusions. Comparable to crude grains, the combined levels of T-2 and HT-2 are considerable lower than the DON level in wheat flour and bran, and were detectable only in a small number of samples. The same applied to nivalenol, which had a low incidence of samples above the LOD and low mean concentrations. Zearalenone is only present in very low concentrations in processed wheat with mean levels from 2.3–6.6 µg/kg. Other *Fusarium* toxins (moniliformin, enniatins, beauvericin) as well as ochratoxin A and aflatoxins have not been analysed in samples of processed wheat products.

Table 17. Mycotoxins in wheat flours on the market in Norway. Mean middle bound concentrations (µg/kg) and sample numbers / number of samples below the LOD in parenthesis. Data are from the Norwegian Food Safety Authoritys monitoring programme for mycotoxins in food, analysed at the Norwegian Veterinary Institute.

Year	DON	T-2	HT-2	NIV	ZON
2008	213 (137/5)	3.1 (137/137)	2.9 (137/137)	3.9 (137/126)	3.0 (137/93)
2009	161 (113/7)	8.5 (113/101)	7.0 (113/111)	15 (113/111)	3.6 (113/63)
2010	152 (120/1)	15 (120/106)	11 (120/109)	16 (111/101)	3.0 (120/83)
2011	224 (90/1)	19 (90/90)	13 (90/90)	19 (72/72)	n.a.

DON: deoxynivalenol, **NIV:** nivalenol, **ZON**: zearalenone. **T-2, HT-2:** trichothecenes, **n.a**.: not analysed. It was not analysed for moniliformin, fumonisin, enniatins, beauvericin, ochratoxin, or aflatoxin. Samples below LOD are considered with a value of ½ LOD for the calculation of the mean values.

DON concentrations were higher in oat flakes than in wheat products, and annual mean concentrations in oat flakes varied from 160 µg/kg in 2011 to 327 µg/kg in 2009. The concentrations of T-2 and HT-2 were also higher in oat flakes than in wheat (Table 17 and 18). The concentrations in oat flakes were, however, considerably lower than the levels found in crude oats , and the total incidence of samples above LOD was very low (Tables 9 and 18). Nivalenol had likewise a low incidence of samples above the LOD, and the levels were low. Zearalenone occurred at slightly higher concentrations in oat flakes than in processed wheat products, but the levels were still low with mean values ranging from 1.4 µg/kg in 2011 to 6.3 µg/kg in 2009 (Table 18).

Other Fusarium toxins (moniliformin, enniatins, and beauvericin) as well as ochratoxin A and aflatoxins had not been analysed in oat flakes.

Table 18. Mycotoxins in oat flakes in Norway. Mean middle bound concentrations (µg/kg) and sample numbers / number of samples below the LOD in parenthesis. Data are from the Norwegian Food Safety Authoritys monitoring programme for mycotoxins in food, analysed at the Norwegian Veterinary Institute.

Year	DON	T-2	HT-2	NIV	OCH	ZON
2008	285 (31/0)	9.7 (31/5)	36 (31/1)	3.2 (31/29)	n.a.	1.9 (31/26)
2009	327 (34/0)	8.0 (34/23)	19 (34/11)	13 (34/34)	n.a.	6.3 (34/21)
2010	168 (31/0)	12 (31/31)	9.4 (31/29)	12 (30/30)	n.a.	1.7 (31/27)
2011	160 (31/1)	15 (31/31)	19 (31/16)	15 (28/28)	0.008 (5/5)	n.a.

DON: deoxynivalenol, **NIV:** nivalenol, **OCH**: ochratoxin, **ZON**: zearalenone. **T-2, HT-2:** trichothecenes, **n.a**.: not analysed. It was not analysed for moniliformin, fumonisin, enniatins, beauvericin, or aflatoxin. Samples below LOD are considered with a value of ½ LOD for the calculation of the mean values.

Trichothecene mycotoxins as well as zearalenone and ochratoxin A were analysed in a number of infant porridges containing different grains in 1999, 2000, and 2008. DON was detected in more than 50% of the wheat- and oat-based porridges although in low concentrations (Table 19). Furthermore, DON was detected in alarmingly high concentrations in a batch maize-based infant porridges in 1999, which was immediately removed from the

market. Analyses in 2000 and 2008 revealed only rather low DON-levels, so that VKM decided to consider the 1999-value as an outlier and not to include maize-based infant porridge in the present risk assessment. The combined results for all years (Table 19) still look high as they are dominated by the 1999-value. T-2, HT-2, and nivalenol were not detected in the infant porridges, whereas ochratoxin and zearalenone were found in wheat-, oat-, and maize-based samples, but only at low concentrations.

Breakfast cereals were analysed in 2010 for trichothecenes and zearalenone (Table 19). DON was detected in 70% of the samples at noticeable concentrations. T-2, HT-2, and nivalenol were not detected in breakfast cereals, whereas relatively low zearalenone-levels were found in about 40% of the samples.

Several retailed maize products such as corn cobs, tinned corn, and tinned kernel were analysed for fumonisins (Table 19), which was not detected in any of the samples.

Table 19. Mycotoxin prevalence in infant porridge, breakfast cereals, and maize food products in Norway mean middle bound values (µg/kg) and sample numbers/number of samples below the LOD in parenthesis. Combined results for 1990-2010.

Food category	DON	T2	HT-2	NIV	OTA	ZON	FUM
Infant porridge wheat	13[a] (4/1)	4.4[a] (4/4)	4.4[a] (4/4)	4.4[a] (4/4)	0.2[b] (45/2)	0.8[a] (49/18)	n.a.
Infant porridge oat	25[c] (8/4)	4.0[a] (5/4)	6.2[a] (5/5)	6.3[c] (8/8)	0.1[b] (26/4)	0.8[d] (31/17)	n.a.
Infant porridge barley[a]	5[a] (1/1)	2.5[a] (1/1)	2.5[a] (1/1)	2.5[a] (1/1)	n.a.	1.3[a] (1/1)	n.a.
Infant porridge spelt	5[a] (1/1)	5.0[a] (1/1)	5.0[a] (1/1)	5.0[a] (1/1)	0.04[e] (2/0)	1.1[f] (3/2)	n.a.
Infant porridge sorghum[a]	5.0[a] (1/1)	5.0[a] (1/1)	5.0[a] (1/1)	5.0[a] (1/1)	n.a.	2.5[a] (1/1)	n.a.
Infant porridge maize	642[c] (11/0)	4.2[c] (3/3)	4.2[c] (3/3)	14.2[c] (11/7)	0.01[c] (26/20)	65[d] (28/3)	n.a.
Infant porridge mixed grains	11[a] (6/1)	2.9[a] (6/6)	2.9[a] (6/6)	2.9[a] (6/6)	0.45[e] (9/0)	1.0[f] (15/0)	n.a.
Breakfast cereals mixed grains[g]	67[g] (29/8)	13[g] (29/29)	9.3[g] (29/27)	14[g] (27/25)	n.a.	2.5[g] (29/18)	n.a.
Maize food products (corn cob, tinned corn, tinned kernel)	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	10[h] (79/79)

DON: deoxynivalenol, **NIV:** nivalenol, **OTA:** ochratoxin A, **ZON:** zearalenone, **FUM:** fumonisin, **n.a.:** not analyzed; Samples below LOD are considered with a value of ½ LOD for the calculation of the mean values. [a] Data from 2008, [b] data from 1999 and 2000, [c] data from 1999 and 2008, [d] data from 1999, 2000 and 2008, [e] data from 2000, [f] data from 2000 and 2008, [g] data from 2010, [h] data from 2000 and 2001.

6.5 Occurrence of mycotoxins and mycotoxin-producing fungi in imported cereal grain

The occurrence of mycotoxin-producing fungi in small grain cereals (e.g., wheat, barley, and oats) is similar in the Scandinavian countries, Germany, France and the Netherlands. There are several European reports of an increase in *F. graminearum* parallel to the development in Norway. The pattern of mycotoxins in the cereals may differ with geographical area. T-2 and HT-2 are more common in Norway, Sweden and England compared with southern Europe, where DON and zearalenone traditionally are of largere problems. In USA and Canada is

DON the great problem in small cereal grains. The cereal species which is most contaminated with mycotoxins do also show differences around the world.

Table 20 presents data on cereal grain (of wheat, barley, maize, rice and some other cereals) and cereal grain products imported into Norway during the years 2007-2010. The exporting countries are ranked according to the total value of cereal grain import during that period. Countries with a total sum (2007-2010) less than 30,000 tons are excluded from the table. Data on import from India, Pakistan, Thailand, Vietnam and other countries, which are mainly exporting rice, are not included. Cereal grain products include breakfast cereals, pasta, bread, pastry, cakes, biscuits and a number of other commodities. The value of the import of these products is highest from Denmark and Sweden. Table 20 shows that the four North-European countries Germany, Sweden, Denmark and Finland in addition to France have been the main sources of import of cereal grain and cereal grain products into Norway the last four years. Also, USA, Latvia, Great Britain and the Netherlands have exported rather large amounts of cereals to Norway in this period. The traditional trading partners Argentina, Canada and Russia, however, have exported lower volumes of cereals to Norway in this period than during previous years. When considering the total exposure to mycotoxins this shift in trading patterns is a change that potentially could have impacts, if this corresponds to a shift in mycotoxin contamination levels in imported cereals.

Table 20. Amount (1000 tons)) of imported cereal grains (of wheat, barley, maize, rice and some other cereals) and cereal grain products into Norway during 2007, 2008, 2009, and 2010. Source: Statistics Norway.

Country	2007	2008	2009	2010	Sum 2007-2010
Germany	145.0	199.3	249.4	285.4	879.1
Sweden	93.4	124.4	86.2	94.6	398.5
Denmark	82.0	73.0	40.8	66.0	261.7
France	50.5	84.0	36.6	63.3	234.4
Finland	84.3	41.6	3.5	15.8	145.2
USA	47.1	71.1	1.6	1.6	121.4
Latvia	36.2	11.8	28.8	24.7	101.5
Nederland	18.8	29.8	15.9	12.5	76.9
Great Britain	20.5	32.4	9.5	13.9	76.4
Kazakhstan	39.2	10.2	3.1	17.0	69.6
Italy	17.4	17.1	17.2	16.7	68.4
Lithuania	19.9	15.1	16.0	14.3	65.4
Poland	5.0	4.8	13.3	32.9	56.1
Canada	28.6	2.2	2.5	5.8	39.0
Spain	6.7	10.0	9.9	10.0	36.6
Russia	0	2.2	3.5	29.9	35.7
Belgium	9.0	8.6	9.3	8.3	35.2
Argentina	1.9	11.5	0.3	17.3	31.0

Maize for animal feed and maize products imported into Norway may originate from the Mediterranean countries or overseas. A range of fungi commonly found on small grain are also producing mycotoxins in maize. Trichothecenes and zearalenone are commonly present in maize. In addition, fumonisins are commonly detected in maize grown to maturity in Southern Europe and worldwide. Maize may also contain toxins from storage fungi, ochratoxin A and aflatoxins. In an evaluation of fumonisins in the Nordic countries Petersen and Thorup (2001) reported that in 70 samples of maize-based food on the Danish market 37% contained fumonisin B1 and 21 % contained fumonisin B2. Fumonisin was not detected

in sweet maize, maize starch or maize-based infant porridge. Fumonisin B1 was detected in half of the samples of maize flakes, maize snack and popcorn. Both fumonisin B1 and fumonisin B2 were found in 75% of the samples from maize flour, tacos and polenta. No data were presented on microbiological examination of the samples, but it is probable that the fumonisin were produced by *F.verticillioides*. The level of fumonisins in maize products on the Norwegian market is likely to be the same as in Denmark. The samples analysed in Norway (Table 19, Chapter 6.4) did not contain detectable fumonisin.

7 Animal exposure to mycotoxins

Experimental studies on the effects of mycotoxins in domestic animals most often report and focus on fixed feed concentrations, rather than doses pr kg body weight. Published reports frequently do not contain enough information to calculate the doses from the feed concentrations used in the feeding experiments and any conversions would have to be based on tabulated values, introducing additional uncertainty. Furthermore, it is easier for stakeholders to understand feed concentrations than to body weight doses. VKM therefore decided to use dietary concentrations in the risk assessment of mycotoxins in domestic animals. Animal exposure of mycotoxins in this report is hence expressed as their occurrence concentrations in the cereal feed (Chapters 6.2 and 6.3) and its ratio in the total diet.

The level of cereal grain which in the present context is the source of mycotoxins varies among the different animal species. Pigs and poultry eat cereal-based compound feed as their main diet. Ruminants and horses eat roughage and usually complementary amounts of cereal compound feed. In intensive ruminant production (high milk production and feedlots), the ratio of cereal-based feed may reach about 50% of dry matter intake. Horses in particular, may ingest considerable amounts of crude oats. Rabbits may eat considerable amounts of cereal grains. Pet animals, particularly dogs, but also cats usually receive commercial compound feed where cereal grain is a considerable ingredient. As marine protein and fat are limited resources, also farmed fish receive cereal grains in their feed (up to about 30%).

For animals in commercial production feed energy density is fairly constant and feed intake per day per unit of body weight, daily growth and/or other productions such as milk and eggs are well established, including normal variation. Conversion from concentration of mycotoxins in feed to exposure can therefore be be modeled for the various productions.

8 Human exposure to mycotoxins

8.1 Norwegian data on mycotoxin levels in food used in the exposure assessment

The mycotoxin concentrations in various grain and flour products in Norwegian mills in 2008, 2009, 2010 and 2011 have been analysed. For the human exposure assessment, VKM has chosen to use the lowest and highest mean values from the four years (2008-2011) for each of the four flour products: sieved wheat flour, milled wheat flour, wheat bran and oat flakes. This choice was made in order to take into account the annual variations in fungal infections and toxin concentrations. Since the samples represent the food available on the market, this allows for potential mixture of grains from several harvest years.

The mycotoxin concentrations in infant porridges have been retrieved from 2008. The concentrations of mycotoxins in oats-based infant porridge were at least twice the levels in infant porridges based on other grains. Oat-based infant porridge was therefore chosen for exposure calculation for all infant porridges, in the absence of any detailed information about the different porridges consumed by infants.

Mycotoxin concentrations in breakfast cereals were only available for 2010. For the intake calculations performed in this risk assessment, breakfast cereals were divided into the different grain/flour types used, and intake was estimated on the basis of the occurrence of mycotoxins in the respective grain/flour types.

The calculated mycotoxin exposure is based on the mean concentrations of DON and zearalenone (analysed from both imported and Norwegian grain) found in the four different flour categories included in the Norwegian monitoring data. It should be noted that for mycotoxin contents below the limit of detection (LOD), then half of the LOD is used for calculating the mean values of mycotoxins in the different categories. DON is present in levels above the LOD in more than 95% of the samples and the use of half of the LOD in samples below the LOD has only a minor influence on the estimated mean concentrations.

The exposures to the sum of T-2 and HT-2 toxins and nivalenol could not be estimated due to a high proportion of samples below the LOD. Scenarios of potential exposure have therefore been made for these toxins (see Chapter 8.4).

8.2 Methodological description of the national consumption surveys

In the present opinion, the calculated mycotoxin exposures from food are based on data from the national food consumption surveys in Norway; Spedkost, Småbarnskost, Ungkost and Norkost. The consumption for each relevant food and food category in the dietary surveys was multiplied by the corresponding mycotoxin concentrations given in Table 22 and summarised for each individual. Short descriptions of the consumption surveys and the different methodologies used are given below:

1-year-old infants; Spedkost 2006-2007 is based on a semi-quantitative food frequency questionnaire covering the last 14 days. In addition to predefined household units, food amounts were also estimated from photographs. The study was conducted in 2007, and a total of 1635 1-year-old children participated (Øverby et al. 2009).

2-year-old children; Småbarnskost 2007 is based on a semi-quantitative food frequency questionnaire covering the last 14 days. In addition to predefined household units, food

amounts were also estimated from photographs. The study was conducted in 2007, and a total of 1674 2-year-olds participated (Kristiansen et al. 2009).

4-year-old children; Ungkost 2000 is based on a 4 consequetive days food intake registration with a precoded food diary. Food amounts were presented in predefined household units or as portions estimated from photographs (Pollestad et al. 2002). The study was conducted in 2001, and 391 4-year-olds participated.

9- and 13-year-old children/adolescents; Ungkost 2000 is based on a 4 consequetive days food intake registration with a precoded food diary. Food amounts were presented in predefined household units or as portions estimated from photographs (Øverby and Andersen 2002). The study was conducted in 2000, and 810 9-year-old children and 1005 13-year-old adolescents participated.

Adults; Norkost 3 is based on two 24-hour recalls by telephone at least one month apart. Food amounts were presented in household measures or estimated from photographs (Totland et al. 2012). The study was conducted in 2010/2011 and 1787 men and women aged 18-70 years participated.

Daily consumption of flour was computed by using food databases in the software system (KBS) developed at the Institute of Basic Medical Sciences, Department of Nutrition, at the University of Oslo. The food databases are mainly based on various versions of the official Norwegian food composition table (Matvaretabellen 2012, 2006, 1995; Rimestad et al. 2000). The three dietary surveys used in this risk assessment were conducted at three different time points, Ungkost-2000 in 2000-2001, Sped- and Småbarnskost in 2007 and the data for Norkost 3 were collected in 2010-2011. The mycotoxin analyses used in the exposure assessment were conducted in the time period 2008-2011.

8.2.1 Body weight

The individual body weights reported in the different dietary surveys were used to calculate the exposure in microgram (µg) mycotoxin/kg bw. In cases where an individual body weight is missing, the mean body weight for the age group in question has been used.

Among the 4-year-old children, body weight data for 23.8% (i.e. 93 individuals) were not reported and thus the group's mean body weight of 18 kg was used. Correspondingly, 14.3% of the 9-year-olds were allocated the group mean body weight of 32 kg, and 14.4% of the 13-year olds were allocated the group mean body weight of 49.4 kg.

Among adults, 30 (1.7%) persons did not report their individual body weights and the group's mean body weight of 77.5 kg was used.

An overview of the mean body weights for the different age groups is given in Table 21.

Table 21. Mean body weight of infants, children, adolescents and adults in the Norwegian population.

Age	Mean body weight (kg)
12 months	9.9
24 months	12.8
4 years	18.0
9 years	32.0
13 years	49.5
Adults	77.5

8.3 Chronic dietary exposure to mycotoxins

8.3.1 Overview of the mycotoxin levels in the food categories included in the exposure calculations

An overview of the mycotoxin concentrations used in the exposure calculations is presented in Table 22. In this risk assessment mycotoxin concentrations in four different flour products and in oat-based infant porridge are used. The highest and lowest mycotoxin concentrations during the four years 2008-2011 for each flour type are presented to illustrate the variations between years. The data for infant porridge are from 2008 only.

Table 22. Annual lowest and highest mean concentrations of deoxynivalenol (DON) and zearalenone for four years in the different flour categories included in the exposure calculations. Data are based on the same data as Tables 17 and 18.

Mycotoxin	Flour category	Lowest mean concentration			Highest mean concentration		
		Year[a]	n	concentration[a] (µg/kg)	Year[b]	n	concentration[b] (µg/kg)
DON	Sieved wheat flour	2010	49	140	2008	65	210
	Milled wheat flour	2010	42	121	2011	42	240
	Wheat bran	2009	20	141	2008	23	383
	Oat flakes	2011	30	165	2009	34	327
	Infant porridge[c]	2008	21	34	2008	21	34
Zearalenone	Sieved wheat flour	2008	65	1.5	2009	49	2.3
	Milled wheat flour	2008	52	2.6	2009	42	3.9
	Wheat bran	2010	20	5.5	2008	23	7.7
	Oat flakes	2010	34	1.7	2009	34	6.3
	Infant porridge[c]	2008	21	2.0	2008	21	2.0

[a] The lowest mean concentration of four years (2008-2011).
[b] The highest mean concentration of four years (2008-2011).
[c] Infant porridge has only concentrations for one year (2008). The highest flour concentration was chosen, which was for oat.
Limits of detection (LODs, µg/kg): DON: 5-20, zearalenone: 2.0.

8.3.2 Calculation of flour consumption

Flour concentrations and types were estimated for all foods reported as being eaten according to the different national surveys. Dishes and foods for which recipes were available in KBS (food database and software system, Department of Nutrition, University of Oslo, Norway) were divided into ingredients. For dishes and foods that contained flour, but recipes were not in the database, the quantitites and types of flour were estimated. Flour amounts were calculated for each person and reported in grams per day. Mean and 95-percentile consumption in grams per day of the different flour categories are presented for each age group in Table 23. Wheat flour is most commonly consumed while wheat bran and oat flakes are consumed less frequently.

Table 23. Consumption of different flour categories in different age groups.

Age	Flour category	Mean (g/day)	95-percentile (g/day)
1-year-olds	Sieved wheat flour	23	55
	Milled wheat flour	24	75
	Infant porridge	58	149
	Oat flakes	3	17
2-year-olds	Sieved wheat flour	51	83
	Milled wheat flour	48	110
	Infant porridge	5	39
	Oat flakes	8	32
4-year-olds	Sieved wheat flour	69	115
	Milled wheat flour	16	33
	Wheat bran	0	1
	Oat flakes	7	29
9-year-olds	Sieved wheat flour	98	167
	Milled wheat flour	20	44
	Wheat bran	0	1
	Oat flakes	9	46
13-year-olds	Sieved wheat flour	109	204
	Milled wheat flour	5	32
	Wheat bran	0	1
	Oat flakes	7	29
Adults	Sieved wheat flour	94	208
	Milled wheat flour	47	117
	Wheat bran	1	4
	Oat flakes	9	45

8.3.3 Exposure calculations

The exposures to mycotoxins given in Table 24 are derived from the product of mycotoxin concentration and consumption for each age group. Two calculations are presented, one with the lowest mycotoxin level and one with the highest mycotoxin level in the years 2008-2011. Both mean and 95-percentiles are presented for low and high mycotoxin levels. The last two columns in Table 24 present exposure to each mycotoxin per kilogram body weight per day. In agreement with the occurrence data (Tables 17, 18, 19, and 22), exposures were calculated for DON and zearalenone.

Table 24. Estimated mean and 95-percentile exposure of the mycotoxins deoxynivalenol and zearalenone for different age groups. Mean low is estimated exposure based on the lowest annual mean mycotoxin concentration in each food product and mean high is estimated exposure based on the highest annual mean mycotoxin concentration in each food product.

Mycotoxin	Mean low (95-perc) µg/day	Mean high (95-perc) µg/day	Mean low (95-perc) µg/kg bw/day	Mean high (95-perc) µg/kg bw/day
1-year-olds (n=1635)[1]				
DON	8.6 (18)	14 (30)	0.89 (1.8)	1.4 (3.1)
Zearalenone	0.22 (0.43)	0.77 (1.5)	0.022 (0.044)	0.078 (0.16)
2-year-olds (n=1674)[2]				
DON	15 (24)	25 (43)	1.1 (1.9)	2.0 (3.5)
Zearalenone	0.23 (0.42)	1. 4 (2.2)	0.018 (0.032)	0.11 (0.18)
4-year olds (n=391)[3]				
DON	13 (21)	21 (36)	0.73 (1.2)	1.1 (2.0)
Zearalenone	0.16 (0.27)	1.7 (2.7)	0.009 (0.016)	0.096 (0.16)
9-year-olds (n=810)[4]				
DON	18 (31)	28 (51)	0.56 (1.02)	0.90 (1.6)
Zearalenone	0.21 (0.39)	2.4 (4.1)	0.007 (0.012)	0.076 (0.13)
13-year-olds (n=1005)[5]				
DON	18 (35)	29 (56)	0.38 (0.72)	0.60 (1.1)
Zearalenone	0.22 (0.40)	2.6 (4.9)	0.005 (0.009)	0.054 (0.10)
Adults (n=1787)[6]				
DON	20 (42)	34 (69)	0.27 (0.55)	0.45 (0.93)
Zearalenone	0.28 (0.58)	2.4 (5.2)	0.004 (0.008)	0.031 (0.067)

[1] 103 (6%) 1-year-olds did not report their individual body weight (bw) and given the group's mean bw 9.9 kg.
[2] 620 (37%) 2-year-olds did not report their individual bw and were given the group's mean bw of 12.8 kg.
[3] 93 (24%) 4-year-olds did not report their individual bw and were given the group's mean bw of 18.0 kg.
[4] 116 (14%) 9-year-olds did not report their individual bw and were given the group's mean bw of 32.0 kg.
[5] 145 (14%) 13-year-olds did not report their individual bw and were given the group's mean bw of 49.5 kg.
[6] 30 (2%) adults did not report their individual bw and were given the group's mean bw of 77.5 kg.
DON – deoxynivalenol (type B trichothecens).

As an example, the relative contributions from the various flour categories are illustrated for DON in 2- and 4-year-olds, and adults in Figure 12. Despite wheat bran and oat flakes having higher mycotoxin concentrations than wheat flour, the consumption of sieved wheat flour is a relatively large contributor to DON exposure among all age groups.

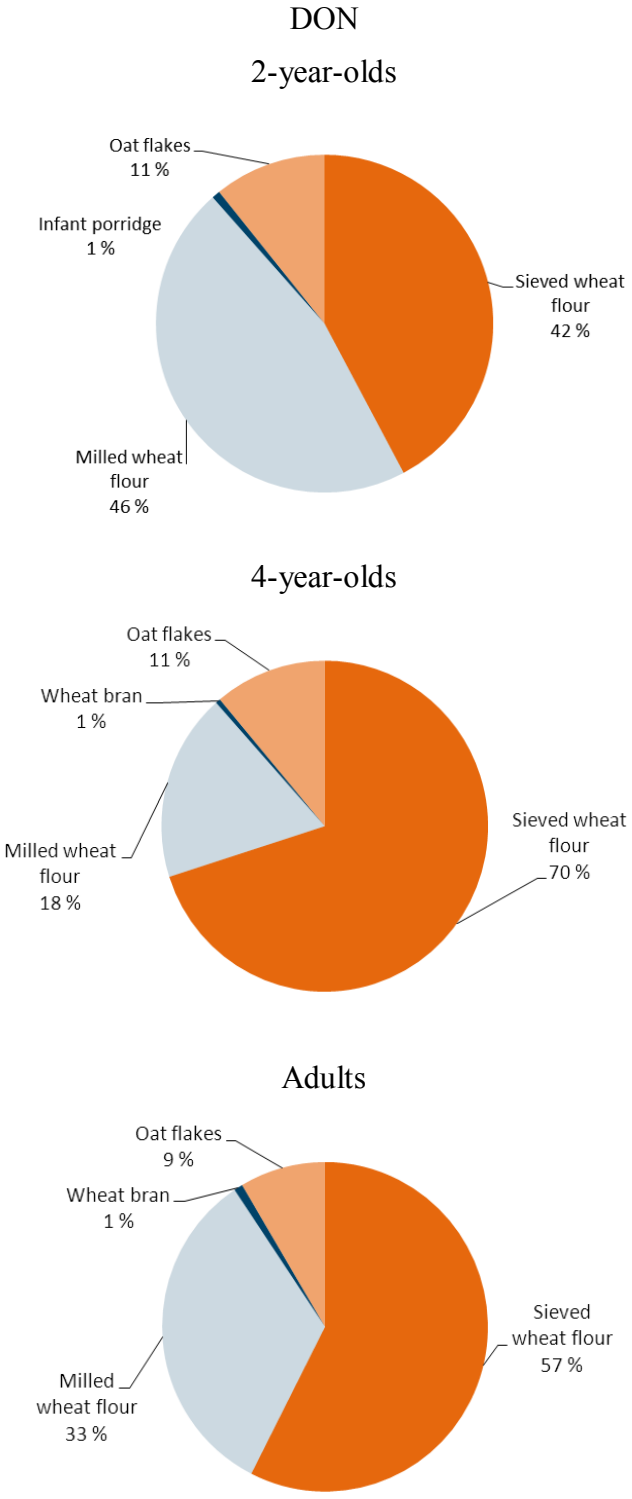


Figure 12. The relative contribution from various flour categories (sieved wheat flour, milled wheat flour, wheat bran, oat flakes and infant porridge) to DON exposure in 2- and 4-year-olds and in adults (Table 24).

8.4 Scenario for chronic exposure to nivalenol and T-2 and HT-2 toxins

Since the concentrations of nivalenol, T-2 and HT-2 toxins were below the LOD in more than 90 % of the wheat flour and oat flakes samples (Tables 17 and 18), exposure to these toxins could not be calculated from measured data. Chronic exposure scenarios were therefore estimated using the concentrations given in Table 25.

None of the samples of *sieved wheat flour* contained measurable concentrations of nivalenol, T-2, and HT-2. Even when LODs were as low as 2.5 µg/kg for T-2 and HT-2 and 5 µg/kg for nivalenol for some samples (see Table 7), the mycotoxins were not detected. Furthermore, T-2 and HT-2 toxins were hardly detected in unprocessed wheat grain (See Table 8). VKM therefore assumed that the use of half of the lowest LOD is an overestimate of the nivalenol, T-2, and HT-2 concentrations in sieved wheat flour. With the aim of providing a conservative scenario, i.e. implementing optimal consumer protection, for chronic exposure from sieved wheat flour, half of the lowest LODs for nivalenol (2.5 µg/kg) and the sum of half of the lowest LODs for T-2 and HT-2 toxins (1.25 + 1.25 µg/kg) were used.

A few samples of *milled whole wheat flour* and *wheat bran* contained T-2 and HT-2 concentrations above the LODs, but the mean values were still below the LOD (See Table 17). VKM therefore assumed that concentrations equal to the lowest LOD (5 µg/kg for nivalenol and 5 µg/kg for the sum of T-2 and HT-2) in milled wheat flour and wheat bran are relevant for calculating the chronic exposure scenarios from these toxins.

A few samples of oat flakes contained levels of T-2 and HT-2 toxins and nivalenol above LOD (See Table 18). Therefore, the sum of the measured concentrations of T-2 and HT-2 were used in the scenario. Data from 2011, the year with the highest mean concentrations of the years 2008 to 2011, were used. In samples in which T-2 and HT-2 toxins and nivalenol were not detected, the concentrations were set to half the respective LODs.

Table 25. Mycotoxin concentrations of nivalenol and sum T-2 and HT-2 toxins in the different flour categories used in the chronic exposure scenario calculations.

Mycotoxin	Flour category	Selected concentrations used for calculations of the scenarios (µg/kg)
Nivalenol	Sieved wheat flour	2.5
	Milled wheat flour	5.0
	Wheat bran	5.0
	Oat flakes[1]	5.0
	Infant porridges[2]	4.4
Sum T-2 and HT-2	Sieved wheat flour	2.5
	Milled wheat flour	5.0
	Wheat bran	5.0
	Oat flakes[1]	45.7
	Infant porridges[2]	10.2

[1] Oat flakes: The highest mean concentration of four years (2008-2011).
[2] For infant porridge, concentration data were only available (2008). Oat based porridges was selected. As it had the highest concentration.

The exposure scenarios based on the chosen mycotoxin concentrations shown in Table 23 are given in Table 26.

Table 26. Chronic exposure scenarios to nivalenol and sum T-2 and HT-2 toxins.

	Scenario Mean (95-percentile) µg/day	Scenario Mean (95-percentile) µg/kg bw/day
1-year olds (n=1635)[1]		
Sum T-2 and HT-2	0.90 (2.0)	0.091 (0.21)
Nivalenol	0.45 (0.89)	0.046 (0.092)
2-year olds (n=1674)[2]		
Sum T-2 and HT-2	0.81 (1.9)	0.063 (0.15)
Nivalenol	0.44 (0.80)	0.034 (0.063)
4-year olds (n=391)[3]		
Sum T-2 and HT-2	0.57 (1.7)	0.033 (0.093)
Nivalenol	0.29 (0.53)	0.016 (0.031)
9-year-olds (n=810)[4]		
Sum T-2 and HT-2	0.74 (2.6)	0.023 (0.077)
Nivalenol	0.39 (0.72)	0.012 (0.023)
13-year-olds (n=1005)[5]		
Sum T-2 and HT-2	0.61 (2.0)	0.013 (0.041)
Nivalenol	0.39 (0.74)	0.008 (0.016)
Adults (n=1787)[6]		
Sum T-2 and HT-2	0.88 (2.6)	0.017 (0.036)
Nivalenol	0.52 (1.1)	0.007 (0.014)

[1] 103 (6%) 1-year-olds did not report their individual body weight (bw) and given the group's mean bw 9.9 kg.
[2] 620 (37%) 2-year-olds did not report their individual bw and were given the group's mean bw of 12.8 kg.
[3] 93 (24%) 4-year-olds did not report their individual bw and were given the group's mean bw of 18.0 kg.
[4] 116 (14%) 9-year-olds did not report their individual bw and were given the group's mean bw of 32.0 kg.
[5] 145 (14%) 13-year-olds did not report their individual bw and were given the group's mean bw of 49.5 kg.
[6] 30 (2%) adults did not report their individual bw and were given the group's mean bw of 77.5 kg.
T-2 ((2α,3α,4β,8α)-4,15-bis(acetyloxy)-3-hydroxy-12,13-epoxytrichothec-9-en-8-yl 3-methylbutanoate).
HT-2 (15-Acetoxy-3α,4β-dihydroxy-8α-(3-methylbutyryloxy)-12,13-epoxytrichothec-9-ene).

8.5 Scenario for acute exposure of DON

DON is the only mycotoxin for which an acute reference dose (ARfD) has been established (JECFA 2011).

Two different scenarios of exposure to DON from a single meal were calculated in order to estimate the acute exposure to DON for comparison with the ARfD. One scenario was made for exposure to DON from a single meal of oat flakes or oat porridge and another scenario was made for the exposure to DON from one single meal of wheat-based bread for 2-year-olds and adults, respectively.

Oat flakes have relatively high DON concentrations (Table 18), and even if oat flakes are not the cereal species that contributes most to the average mean mycotoxin exposure, it is considered easy to eat large portions of oat flakes as breakfast cereal or oatmeal porridge.

The largest single portion of oatmeal porridge registered in the dietary survey (Ungkost) was 500 gram (g) and is based on 60 g dry oat flakes. According to the recipe in KBS, 100 g oatmeal porridge correspond to 12 g dry oat flakes.

A portion of 500 g oatmeal porridge is considered large for 2-year-old children, while adults will be able to eat twice or even three times as much. A 1500 g oatmeal porridge portion contains 180 gram dry oat flakes. VKM considers a 500 g oatmeal porridge portion for 2-year-old children and a 1500 g oatmeal porridge portion for adults to be worst-case scenarios. These portions were used in comparision to the ARfD of DON.

The following scenario calculations for children and adults are also shown in Table 27:

- Acute exposure to DON from a 500 g oatmeal porridge portion, children:
  (60 g oat flakes x 327 µg DON/kg) / 12.8 kg (2-year-olds) = 1.5 µg DON/ kg b.w.
- Acute exposure to DON from a 1500 g oatmeal porridge portion, adults:
  (180 g oat flakes x 327 µg DON/kg) / 77.5 kg (adults) = 0.76 µg DON/ kg b.w.

Table 27. Acute exposure scenarios to deoxynivalenol in children and adults.

Age	Portion oatmeal porridge (gram)	Dry oat flakes[1] per portion oatmeal porridge (gram)	DON concentration[2] (µg/kg oat flakes)	Exposure[3] (µg/kg bw/day)
2-year-olds	500	60	327	1.5
Adults	1500	180	327	0.76

[1] 100 g oatmeal porridge is based on 12 g dry oat flakes (KBS 2011).
[2] The year of highest mean deoxynivalenol (DON) concentration was 2009 (see Table 22).
[3] Body weight used for 2-year-old children is 12.8 kg and for adults 77.5 kg.

In Table 27, the exposure scenarios are based on consumption data and measured DON-concentrations in the consumed food. Estimates can also be made of how much consumers in different age groups would have to eat in order to reach or exceed the ARfD of DON by referring to their respective body weights, as follows:

Assuming a body weight of 12.8 kg, a 2-year-old child would have to consume 313 g oat flakes or 585 g wheat-based bread (approximately 15 slices of bread) in order to exceed the ARfD.

Similarly, an adult of 77.5 kg body weight would have to consume 1713 g of oat flakes or 3200 g bread (approximately 80 slices of bread) to exceed the ARfD.

9 Hazard characterisation – mycotoxins in cereals for feed and food

The mycotoxins known to be present in cereals are mainly produced by the *Fusarium*, *Aspergillus*, *Penicillium* or *Alternaria* genera (Chapter 2). These fungi produce a range of mycotoxins with large diversity in both chemical structures and biological effects. Some mycotoxins are well-studied and several risk assessments of these have been made. This risk assessment is based on the available hazard characterization from international organizations such as JECFA and EFSA when such evaluations are available. Other mycotoxins have been less studied and the risks related to these mycotoxins may not have been assessed previously. Available effects levels (No Observed Adverse Effect Levels (NOAEL), No Observed Effect Levels (NOEL), LOAEL, LOEL, and effects thresholds as Benchmark dose level are derived from other risk assessments or reviews, or effect levels were identified in the present assessment based on original available scientific studies..

The following mycotoxins have been included in the present assessment and are described: trichothecenes (deoxynivalenol (DON), HT-2, T-2, nivalenol), fumonisins, enniatins, beauvericin, moniliformin, zearalenone, ochratoxin, aflatoxins.

Some of these toxins, such as aflatoxins and ochratoxin A, have been extensively studied over several years and a vast amount of information is available. Other toxins, such as enniatins and moniliformin, are currently termed “emerging mycotoxins” and the available information is scarce.

Several feeding studies with the main mycotoxins have been conducted on pig, which is considered to be a sensitive species for several mycotoxins. Even for poultry, there is a reasonable amount of information for the main mycotoxins in cereals. For other species, the available information is much more limited, probably partly because species such as cattle seems to be rather tolerant to several mycotoxins, and partly due to the cost of performing feeding studies with large animal species such as cattle and horses.

The information available about the effects of mycotoxins on dog and cat is also limited.

Most dietary studies on farmed finfish include exposure studies on rainbow trout (*Oncorhynchus mykiss*), Nile tilapia (*Oreochromis niloticus*), channel catfish (*Ictalurus punctatus*), Indian major carp (*Labeo rohita*) and common carp (*Cyprinus carpio L*). Few studies have been performed for the finfish species that is of most importance for Norwegian aquaculture, namely Atlantic salmon. Most dose response dietary exposure studies evaluate acute and general sub-chronic (growth, feed conversion, haematology and organ histopathology) toxicity. Studies on mycotoxin-specific toxic effects (e.g. oestrogenic disruption for zearalenone, and sphingolipid signalling pathways for fumonisin B1) have been mostly studied in *in vitro* trials or intraperitoneal (*i.p.)* injected fish that makes them less relevant for dietary exposure assessment. There is limited scientifically published information on the absorption and toxicokinetics of several mycotoxins in farmed finfish species relevant for Norwegian aquaculture, and on the carry-over of dietary mycotoxins into the edible parts of the fish.

9.1 Trichothecenes

The trichothecenes are a large group of toxins, frequently occurring in mixtures. The group includes the widespread mycotoxins DON, acetylated forms of DON, T-2 and HT-2 toxins, nivalenol as well as several other far less commonly occurring compounds. The toxins are

closely related sesquiterpenoids (ring structure) with a 12, 13 epoxyring and a variable number of hydroxyl, acetoxy or other substituents. Trichothecenes are heat-stable and do not degrade during cooking or baking. Polarity and water solubility vary among the toxins. Nivalenol is the most water-soluble and T-2 the least water soluble of the trichothecenes regularly occurring in grains. This affects the loss of toxins during certain food processing procedures, such as cooking of pasta. The trichothecene of most concern is DON, along with HT-2, T-2 and nivalenol. DON, T-2 and HT-2 are the trichothecenes most frequently found in Norwegian cereal grain. Nivalenol and the acetylated forms of DON, 3-acetyl DON and 15-acetyl DON, are less frequently found (Tables 8-10; Langseth and Rundberget 1999). Other trichothecenes are only rarely found and in low concentrations in Norwegian grains (Langseth and Rundberget 1999). DON, T-2 and HT-2 have previously been assessed by SCF, EFSA and JECFA. SCF and Nordic Council of Ministers have evaluated only nivalenol.

9.1.1 Deoxynivalenol (DON)

DON is the most frequently occurring mycotoxin in Norwegian grains (see occurrence, Chapter 6). This toxin has previously been assessed as a contaminant in food by Nordic Council of Ministers (Eriksen and Alexander 1998), the European Scientific Committee for Food (SCF 1999) and JECFA (2001 and 2011), as well as by EFSA as a contaminant in feed (EFSA 2004c). In the latest evaluation of DON, JECFA decided to change the provisional maximum human tolerable daily intake (PMTDI) for DON set by SCF (1999) and JECFA (2001) into a group TDI covering DON and the acetylated forms (mainly 3- and 15-acetyl DON). It was decided not to include the glucoside conjugates in the group TDI due to lack of data (JECFA 2011). JECFA also established an acute reference dose (ARfD) for DON, based on the acute vomiting observed in pigs. This risk assessment will be based on the TDI and ARfD set by JECFA for human risk assessments. EFSA has also made an evaluation of DON as a contaminant in animal feed, but concluded that no safe level of DON in animal feed could be established based on the available data (EFSA 2004c).

In addition to DON, there are international reports on the occurrence of conjugated forms of DON in grains (see JECFA 2011). The occurrence of glucoside conjugates is not well studied. According to Danish studies, the occurrence of DON 3-glucoside in grains may constitute 0 – 40 % (mean ca. 15 %) of the DON levels in grains (Rasmussen 2010). There is currently no published information about the occurrence of DON glucosides in Norwegian grain. 3- and 15-acetyl DON were previously included in the multi-toxin method used for trichothecenes in Norwegian grains (Langseth and Rundberget 1999). 3-Ac-DON was frequently found in levels of approximately 10 – 15 % of the DON concentration in the same samples. 3-Ac-DON is rapidly deacetylated *in vivo* in pigs and the acetylated forms are included in the PMTDI set by JECFA in the latest evaluation (JECFA 2011). The acetylated forms are, however, not included in the occurrence data used in the exposure estimations in this report.

The critical effects of DON are reductions in feed intake and body weight gain, and impairment of the immune system. These effects have been observed in both laboratory and farm animals (SCF 1999; JECFA 2001, 2010; Pestka 2010). Many other organs are also affected by DON, but only at higher doses.

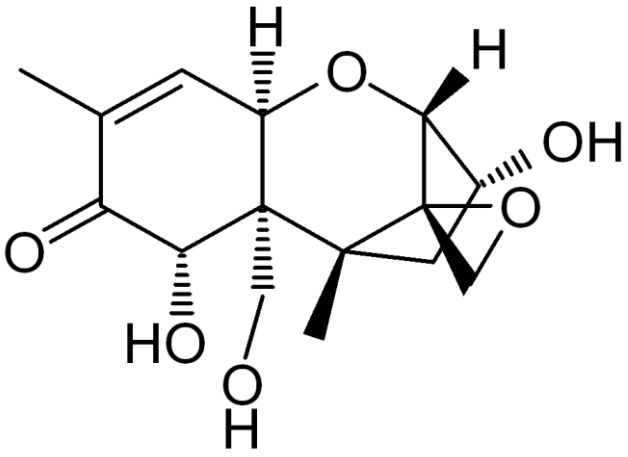


Figure 13. Chemical structure of deoxynivalenol

9.1.1.1 *Mode of action of DON*

The mode of action of DON has been reviewed several times, including by EFSA (2004c) and Pestka (2010). Early on, it was described that trichothecenes, including DON, bind to the 60S ribosomal subunit and inhibit protein synthesis by interfering with the activity of the enzyme peptidyl transferase, an integral part of the ribosome. Various other effects, including inhibition of DNA synthesis, were considered to be secondary to the inhibition of protein synthesis. During recent years, however, there is accumulating evidence that the principal effects of low concentrations of DON are related to up-regulation of proinflammatory cytokines such as TNFα and IL-6, both mediated through increasing cellular levels of transcription factors, such as AF-1, NFκB and C/EBP, and by stabilization of TNFα and IL-6 mRNA (EFSA 2004c; Pestka 2010).

9.1.1.2 *Toxicokinetics of DON*

The toxicokinetics of DON have been reviewed in previous risk assessments (Eriksen and Alexander 1998; EFSA 2004c; JECFA 2001, 2011), as well as in recent review papers such as Dohnal et al. (2008) and Wu et al. (2010).

DON is rapidly absorbed from the gastrointestinal tract after oral ingestion and in most studies is already detectable at the first sampling time (< 30 min). DON is distributed to most of the body, with no particular accumulation in any specific organs. The higest concentrations are found in the intestines and lymphoid tissues. The main metabolic transformation of DON in monogastric animals is by glucuronide conjugation, but a de-epoxide metabolite has also been reported from rat plasma and traces of the de-epoxide metabolite have even been reported from human urine. Several studies have shown that DON is transformed to the de-epoxide when incubated with faeces or content from the large intestine, while much less de-epoxide, or none at all, was found following incubation with the content from the upper part of the gut. Rats have a higher absorption from the lower parts of the guts than other species, which may be the reason for the detection of de-epoxide metabolite in rat plasma. It may, however, also be an effect of the coprophagous activity of rats. DON is rapidly excreted in urine and faeces and only trace amounts are found after more than 24 hours. Due to rapid elimination, steady state is not reached in the plasma of pigs fed DON in the diet twice daily. The ratio of DON and DON glucoronides in faeces versus urine varies between species. The

human data are not known, even though DON and metabolites are found in human urine, and urine concentration has been found to be a suitable biomarker of exposure (Turner et al. 2008 2010, 2011).

Ruminants are less sensitive to trichothecenes than other terrestrial monogastric species. This is considered to be mainly due to the extensive metabolic transformation to the very low toxic de-epoxide metabolite in the rumen prior to absorption (EFSA 2004c; Eriksen and Pettersson 2004).

Preliminary results from an ongoing project on toxicokinetics of DON in salmon indicate no accumulation of the mycotoxin in the fish after an eight-week exposure of up to 6 mg/kg feed (Bernhoft et al., manuscript in preparation). Only traces of DON slightly above the level of quantification (30 µg/kg) were measured in muscle, liver, kidney and brain. *In vitro* studies showed that the gut microbe community from one of nine different fish species was able to transform DON to the DON–de-epoxide, indicating that this detoxification ability may occur even in fish (Guan et al. 2009).

9.1.1.3 *Effects of DON in domestic animals*

The effects of DON in farm animals have been reviewed (Eriksen and Pettersson 2004) and a risk assessment of DON in animal feed was published by EFSA (2004c). EFSA concluded that pig is the most sensitive species, but that no dose-effect relationship could be established for DON in the feed based on the available information (EFSA 2004c).

Pig

Pigs are generally regarded as being the most sensitive species to trichothecenes and a large number of feeding studies with DON have been published. Studies have been conducted with either pure DON, culture material, or naturally or artificially contaminated grains mixed into the diet. Effects of pure DON added to feed and of naturally contaminated material containing similar levels of DON have been compared, and greater effects when naturally contaminated materials are used have been demonstrated (reviewed by EFSA 2004c). The explanation for this is currently not properly understood.

A reduced feed intake and subsequently slower growth are normally the most sensitive endpoints in feeding studies with growing pigs. Trials in which naturally or artificially contaminated grain has been mixed into the diet have shown decreased feed consumption, visible feed aversion, and reduced weight gain, with LOAELs varying from 0.35 to 2 mg DON/kg feed in different studies (reviewed by Eriksen and Pettersson 2004; EFSA 2004c). In several of these studies the reduction of feed intake at the lowest dose was temporary, and the animals managed to compensate for some or most of the reduction in weight gain later in the growing period. In other experiments, no effects on feed intake or weight gain have been demonstrated in pigs given 0.6-0.9 mg/kg feed. Norwegian farmers and feed industry have claimed there is a temporary reduction in appetite and increased stress when new feed batches with DON levels above approximately 0.4 mg/kg are introduced in the pig sty.

Emesis is usually the most sensitive functional manifestation of acute toxicity of DON. Young et al. (1983) showed that pigs vomit when fed DON at 20 mg/kg feed and above, and Pollmann et al. (1985), observed slight evidence of vomiting at 2.8 mg/kg feed. Reduced feed intake was shown at the lowest concentrations of DON contaminated feed used, 1.3 and 1.2 mg/kg. These studies were used by JECFA (2011) for calculating the benchmark dose level

($BMDL_{10}$) between 0.21 and 0.74 mg/kg bw per day for emesis as a basis for establishing a human acute reference dose (ARfD) of DON.

The results of feeding DON during gestation or lactation indicate that sows, as well as their offspring, are less sensitive to DON than directly DON-fed growing pigs (reviewed by Tiemann et al. 2007).

A few studies have been carried out on immune responses towards vaccination, and a delayed response has been demonstrated when pigs are fed 1.8 mg/kg feed (EFSA 2004c). More recently, pigs fed DON contaminated feed at 2.2-2.5 mg/kg for 9 weeks and immunised with ovalbumin showed an increase in total and ovalbumin-specific IgA and IgG, and reduced expression of cytokine mRNA when compared with control pigs (Pinton et al. 2008), indicating an immuno-stimulatory effect at this dosage level.

In sum, in 2004 EFSA concluded in their evaluation of DON that no dose-effect relationship could be established due to the large variation in response from the different experiments. This is still valid. For the critical effect of reduced feed intake, LOEALs may vary from 0.35-2 mg/kg, and NOAELs from 0.6-0.9 mg/kg feed.

Poultry

Many studies of the effects of DON on poultry have been published. The main effects reported are reduction in feed intake and weight gain, and reduced immune response. Chickens have been demonstrated to be far less susceptible than growing pigs, and effects on performance have usually been observed when the feed concentrations reach 16-20 mg DON/kg (reviewed in Eriksen and Pettersson 2004; EFSA 2004c). Increased liver and gizzard weights were measured in chickens fed 9 mg/kg feed (Kubena et al. 1985). A reduced immunological response towards vaccinations has been reported from chickens fed 18 mg DON/kg feed or above (reviewed in EFSA 2004c). EFSA concludes that the available data do not allow the estimation of a maximum tolerance level.

However, recently Yunus et al. (2012a, 2012b) showed significant effects on performance, vaccine response, and intestinal development in male broiler chickens fed DON at 1.68 and 12.2 mg/kg feed from age 7 days, compared with comparable chickens fed a control diet containing 0.265 mg/kg of DON. The DON exposure resulted in a linear decrease in the weekly weight gain during the first 3 weeks of exposure, a linear increase in the titres against Newcastle disease vaccination, a linear reduction in the titres against infectious bronchitis vaccination, and a linear or quadratic increase in the serum protein concentration. In the small intestine, DON at 1.68 and 12.2 mg/kg progressively decreased its relative density (weight : length) with increasing exposure length to DON, which could be correlated with a decrease in villus height in the small intestine.

In a summary of studies on laying hens (EFSA 2004c), it was noted that contaminated maize containing 12 mg/kg of DON temporarily depressed feed intake at the beginning of the experimental period. DON up to 83 mg/kg feed did not have a significant effect on egg production, and hatchability was not affected up to 18 mg/kg. No essential new information has been found regarding the effect of DON on laying hens since that study.

In chick foetuses, an increased incidence of delayed ossification and unwithdrawn yolk sac have been reported when the breeder hens were fed 2.5 and 3.1 mg DON/kg as naturally contaminated material. The effects were considered as delayed foetal maturation. No malformations were observed in chicks from hens fed 4.9 mg/kg feed (Bergsjø et al. 1993).

To summarise, recent studies of chickens fed 1.7 mg DON/kg feed have demonstrated a reduction in the size of gut villi, lower weight gain, and altered immune responses to vaccinations compared with controls. Thus a LOAEL of 1.7 mg/kg for chicken is identified. For laying hens, a LOAEL of 12 mg/kg is derived. However, greater caution should be taken for breeder hens due to possible foetal effects at 2.5 mg/kg feed.

Ruminants

Ruminants are regarded as being rather resistant to the effects of trichothecenes, including DON. DON is extensively metabolised to the considerably less toxic de-epoxide in the rumen prior to absorption. Seeling and Dänicke (2005) reviewed the relevance of DON in ruminal nutrition. Most feeding studies of DON in ruminants have not observed adverse clinical health or performance effects. This includes studies with dairy cows where DON up to 12 mg/kg concentrate (6.5 mg/kg total diet) was given to low-producing cows (about 20 kg milk daily) over 70 days (Charmley et al. 1993), and where DON up to 14.6 mg/kg concentrate (8.5 mg/kg total diet) was given to medium-producing cows (about 30 kg milk daily) over 21 days (Ingalls 1996). Short-term feeding of DON at 66 mg/kg diet during five days did not affect feed intake or milk production (Cote et al. 1986). In heifers and steers, up to 10 mg/kg feed was given for a period of 126 days without apparent effects on performance, clinical health, serum biochemistry or tissue histology (Nelson et al. 1984). Similarly no effects on feed consumption, weight gain, feed efficiency, haematology, serum biochemical variables, or- gross and- microscopic pathology were found in lambs fed DON at 15.6 mg/kg feed for 28 days (Harvey et al. 1986).

There are, however, reports (not peer reviewed) suggesting that lower doses of DON may decrease feed intake in fattened beef cattle (Schuh 1996) and milk production in dairy cow herds (Whitlow et al. 1986). Seeling and Dänicke (2005) indicated that sheep and beef cattle are normally less sensitive to DON than dairy cows, and that low-producing dairy cattle are more tolerant to DON than high milk producers. This may be due to greater stress, lower immunity and faster rumen turnover in high-producing animals. Furthermore, diets containing high levels of concentrate increase ruminal turnover and reduce ruminal pH, which inhibits DON biotransformation. Thus, ruminants on intensive concentrate feeding that results in chronical ruminal acidosis, or animals with other metabolic diseases, as well as animals that in other ways are forced to provide high production rates, may be more sensitive to adverse DON effects than ruminants in normal rumen-physiological balance.

Young pre-ruminanting calves, lambs and kids should be regarded as similarly sensitive to DON as monogastric mammals (Seeling and Dänicke 2005). Thus, before the development of a functional rumen at about 2-3 months of age their tolerance may be rather low. However, cereal intake is usually very limited in these young animals.

It is difficult to draw conclusions for the practical feeding with DON contaminated feedstuff to ruminants. However, DON in total rations of up to 6 mg/kg appears to be safe under normal rumen-physiological circumstances, but intensive feeding with compound feed may increase susceptibility.

Horse

There are few studies on the effects of DON in horses, but the results available indicate that this species is moderately sensitive – more susceptible than ruminants, but less sensitive than pigs. Raymond et al. (2003, 2005) conducted two studies in horses, which were fed

concentrate naturally contaminated with mycotoxins and containing DON at 14 and 11 mg/kg respectively (5.0 and 4.5 mg DON/kg total diet, respectively) and lower concentrations of 15-acetyl-DON and zearalenone for 21 days. The first study tested serum chemistry and haematology, and the second study used exercised horses and tested for athletic ability. In both studies, feed intake was reduced compared with controls, and in the second study, reduced body weight was also found. Except for gamma-glutamyl transferase, which was higher in horses fed contaminated feed, no significant effects were observed on haematological parameters, serum chemical parameters, and athletic ability.

Barley naturally contaminated with DON at 36-44 mg/kg (other trichothecenes were not detected) was fed to adult horses for 40 days (Johnson et al. 1997). The mean barley intake was 1.3 kg per day. The DON content of the whole diet was not measured by the authors, but is assumed to be approximately 6-7 mg/kg diet. No evidence of physical or haematological effects was found, but there were some statistically significant changes in serum biochemistry within the normal variation. Recently, a study of immune parameters in mares fed 2 kg oats with DON at 20.2 mg/kg for 2 weeks compared with corresponding amounts of oats with 0.49 mg/kg using a crossover design was reported (Khol-Parisini et al. 2012). The DON level in the contaminated oats corresponds to 5 mg/kg total diet. No effect was observed on general health. Serum haptoglobin concentration was significantly elevated, but no effects were shown on lymphocyte subsets or lymphocyte proliferation.

In sum, reduced feed intake, body weight gain or biochemical effects have been shown in studies of horses fed DON corresponding to approximately 5 mg/kg total diet.

Rabbit

Concentrations of up to 3 mg DON/kg feed for 5 weeks (highest concentration used) did not induce any overt clinical signs, reduced performance, or immunosuppression in young rabbits (Biró 2003). In rabbits fed diets containing DON on days 0-30 of gestation, decreased foetal weight was shown at 30 mg/kg diet (1.0 mg/kg bw) and above, and maternal body weight was reduced at 7.5 mg/kg diet (0.3 mg/kg bw; lowest dose) and above (Khera et al. 1986).

The available data for rabbits are too few to allow establishment of a safe limit for DON in feed. So far no effects have been reported from rabbits fed up to 3 mg/kg feed, while a reduced maternal body weight was reported from pregnant female rabbits fed a diet containing from 7.5 mg/kg diet.

Dog and cat

Groups of dogs were fed DON-contaminated wheat mixed into their diet at concentrations of 0, 1, 2, 4, 6, 8 and 10 mg/kg feed for 14 days, with two to fourteen dogs per diet. Feed intake was reduced in dogs fed 6 mg/kg and above, and vomiting occurred in dogs fed 8 and 10 mg/kg (Hughes et al. 1999). A threshold for reduced feed intake in the dogs was estimated at 4.5 mg/kg diet. In a similarly designed study for cats (2 cats per group, except the group fed 10 mg/kg with 8 cats), reduced feed intake was observed, with vomiting only observed at 10 mg DON/kg (Hughes et al. 1999). A threshold for reduced feed intake in the cats was estimated at 7.7 mg/kg diet.

In a study with 12 dogs in a 3x3 Latin square study design, exposure to 2.7 mg DON/kg feed plus traces of 15-acetyl DON (0.2 mg/kg) and zearalenone (0.3 mg/kg) for 14 days caused a significant reduction in feed intake and body weight, and also reduced serum total protein and fibrinogen alkaline phosphatase (within reference limits) compared with dogs on a control

diet with 0.3 mg/kg of DON and not quantifiable 15-acetyl DON and zearalenone (Leung et al. 2007). The diets also contained detectable amounts of fusaric acid. Although the fusaric acid levels were far below those expected to produce adverse effects, the authors discuss the possibility of a potentiation of the effect from fusaric acid on the DON-induced anorexia. A third diet was mycotoxin-contaminated feed with additional polymeric glucomannan mycotoxin adsorbant, but this was not shown to ameliorate the mycotoxin effects.

To summarise for dogs, 2.7 mg/kg of DON in the diet resulted in reduced feed intake and body weight, but the effect might have been influenced by other toxins in the feed. Another study estimated a threshold for reduced feed intake in dogs at 4.5 mg/kg diet. In cats, a threshold for reduced feed intake was estimated at 7.7 mg/kg diet.

Fish

Rainbow trout (24 g) were fed diets with increasing levels of DON (0.3 (basal), 0.8, 1.4, 2.0, and 2.6 mg/kg) for 8 weeks (Hooft et al. 2011). Weight gain and growth rate decreased linearly with increasing dietary levels of DON ranging from 0.3 mg/kg to 2.6 mg/kg. Cocurrently, there was a significant effect of feeding diets containing increasing levels of DON on the linear and quadratic decrease of feed intake and feed efficiency. Fish pair-fed 2.6 mg/kg had significantly lower growth rate (expressed as thermal-unit growth efficiency), feed efficiency, and whole body crude protein than the basal diet group. In addition, morphological liver changes were observed in the 2.6 mg/kg group. The other diets were not pair-fed with the basal diets. Adult (405 g) Atlantic salmon were fed DON at 0.3-3.7 mg/kg for 15 weeks; those with the higher DON intake showed reduced feed intake, lower specific growth rate, decreased clinical plasma parameters, and altered histology (Döll et al. 2010).

Smoltified Atlantic salmon (start weight 50-70 g) were fed DON at 0, 0.5, 1, 2, 4 and 6 mg/kg diet for eight weeks (Bernhoft et al., manuscript in preparation). The fish fed DON at 4 and 6 mg/kg showed decreased weight gain, reduced feed intake, and increased relative liver weight. The fish at 6 mg/kg also showed reduced plasma total protein and albumin, as well as decreased blood haematocrit. No effects were observed clinically, measured clinical-chemically, or detected pathologically due to DON at lower concentrations.

In conclusion, a NOAEL for DON in Atlantic salmon at 2 mg/kg diet is identified.

Table 28. Critical effect (most sensitive endpoint) for DON in feed for different animal species at concentrations where NOAEL or other available effect levels are derived/identified.

	Critical Effect	Concentration in total diet	Available effect level	Pivotal references
Pig	Reduced feed intake	0.35 – 2.0 mg/kg 0.6 – 0.9 mg/kg	LOAEL NOAEL	EFSA 2004c Eriksen and Pettersson 2004
Broiler chicken	Reduced size of gut villi, affected immune response, reduced weight gain	1.7 mg/kg	LOAEL	Yunus et al. 2012a, 2012b
Laying hen	Reduced feed intake	12 mg/kg	LOAEL	EFSA 2004c
Breeder hen	Foetal effects	2.5 mg/kg	LOAEL	Bergsjø et al. 1993
Ruminants	Reduced feed intake	6 mg/kg	NOAEL	Seeling and Dänicke 2005
Horse	Reduced feed intake, body weight gain or biochemical effects	Approximately 5 mg/kg	LOAEL	Raymond et al. 2003, 2005
Rabbit	Reduced maternal body weight	7.5 mg/kg	LOAEL	Khera et al. 1986
Dog	Reduced feed intake and body weight	2.7 mg/kg	LOAEL	Leung et al. 2007
Cat	Reduced feed intake	7.7 mg/kg	Threshold	Hughes et al. 1999
Rainbow trout	Reduced growth rate and feed efficiency, liver changes	2.6 mg/kg	LOAEL	Hooft et al. 2011
Atlantic salmon	Reduced feed intake and body weight gain, increased liver weight	2.0 mg/kg	NOAEL	Bernhoft et al, in preparation.

9.1.1.4 *Carry-over of DON to humans through animal-derived food products*

DON is generally considered to be rapidly excreted from animals, without accumulation in any particular organ (see Toxicokinetics). DON and its metabolites were not detected in milk from cows fed DON at 6 mg/kg in concentrate for 10 weeks. De-epoxide metabolites have mainly been detected in milk from cows fed with extremely high trichothecene levels (66 mg DON/kg concentrate for 5 days). For review see Cavret and Lecoeur (2006).

Small traces of DON and de-epoxy DON were found in eggs from home producers of eggs in Belgium (Tangni et al. 2009), while no DON (Valenta and Däniche 2005) or only traces of DON have been found in eggs from experiments in which laying hens have been given DON in the feed (Sypecka et al. 2004; Prelusky et al. 1989). The authors of these studies, as well as JECFA and EFSA, have all concluded that although low levels of DON may be found in eggs, the consumption of eggs is unlikely to contribute significantly to the total dietary intake of DON in humans.

No accumulation of DON in fish tissues, including muscle, was observed when Atlantic salmon were fed with DON at concetrations up to 6 mg/kg feed for 8 weeks (Bernhoft et al., manuscript in preparation).

VKM supports the conclusions from JECFA and EFSA, that human exposure through consumption of animal-derived food products is minimal in comparison with cereal-based food products (JECFA 2001, 2011; EFSA 2004c).

9.1.1.5 *Effects of DON in humans and animals relevant for human risk assessments*

DON induces acute vomiting and feed refusal in experimental and farm animals at relatively low concentrations. Both these effects have been linked with increased levels of the neurotransmitter substance serotonin and serotonin analogues in the brain, but other mechanisms are currently being studied. Single doses of DON also damage rapidly dividing cells, such as epithelial cells, in the gastrointestinal tract. Reduction in feed intake and weight gain and impairment of the immune system have been the most consistent findings from feeding studies with DON in species such as rodents and pigs. In rodents, activation of the immune system has been found at low doses, while decreases in a range of immune responses have been reported at higher doses (reviewed in Pestka 2010). The feed conversion ratio (ratio between weight gain and feed intake) is often reduced.

Several toxicity studies have been conducted to compare the toxicity between pure DON added to feed and feed containing naturally contaminated material. In these studies, naturally contaminated feed was more toxic. The reason for this has not been elucidated, but hypotheses include: the presence of other, unknown fungal metabolites, alterations in nutritional value, presence of other compounds (for example bacterial polysaccharides) affecting the toxicity, or induction of taste or smell aversion (JECFA 2001; Eriksen and Pettersson 2004; EFSA 2004c).

DON was negative in Ames tests, but chromosomal aberrations, mainly gaps, were found *in vitro*. The overall significance of these findings was considered to be equivocal (JECFA 2001).

The available human risk assessments of DON from JECFA (2001, 2011) and SCF (1999) are based on a 2-year feeding study in which mice were given DON (purity > 95%; no 3-acetyl- or 15-acetyldeoxy-nivalenol) in the feed. No increases in preneoplasts or neoplasts were found. There were, however, significant decreases in feed intake and weight gain in males, and increased serum IgA (56 %) and IgG (< 10 %) in females at 0.7 and 1.6 mg/kg bw (Iverson et al. 1995). In addition, increased relative weights of testis and decreased relative weights of liver and spleen were reported from male mice (Iverson et al. 1995). JECFA also established a ARfD, based on the estimated dose causing vomiting in pigs (JECFA 2011).

9.1.1.6 *Human hazard characterizations of DON*

The available human risk assessments of DON from JECFA (2001, 2011) and SCF (1999) are derived from the 2-year feeding study in which mice were given DON in their feed. The NOEL in this study was 0.1 mg/kg bw. Based on this NOEL, and using a safety factor of 100, JECFA established a provisional maximum tolerably daily intake (PMTDI) of 1 µg/kg bw. JECFA concluded that this PMTDI would also protect against effects on the immune system, growth, and reproduction (JECFA 2001). In the re-evaluation in 2010, JECFA reconfirmed this PMTDI. The PMTDI was converted into a group TDI for DON and the acetylated derivatives since the acetylated forms are rapidly de-acetylated *in vivo* (JECFA 2011). In addition, JECFA derived an acute reference dose (ARfD) of 8 µg/kg bw for DON and the acetylated derivatives using the lowest $BMDL_{10}$ based on emetic effects in pigs, using an uncertainty factor of 25 from the maximum concentration in plasma (JECFA 2011).

The PMTDI of 1 µg/kg bw and the ARfD of 8 µg/kg are used for DON in the assessment of human risks in this report.

9.1.2 T-2 and HT-2 toxin

HT-2 toxin is a deacetylated form of T-2 toxin. The deacetylation of T-2 toxin occurs both in plants/fungi and animals exposed to T-2 toxin. HT-2 toxin is a major metabolite of T-2 toxin in animals and appears rapidly in plasma after exposure to T-2 toxin (Wu et al. 2010; EFSA 2011a). There are very few studies on the effects of HT-2 toxin available, but due to the rapid *in vivo* metabolism of T-2 toxin to HT-2 toxin, it is not possible to distinguish the effects of T-2 toxin from the effects of HT-2 toxin, and therefore these toxins are assessed together.

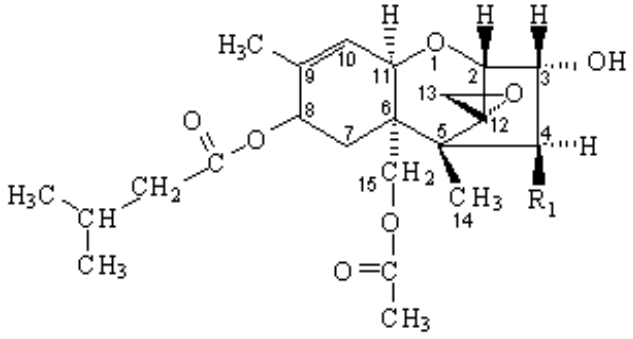


Figure 14. Chemical structures of T-2 toxin (R_1 = OAc) and HT-2 toxin (R_1= OH).

9.1.2.1 *Mode of action of T-2 and HT-2*

T-2 and HT-2 toxins are known inhibitors of protein synthesis. Other cellular effects of T-2 toxin are inhibition of DNA and RNA synthesis, apoptosis, effects on membranes and lipid peroxidation. The exact mechanisms behind all these effects are not completely understood, but may be related to the inhibition of protein synthesis or formation of reactive oxygen species (ROS) (EFSA 2011a). The critical effects of T-2 toxin are impaired function of the immune system and reduced feed intake and growth, but other organs are also affected at higher doses.

9.1.2.2 *Toxicokinetics of T-2 and HT-2*

The toxicokinetics of T-2 and HT-2 have been reviewed in previous risk assessments (EFSA 2011a; JECFA 2001), as well as in review papers such as Wu et al. (2010). Conversion of T-2 toxin starts already in the gastrointestinal tract prior to absorption. The degree of absorption after oral exposure has not been established. T-2 toxin disappears rapidly from plasma, having a plasma half-life of less than 20 minutes (reviewed in JECFA 2001; Wu et al. 2010; EFSA 2011a). Orally administered T-2 toxin in mice and rats is rapidly distributed to the liver, kidneys and other organs without accumulation in any organ. T-2 toxin is also able to cross the placenta and reach foetal tissues. T-2 toxin undergoes extensive metabolism to a large number of metabolites. The metabolic transformations of T-2 toxin are hydroxylations, de-acetylations, hydrolysis, and glucuronide conjugations, and in ruminants, acetylations and de-epoxidation. Except for de-epoxidation, the reactions may occur at multiple sites of the molecule, and consequently several different metabolites have been found. The transformations known to have significant impact of the toxicity are de-epoxidation and glucuronide conjugations. Some of the hydroxyl-metabolites are slightly more cytotoxic than T-2 toxin. T-2 toxin itself has been shown to have a very short plasma half-life, being less than 10 minutes in some studies, but the retention of T-2 and its metabolites seems to be

somewhat longer than for DON, and traces of metabolites have been found in plasma and urine 5 days after exposure. There are no signs of specific accumulation in any tissue in any species. The highest concentrations are found in gastrointestinal and lymphoid tissues.

9.1.2.3 *Effects of T-2 and HT-2 in domestic animals*

T-2 and HT-2 are relatively potent toxins that have shown highly acute toxicity experimentally, although acute lethal toxicity after dietary exposure at relevant concentrations in feed is unlikely. The important issue is the lower dietary concentrations that may influence animal health, welfare and production. The concern is on critical effects (sensitive endpoints), usually after exposure over time.

Pig

Several T-2 feeding studies in piglets have been published, but data on older slaughter pigs and adults are very scarce. Schumacher-Wolz et al. (2010) and EFSA (2011a) have summarized these studies. Rafai et al. (1995a, 1995b) demonstrated effects on immune response variables and lymphoid tissues at low concentrations. Other reports have shown corresponding subclinical effects at similar levels in piglets (Bernhoft et al. 2000; Meissonnier et al. 2008). The most important findings from these studies are given below. In other studies higher concentrations are used, and the results are of less relevance for the risk assessment. The only study of HT-2 in piglets indicated that the immunotoxic potency may be somewhat lower than that of T-2 (Bernhoft et al. 2000).

Rafai et al. (1995a; 1995b) fed seven-week old pigs (9 kg bw) purified T-2 (purity 90 %) at 0.5, 1.0, 2.0, 3.0, 4.0, 10 and 15 mg/kg diet for three weeks. Immunotoxic effects (reduction of T-lymphocyte count, altered response to immunisation with horse globulin and blastogenic response of lymphocytes to the mitogens ConA and PHA, and histological changes of thymus and spleen), as well as reduced feed intake were shown even at the lowest dose (0.029 mg/kg bw per day).

Meissonnier et al. (2008) fed piglets (11 kg bw) with pure T-2 for 28 days and reduced P450 1A-related activities were shown at 0.54 mg/kg diet (lowest concentration, estimated to 0.03 mg/kg bw per day) and above. Reduced antibody production after immunisation with ovalbumin was shown at 1.3 mg/kg diet and above. Body weight gain was reduced only at the highest dosage, 2.1 mg/kg diet.

In piglets orally administered with either HT-2 or T-2 at doses of 0.012 and 0.060 mg/kg bw per day from 1 to 8-weeks of age, increased interleukin 2 and 4 were demonstrated at both T-2 levels (Bernhoft et al. 2000). A reduced lymphocyte response to PHA was found at the highest T-2 level. No corresponding effects were found with HT-2, but reduced IFN-γ was observed at the lowest level of HT-2.

Friend et al. (1992) reported a study on some older pigs (12-13 weeks-old, about 37-39 kg bw) that were fed T-2 at 0.4-3.2 mg/kg diet for 5 weeks. Performance, and pathological, biochemical and haematological variables were examined, and no significant effects were observed, except for a tendency towards lower final body weight and reduced feed intake that appeared to be associated with increasing T-2 content. Immunological tests were not included.

Reproductive effects of T-2 in sows and offspring have been experimentally observed at high concentrations (12 mg/kg diet) (Weaver et al. 1978). In case reports, T-2 is also suspected to cause infertility with inhibited ovary function at lower levels (Glavits et al. 1983; Vanyi et al. 1995), but this has not been confirmed experimentally.

EFSA made an evaluation of T-2 and HT-2 toxins in feed, including pig feed, in 2011. They concluded that the most sensitive endpoints for pigs are immunological or haematological effects that occur from doses of 0.029 mg T-2 toxin/kg bw per day (equivalent to 0.5 mg/kg of T-2 toxin/kg feed), which is a LOAEL. No NOAEL in pigs has been identified to date. EFSA calculated a bench mark dose level ($BMDL_{05}$) of 0.010 mg/kg bw/day from this study, and this is used as a reference point in the risk characterization for pigs. This dose corresponds to approximately 0.17 mg/kg feed. Knowledge on the effects of HT-2 is limited and its effects are regarded to be similar level to those of T-2.

To summarise for T-2 in pigs, 0.5 mg/kg diet is a LOAEL, and EFSA has calculated a $BMDL_{05}$ corresponding to 0.17 mg/kg diet. Knowledge regarding the effects of HT-2 is scarce and its effects are considered to be at a similar level to those of T-2.

Poultry

EFSA (2011a) has reviewed the effects of T-2 toxin in poultry. The critical effects after chronic exposure seem to be lesions in the mucous membranes of the mouth and intestine. Other effects observed at relatively low feed concentrations are reduced body weight gain, reduced egg production and hatchability, and immunomodulatory effects. In chickens and turkeys, the critical effects, lesions in the oral cavity, have been shown at concentrations from 0.5 mg T-2/kg feed. In ducklings, T-2 dietary concentrations from 0.2 mg/kg resulted in reduced weight gain. Immunmodulatory effects have been observed in chickens at concentrations above 1 mg T-2/kg feed. Growth depression in chickens is observed from 2 mg T-2 toxin/kg diet. Effects on reproduction i.e. infertility of eggs and/or reductions in egg production, were observed at concentrations starting from 2 mg T-2 toxin/kg feed (all results from EFSA 2011a). So far, no NOAEL has been derived for poultry.

EFSA concluded in their risk assessment of T-2 and HT-2 toxins in feed that there are no indications that poultry is more sensitive than pigs. As no NOAEL has been identified for poultry, EFSA considered the $BMDL_{05}$ of 0.010 mg/kg bw estimated for pigs to be acceptable for poultry also (EFSA 2011a). Assuming a normal feed consumption for poultry, this would correspond to 0.10 mg/kg feed for chickens and 0.17 mg/kg for laying hens. The estimate for chickens seems to correspond fairly well for actual data, but the estimate for laying hens does not seem to be supported by the observed effects of different concentrations in adult hens.

In summary, no NOAELs have been identified for poultry. However, for chickens and turkeys 0.5 mg/kg feed is considered as LOAEL based on oral lesions. For ducks, a LOAEL has been identified at 0.2 mg/kg feed based on reduced weight gain. For laying hens, a LOAEL at 2 mg/kg feed based on reduced egg production and hatchability has been identified. EFSA considered the $BMDL_{05}$ for pigs acceptable for poultry, and would correspond to 0.10 mg/kg feed for chickens.

Ruminants

There are few studies on the effects of T-2 and HT-2 on ruminants. Purified T-2 toxin was given orally to ewes (0.3 and 0.9 mg/day; 0.005, and 0.015 mg/kg bw per day, estimated to be 0.1 and 0.3 mg/kg total diet) for 21 days to test the effects upon ovarian function in comparison with control animals without T-2 exposure (Huszenicza et al. 2000). Half of the animals were given concentrate-rich feeding to increase the acid condition of the rumen. The T-2 exposed animals on concentrate-rich feeding showed retarded follicle maturation and ovulation. This observation was not generally replicated in ewes fed hay together with T-2

exposure, where only 1 of 5 animals at highest T-2 dose showed reduced reproductive function.

Another study by the same authors was performed on heifers with a single dosage level of T-2 toxin (9 mg/day; 0.025 mg/kg bw per day; estimated to be 1 mg/kg total diet) for 21 days, to test the effect upon ovarian function and compared with control animals without T-2 exposure (Huszenicza et al. 2000). All animals were given concentrate-rich feeding, which increased the acid condition of the rumen. The T-2 exposed heifers showed retarded ovulation and reduced plasma progesterone levels compared with controls.

EFSA (2011a) stated that the effects observed in nutritionally challenged heifers and ewes indicate that rumen detoxification of T-2 toxin may not always be complete and thus effective at preventing the effects on ruminants.

There are also case reports of the effects of T-2 toxin in dairy herds. A high frequency of 5-6 month abortions preceding and coinciding with obvious illness in the affected cows was reported when the feed had been contaminated with T-2 toxin at 2 mg/kg corn concentrate (Hsu et al. 1972). After several days of consumption of 10.5 kg concentrate contaminated with 0.6-1.3 mg/kg T-2 toxin (corresponding to 0.009 mg/kg bw per day or 0.3-0.6 mg/kg total ration), cows on a large-scale dairy unit in Hungary showed anorexia, decreased milk production, bloody diarrhoea and absence of visible oestrus (Kegl and Vanyi 1991). The anorexia and diarrhea normalised rapidly on non-contaminated feed, but ovarian function and milk production first normalised 9-11 days later.

For calves and lambs, EFSA (2011a) concluded that exposure to 0.3 mg T-2 toxin/kg bw per day (approximately 10 mg/kg total diet) or more may result in gastrointestinal lesions, altered serum proteins, and changes in haematological variables. EFSA considered this to be a LOAEL. However, Pier et al. (1976) have reported evidence of dose-response related loose faeces and abomastis at all dosage levels when calves (1 per dose) were exposed to pure T-2 by oral gavage at 0.08, 0.16, 0.32, and 0.6 mg/kg bw per day for 30 days. The calf exposed to the highest dose died after 20 days, and bloody faeces were observed at doses of 0.32 mg/kg and above. From this limited experiment, a LOAEL of 0.08 mg/kg bw per day (approximately 3 mg/kg total diet) may be indicated.

In sum for ruminants, the data are scarce but there are indications of adverse reproductive effects in sheep exposed to T-2 at approximately 0.3 mg/kg total diet (a NOAEL at approximately 0.1 mg/kg total diet) and corresponding effects at approximately 0.1 mg/kg total diet in sheep with induced rumen acidosis. A case report describes health and production effects in dairy cows exposed to 0.3-0.6 mg/kg total feed. Thus, the data indicate that even ruminants are rather susceptible to T-2, particularly when fed high amounts of compound feed. While the data are sparse, those available indicate that pre-ruminants have a similar susceptibility to T-2 as mature ruminants.

Horse

Purified T-2 toxin was mixed into the diet and given to adult mares at 7 mg/horse (0.011—0.013 mg/kg bw per day; estimated to be 0.7 mg/kg total diet) for 32-40 days (Juhasz et al. 1997). No reproductive effects were demonstrated, but feed refusal and skin lesions around the mouth were observed. Haematological and clinical chemical data were not measured.

There are also two reported cases of intoxication in horses (reviewed by EFSA 2011a). The horses had consumed feed with 25 and 204 mg T-2/kg feed, resulting in adverse health effects and also death among the horses exposed to the higher dose.

EFSA concluded in their evaluation that the available data did not provide any basis for establishment of safe levels in horse feed. In the absence of NOAELs or LOAELs for horses, they proposed to use the same $BMDL_{05}$ (0.010 mg/kg bw/day) as that derived for pigs to give an indication on the possible risk, since toxicokinetics of T-2 and HT-2 toxins in horses do not differ substantially from those in pigs. Such a daily dose would correspond to approximately 0.6 mg/kg feed per day. From the study by Juhasz et al. (1997), feed refusal and skin lesions were observed at approximately 0.7 mg/kg total diet, and therefore may indicate that a threshold for effect in horses is lower than 0.6 mg/kg total feed.

To summarise for horses, feed refusal and skin lesions around the mouth were observed at approximately 0.7 mg/kg total diet. EFSA proposed to use the $BMDL_{05}$ (0.010 mg/kg bw per day) as derived for pigs also for horses, corresponding to 0.6 mg/kg total feed.

Rabbit

EFSA concluded in their assessment that the available toxicological data from chronic exposure studies in rabbits show that doses of T-2 ranging from 0.5-2.0 mg toxin/kg bw per day result in a decrease of body weight gain and other signs of toxicity such as gastritis and intestinal necrosis. Only moderate signs, including haematological and hormonal effects, and no signs of toxicity have been observed at doses of 0.2-0.5 mg of T-2 toxin/kg bw per day. A NOAEL of 0.1 mg T-2 toxin/kg bw per day (estimated to be approximately 2 mg/kg diet) was identified.

Dog and cat

Data on the effects on T-2 and HT-2 toxins on dogs were not found. EFSA proposed in their recent evaluation that in the absence of data for dogs, the same reference point ($BMDL_{05}$ at 0.010 mg/kg bw/day) could be used as that derived for pigs to give an indication on the possible risk, since toxicokinetics of T-2 and HT-2 toxins in dogs do not differ substantially from those in pigs. Such a daily dose would correspond to approximately 0.3-0.5 mg/kg feed per day for dogs.

Cats were given oral doses of T-2 toxin on alternate days at 0.06, 0.08 or 0.1 mg/kg bw until death (Lutsky et al. 1978; Lutsky and Mor 1981). Emesis, anorexia, bloody diarrhoea and ataxia were observed. The cats died after 6 to 40 days, and mean survival time was inversely related to the dose. Necropsy revealed haemorrhages in the gut, lymphoid tissues, heart and meninges, as well as pancytopenia. The doses correspond to approximately 1.2-2 mg/kg diet. EFSA (2011a) evaluated the studies and concluded that the particular sensitivity of cats to T-2 toxin may be related to the inability of cats to form excretable glucuronide conjugates.

To summarise, cats are very sensitive to the adverse effects of T-2 toxin. Approximately 1.2 mg/kg diet is lethal. There are no data for dogs. Based on $BMDL_{05}$ for pigs, a corresponding effect threshold for dogs would be approximately 0.3-0.5 mg/kg feed.

Fish

Channel catfish were fed semi-purified diets contaminated with purified T-2 toxin for 8 weeks at levels of 0, 0.625, 1.25, 2.5 and 5.0 mg/kg feed. Fish fed 0.625 mg/kg feed and above had significantly reduced growth, and feed conversion was significantly reduced at the highest dietary level. Haematocrit values were significantly reduced and histopathological anomalies of the stomach, head and trunk kidney were observed at dietary levels of 1.25 mg/kg and

above (Manning et al. 2003a). In another channel cat fish study, fish fed T-2 toxin enriched diets (1.0 and 2.0 mg/kg diet) for 6 weeks were then challenged with the bacteria *Edwarsiella ictaluri*. Catfish fed both T-2 toxin levels had a higher mortality (84.1 and 99.3 %) than control fish (68.3 %) (Manning et al. 2005c).

Juvenile rainbow trout (1 g) fed graded levels (0, 1.0, 2.5, 5, 10 and 15 mg/kg diet) of crystalline T-2 for 4 months showed decreases in: growth, efficiency of feed use, haematocrit, blood haemoglobin concentrations and feed acceptance. A transitory, dose-dependent oedema was observed in fish fed T-2 at levels of 2.5 mg/kg feed and higher (with an assumed feeding rate of 5.5 % the dose is 0.14 mg/kg bw per day) (Poston et al. 1982).

In another study in which rainbow trout were fed 0.2 or 0.4 mg crystalline T-2 per kg diet for 12 months, examination of the liver showed no signs of neoplasia (Marasas et al. 1969).

To summarise for species relevant for fish farming, a NOAEL and a LOAEL for juvenile rainbow trout are derived at 1.0 and 2.5 mg/kg feed, respectively, based on reduced growth rate, feed efficiency, and haematocrit level. In channel catfish, a LOAEL at 0.63 mg/kg feed is derived based on reduced growth rate and feed efficiency.

Table 29. Critical effect (most sensitive endpoint) for T-2 toxin in feed for various animal species at concentrations where NOAEL or other available effect levels are derived/identified.

Animal	Critical effect	Concentration in total diet	Available effect level	Pivotal references
Pig	Immunological effects	0.17 mg/kg 0.5 mg/kg	Threshold LOAEL	EFSA (2011a); Rafai et al. (1995a, 1995b)
Broiler chicken	Lesions in oral cavity	0.5 mg/kg	LOAEL	EFSA (2011a)
Laying hen	Decreased egg production	2 mg/kg	LOAEL	EFSA (2011a)
Duck	Reduced body weight gain	0.2 mg/kg	LOAEL	EFSA (2011a)
Sheep	Reproductive adverse effect	Approxmately 0.1 mg/kg	NOAEL	Huszenicza et al. (2000)
Horse	Feed refusal and skin lesions	Approximately 0.7 mg/kg	Observed effect (one dose)	Juhasz et al. (1997)
Rabbit	Reduced body weight gain, intestinal necrosis	Approximately 2 mg/kg	NOAEL	EFSA (2011a)
Dog	No information found	0.3 – 0.5 mg/kg	Estimated threshold based on pig study	EFSA (2011a)
Cat	Mortality	Approximately 1.2 mg/kg	LOAEL	EFSA (2011a)
Rainbow trout	Reduced growth and feed efficiency	1 mg/kg 2.5 mg/kg	NOAEL LOAEL	EFSA (2011a) EFSA (2011a)

9.1.2.4 *Carry-over of T-2 and HT-2 to humans through animal-derived food products*

Trichothecenes, including T-2 and HT-2 toxins, are considered to be rapidly excreted from animals without accumulation in any particular organ (see Toxicokinetics). After a single administration of 0.25 mg/kg bw of T-2 toxin by gastric intubation to hens, a maximum level of 0.175 % of the administered dose was measured in eggs after 24 hours. The eggs contained only 0.1 and 0.025 % after 2 and 7 days respectively (Chi et al. 1978). Only the de-epoxide has been found in milk.

VKM supports the conclusions reached by SCF, JECFA and EFSA, that human exposure to trichothecenes through consumption of animal-derived food products is minimal in comparison to exposure via cereal-based food products (SCF 2001; JECFA 2001; EFSA 2011a). No information is available on the toxicokinetics and carry-over in Norwegian farmed fish species.

9.1.2.5 *Effects of T-2 and HT-2 in humans and animals relevant for human risk assessments*

The toxicity of the metabolites of T-2 toxin have not been studied in any detail, but, in general, most of the primary metabolites have similar or less toxic effects than T-2 toxin in different *in vitro* systems. The de-epoxy metabolites and probably the conjugated forms are considerably less toxic. Cats have been shown to be very sensitive towards T-2 toxin, probably due to limited glucuronide conjugation capacity.

Tests for genotoxicity are negative in microorganisms, while DNA strand breaks, unscheduled DNA synthesis, mutations, chromosomal aberrations and inhibition of gap-junctional intercellular communication have been reported from mammalian cells *in vitro*. No evidence of direct interaction with DNA has been reported, and hence, an approach with threshold for toxicological effects has been used in the risk assessment. No increase in tumours was found in a 71-week feeding study on mice, but an increase in pulmonary and hepatic adenomas in males was reported. However, a small increase in benign adenomas in one sex was not considered to be evidence of carcinogenicity. Therefore, T-2 toxin is not considered to be a potent carcinogen or to be directly genotoxic. Both SCF (2001) and JECFA (2001) based their evaluation on a short-term study of the haematological and immunological effects in pigs (Rafai et al. 1995a, b). 7-week-old pigs were fed 0.029 – 0.13 mg T-2 toxin/kg bw/day for 3 weeks. Decreased feed intake, decreased immunological response towards immunisation with horse globulin, decreased leukocyte counts, decreased mitogen response and decreased haemoglobin concentrations were observed in exposed animals. In addition, other effects, such as reduced weight gain and lower erythrocyte counts, were observed at the higher doses.

9.1.2.6 *Human hazard characterizations of T-2 and HT-2*

T-2 toxin has been evaluated by SCF (2001), JECFA (2001) and EFSA (2011a). All the assessments were based on an evaluation of a short-term study of the haematological and immunological effects in pigs given from 0.029 mg/kg bw per day (Rafai et al. 1995a, b). SCF and JECFA used a safety factor of 500, due to use of the lowest observed effect level (LOEL) and deficiencies in the available database. EFSA re-evaluated T-2 and HT-2 toxins and based their evaluations on the same study, but used a BMDL modelling of the data in the same study and an uncertainty factor of 100. EFSA proposed a TDI of 100 ng/kg bw/day. This TDI applies to the sum of T-2 toxin and HT-2 toxin. This TDI is used in this risk assessment.

9.1.3 Nivalenol

Nivalenol (NIV) occurs less frequently in Norwegian grains than DON and T-2 and HT-2 toxin (see occurrence, Chapter 6). The toxin has previously been assessed as a contaminant in food by the Nordic Council of Ministers (Eriksen and Alexander 1998) and the European Scientific Committee for Food (SCF 2000b). Nivalenol has been studied less than other trichothecenes like DON and T-2 toxin, but is generally considered to cause the same types of effects.

9.1.3.1 *Mode of action of nivalenol*

The mode of action of nivalenol is less studied than those of DON and T-2 toxin. Like DON and T-2 toxin, nivalenol binds to ribosomes and inhibits protein synthesis (reviews in Eriksen and Alexander 1998; SCF 2001b). Nivalenol has also been demonstrated to cause apoptosis in human Jurkat cells (Pestka et al. 2005; Nasri et al. 2006) and also *in vivo* in thymus, spleeen and Peyer´s patches of mice (Poapolathep et al. 2002). It also increases cytokine secretion in murine cells (Ouyang et al. 1995). As for other trichothecenes, the general toxicity and immunotoxicity/haemotoxicity were the critical effects in the SCF evaluation; the critical effects are considered to be reduced feed intake and growth, and impairment of the immune system (SCF 2000c).

9.1.3.2 *Toxicokinetics of nivalenol*

The toxicokinetics of nivalenol were reviewed by SCF (2000c) and Wu et al. (2010). Nivalenol is rapidly absorbed in pigs after oral exposure, and was present in plasma 20 min after feeding. Hedman et al. (1997) found that in pigs given 0.05 nivalenol/kg bw twice daily, 11-48 % was absorbed during the first 7.5 hours. The maximum plasma concentrations were low (3-6 µg/l), mostly occurring 2.5-4.5 hours after feeding. However, it was also found that absorption from the intestine was still occurring 16 hours after feeding.

Nivalenol was rapidly distributed to, and eliminated from, tissues in mice given a single oral dose of radioactively labelled nivalenol (Poapolathep et al. 2003). Unchanged nivalenol was transported to foetal or suckling mice via the placenta or milk. The levels in the foetus were comparable to maternal levels (Poapolathep et al. 2004).

Nivalenol was metabolised to de-epoxy nivalenol in rats (Onji et al. 1989), but not in mice (Poapolathep et al. 2003). No metabolites were found in pigs (Hedman et al. 1997). No *in vivo* studies of the metabolism in ruminants have been found, but nivalenol was transformed to de-epoxy nivalenol when incubated in ruminal fluids from cows (Hedman and Pettersson 1997).

Nivalenol is rapidly eliminated from animals. Nivalenol was mainly excreted in faeces in rodents and pigs, with no accumulation in any tissue (Wu et al. 2010).

9.1.3.3 *Effects of nivalenol in domestic animals*

The effects of nivalenol in farm animals have been reviewed (Eriksen and Pettersson 2004). The toxicity of nivalenol is far less studied than that of DON and T-2 toxin.

Pig

No effects on feed intake and body weight gain were found in pigs given 2.5 - 5 mg nivalenol/kg feed for 3 weeks, but feed intake and body weight gain were reduced in pigs given 5.8 mg nivalenol/kg feed from naturally maize. The feed in the experiments using naturally contaminated maize also contained zearalenone, and interactions with zearalenone or other unknown compounds present in the maize cannot be excluded (reviewed in Eriksen and Pettersson 2004).

Pathological examinations of pigs given 2.5 or 5 mg pure nivalenol/kg feed revealed haemorrhages, gastrointestinal erosions, and signs of nephropathy (pale kidneys with narrow cortex and dilated pelvis) in 4 and 5 out of 6 pigs in the group fed the diets containing 2.5 and 5 mg nivalenol/kg feed, respectively.

A statistically significant reduction in spleen cell numbers, as well as in CD4+ and CD8+ cells in the spleen, were also observed in the pigs given 5 mg nivalenol/kg feed (Hedman et al. 1997).

In sum, gastrointestinal haemorrhages and erosions have been reported in pigs given 2.5 mg/kg feed, which was the lowest dosage level used.

Poultry

The effects of nivalenol on poultry have been reviewed (Eriksen and Pettersson 2004). Few feeding experiments have been performed on poultry species: only two publications are available describing experiments in which broiler chickens were fed nivalenol and one publication is available for laying hens. Nivalenol concentrations of 0, 3, 6, and 12 mg/kg were used for chickens (12 birds per group fed for 20 days). Gizzard erosions were found in 33 % of the birds fed 12 mg/kg nivalenol and in 8 % of those fed 3 or 6 mg/kg of nivalenol (Hedman et al. 1995; Pettersson et al. 1995). Thus, gizzard erosions were reported from 3 mg/kg feed, and, furthermore, reduced liver and gizzard weights were shown in chickens fed nivalenol in concentrations from 3 mg/kg feed. Decreased weight gain and reduced feed conversion were shown in birds fed from 6 mg nivalenol/kg feed. In a study of laying hens, three of the five hens fed 1 mg nivalenol/kg diet had light brown coloured and fragile livers (Garaleviciene et al. 2002). The kidney of one bird fed 1 mg nivalenol/kg diet appeared to be enlarged and pale. Such pathological changes or erosions were not found in the control birds. Pathological examination revealed that 40-75 % of hens fed nivalenol supplemented diet at 3 and 5 mg/kg showed gizzard lesions, haemorrhages in the duodenum, and swollen cloaca and oviducts with immature eggs. The feed intake was reduced in the nivalenol-treated groups, but there were no apparent effects on body weight, egg production and egg quality. However, in addition to the pure nivalenol added to the diet, it also contained 66 µg nivalenol/kg and 100 µg DON/kg.

In sum, nivalenol has been shown to have various effects on poultry, depending on age and doses. In chicken, 3 mg/kg feed (lowest dosage level) produced gizzard erosions. In laying hens fed from 1 mg/kg feed, pale and fragile livers were observed, with more pronounced pathological changes at higher doses.

Ruminants

No *in vivo* information on nivalenol from ruminants was identified.

Rabbit

No data on the effects of nivalenol on rabbits were identified.

Dog and cat

No information on the effects of nivalenol on dogs and cats was identified.

Horse

No information on the effects of nivalenol on horses was identified.

Fish

No information on the effects of nivalenol on fish was identified.

Table 30. Critical effect (most sensitive endpoint) for nivalenol in feed at concentrations where NOAEL or other available effect levels are derived for various animal species.

Animal	Critical Effect	Concentration in feed	Available effect level	Pivotal reference
Pig	Gastrointestinal haemorrhages and erosions	2.5 mg/kg	LOAEL	Hedman et al. 1997
Broiler chicken	Gizzard erosions	3 mg/kg	LOAEL	Hedman et al. 1995
Laying hen	Pale and fragile livers	1 mg/kg	LOAEL	Garaleviciene et al. 2002
Ruminants	No information found			
Horse				
Rabbit				
Dog				
Cat				
Fish				

9.1.3.4 *Carry-over of nivalenol to humans from animal-derived food products*

No studies on the carry-over of nivalenol are available. Similarities with the more studied trichothecenes, DON and T-2 toxin, and the rapid excretion from animals suggest that it is reasonable to assume that human exposure through consumption of animal-derived food products is of little significance compared with exposure from consumption of grains.

9.1.3.5 *Effects of nivalenol in humans and animals relevant for human risk assessment*

SCF (2000c) established a temporary TDI (t-TDI) based on LOAEL of 0.7 mg kg bw/day from two long-term feeding studies on mice. Growth retardation and leukopenia were considered to be the critical effects, while other effects, including interauterine growth retardation, were seen at higher doses. Due to the limited data and a TDI based on the LOAEL, a large uncertainty factor of 1000 was used to derive a t-TDI of 0-0.7 μg/kg bw/day (SCF 2000c).

A few studies on the effects of nivalenol in rodents have been published since the opinion of SCF, mainly focusing on immunological effects, but other parameters, including pathology and clinical chemistry have also been included. Choi et al. (2000) found that 6 ppm nivalenol in drinking water for 2 and 4 weeks inhibited total and antigen-specific IgE production. The production of IL-4 was suppressed, while the production of IL-2 was enhanced (Choi et al. 2000). However, water consumption data are not provided and doses could not be estimated.

In mice given 0 – 8.87 mg nivalenol/kg bw, 3 days a week, for 4 weeks, alterations in immunological parameters were detected in mice given the highest dose. At this dose, even the activity of important biotransformation enzymes, such as some CYP 450 isoforms and glutathione transferase, were altered, but the protein expression of these enzymes was no different from that in controls (Gouze et al. 2007). The authors concluded that 1.8 mg nivalenol/kg bw was a NOEL in the study. Mice given 24 mg nivalenol/kg feed for 8 weeks

had an increased level of IgA in plasma compared with control mice. No effect was found in mice given 12 mg nivalenol/kg feed (Kubosaki et al. 2011). The authors concluded that nivalenol increased the level of IgA in plasma, but did not contribute to the deposition of the plasma IgA in the kidney glomeruli observed in IgA nephropathy.

Three papers describing experiments on rats given 0-100 ppm pure nivalenol/kg feed (0-6.9 mg/kg bw/day) for 90 days have been published. The number of white blood cells in the blood was decreased in females from the lowest dose group (0.4 mg/kg bw/day). Furthermore, an increase in NK cell activity against YAC-1 target cells was reported at this dose. Other haematological and serum biochemical parameters were affected at 1.5 mg nivalenol/kg bw/day, while histopathological changes were only observed at the highest dose group (Kubosaki et al. 2008; Takahashi et al. 2008). At this dose, histopathological alterations in the uterus, ovaries and oviducts were also reported (Sugita-Konishi et al. 2008).

The available data regarding the effects of nivalenol on most animal species are limited, but the current lowest LOAEL is from a sub-chronic feeding study on mice, where a LOAEL of 0.7 mg/kg bw/day was found. The critical endpoints were growth retardation and leukopenia. The current t-TDI set by SCF is based on this study, with a safety factor of 1000 due to the limited data and the use of a LOAEL.

9.1.3.6 *Human hazard characterizations of nivalenol*

SCF established a temporary TDI (t-TDI) for nivalenol of 0.7 μg/kg bw/day in 2000. Later studies have shown a lower LOAEL level in rodents than the one that the current t-TDI is based upon. However, a large uncertainty factor (1000) was used by the SCF when the t-TDI was established due to the very limited data. Several feeding studies have been published since then, but data gaps still remain. The t-TDI set by SCF in 2000 is used as a reference point in this risk assessment, but VKM is aware that EFSA is currently conducting a risk assessment of nivalenol in feed and food.

9.1.4 Other trichothecenes

A large number of other trichothecenes are occasionally reported from grains internationally, including diacetoxyscirpenol (DAS), monoacetoxyscirenol (MAS) and scirpentriol. These toxins are, however, found only infrequently in grains, and normally at low concentrations. Previous results from a screening of the occurrence of other trichothecenes in Norwegian grains also showed that the levels in these years were low (Langseth and Rundberget 1999). VKM therefore considers the risk from these toxins *per se* to human and animal health in Norway to be negligible.

9.2 Zearalenone

Zearalenone is commonly found in cereals worldwide. The highest concentrations are normally found in wheat bran and maize products. The effects of zearalenone have previously been assessed by the Nordic Council of Ministers (Eriksen and Alexander 1998), SCF (2001), JECFA (2001), and EFSA (2004b, 2011b). The effects of zearalenone have also been reviewed in Fink-Gremmels and Malekinejad (2007). The minor metabolite α-zearalanol is also used as a growth promoter in cattle. Such use is illegal in Norway and within the EU.

Zearalenone is a macrocyclic b-resorcyclic acid lactone. It is heat-stable up to 150°C. Degradation has been measured at higher temperatures or under alkaline conditions. Several

analogues have been shown in fungal cultures, but the natural occurrence of these analogues in feed and food has not been demonstrated (EFSA 2011b).

9.2.1 Mode of action of zearalenone

Zearalenone binds to, and activates, both the α– and β-oestrogen receptors (Fink-Gremmels and Malekinejad 2007). The metabolite α–zearalenol is more potent than the parent compound in receptor binding assays, while β-zearalenol is less potent. The minor metabolite α–zearalanol is even more potent than α–zearalenol.

9.2.2 Toxicokinetics of zearalenone

Zearalenone is rapidly absorbed in most species, and absorption has been estimated to be 80-85 % (JECFA 2000; Fink-Gremmels and Malekinejad 2007; EFSA 2011b). After oral administration to male rats, zearalenone is widely distributed to a range of tissues, including (in descending order of distribution) kidney > liver > adipose > lung > heart, spleen, muscle, brain, and testes (Shin et al. 2009), but without any apparent accumulation in any organ. Placental transfer has been demonstrated in both rats and pigs (Bernhoft et al. 2001; Däniche et al. 2007). The compound is extensively metabolised, primarily in the GI tract and the liver.

Zearalenone is enzymatically reduced to α- and β-zearalenol by 3α- and 3β-hydroxysteroid dehydrogenases (HSD). Smaller amounts are further reduced to form the corresponding zearalanols. In addition, minor concentrations of some hydroxylated metabolites have been found. Both the parental compound and the metabolites are glucoronide conjugated. The unconjugated zearalenols have different oestrogenic activities relative to the parent compound, in descending order of activity: α-zearalenol > zearalenone > β-zearalenol (reviewed in EFSA 2011b). The proportion of the different metabolites formed varies between species, and a larger proportion of α-zearalenol is formed in pigs than in other species investigated, giving a potential pharmacokinetical explanation for the sensitivity of this species to zearalenone (EFSA 2011b). Very limited human data indicate that more α-zearalenol than β-zearalenol is formed in humans (Mirocha et al. 1981).

Zearalenone is excreted through urine and bile, and the proportion seems to be species-dependent. Zearalenone and metabolites are mainly excreted in the faeces in most species, but urine is the main route of excretion in sheep. Zearalenone undergoes an extensive enterohepatic recycling, as demonstrated by the plasma half-time reduction from 87 to 3 hours in pigs when the bile was cannulated (Biehl et al. 1993). Excretion through milk has been found in pigs (Dänicke et al. 2007). Only conjugated forms of zearalenone and metabolites were found in milk from a cow given 6 g of zearalenone over one day (Prelusky et al. 1990). No information was found on the toxicokinetics and carry-over in Norwegian farmed fish species.

9.2.3 Effects of zearalenone in domestic animals

Zearalenone is a potent agonist of the oestrogen receptors, and the clinical symptoms of exposure to zearalenone are primarily oestrogenic effects. The mycotoxin has low acute toxicity, with oral LD50 values >4000 mg/kg bw (EFSA 2004b).

Pig

Pig is a particularly sensitive species for zearalenone. The effects in pigs are typically oestrogenic, such as effects on cycling time, hormone levels, swelling of cervix, increased uterus weights etc., as well as embryonic resorption and reduced foetal weight and smaller litter size. The effects are dependent on toxin source, potential interactions with co-occurring fungal metabolites, and pig age. Young pre-pubertal female pigs seem to be particularly sensitive to zearalenone.

The majority of studies have been conducted with rather high concentrations of pure crystalline zearalenone mixed into the feed at doses that are not regarded as realistic exposure scenarios (EFSA 2004b). However, experiments have also been conducted using lower exposure levels, and naturally contaminated grain. Döll et al. (2003) conducted a study on young female pigs fed diets with zearalenone from naturally contaminated maize at 0.01, 0.06, 0.15, 0.22 and 0.42 mg/kg diet for 5 weeks, with 20 pigs per group. The study resulted in an apparent dose-related increase in the number of pigs with swollen and reddened vulva and cervix, but the effects were only statistically significant at the highest dosage level (LOEL). Increases in the absolute and relative weights of the uterus were also found at this level. The maize was co-contaminated with 0.2 – 3.9 mg DON/kg feed and the body weight was significantly decreased at highest dosage level. The DON contamination may have influenced the results, but as the zearalenone effects are rather specific, the dose response relationship found is considered appropriate for using for hazard characterization. EFSA (2011b) has based the human TDI in the hazard characterization of zearalenone on this pig study, where a NOEL at 0.01 mg/kg bw per day, corresponding to 0.22 mg/kg diet, was derived.

Few studies on the effects of zearalenone on boars have been conducted, but they seem to be rather resistant.

To summarise for pigs, a NOAEL for zearalenone in females at 0.2 mg/kg diet has been derived.

Poultry

A few feeding studies on the effects of zearalenone in the diets of poultry have been published. The feeding experiments have generally exposed the birds to unrealistically high concentrations of zearalenone in the feed, with concentrations up to 800 mg/kg.

In broilers given 50 – 800 mg /kg feed for 7 days, a dose-related increase in oviduct weights and in ovarian cystic development have been reported, but only at the higher concentrations (Chi et al. 1980a, 1980b).

No effects on laying performance or reproduction were reported from hens and cocks given 10-800 mg zearalenone/kg feed (Allen et al. 1981b).

EFSA concluded in their evaluation that poultry seem to be quite resistant to zearalenone, as symptoms of hormonal effects were observed only at very high doses that are unlikely to occur under normal practical feeding conditions (EFSA 2004b).

Ruminants

Few studies of the effects of zearalenone in ruminants have been published, but the available information indicates that sheep are rather sensitive. In ewes fed 1.5, 3, 6, 12 and 24 mg

zearalenone/animal (0.03-0.45 mg/kg bw per day, derived to be approximately 0.9-14.4 mg/kg diet) for 10 days from day 7 in oestrus, a reduced ovulation rate was observed at the lowest dosage (Smith et al. 1990). A range of other oestrogenic effects were observed at the higher doses. No effects were seen in ewes given the same doses from day 5 after mating. In heifers fed 250 mg crystalline zearalenone (approximately 50 mg/kg diet) per day a decreased conception rate over 3 oestrus cycles was found (Weaver et al. 1986). No deviations in the oestrus cycle and no pathological and histological effects on the reproductive organs were observed when contaminated oats with a concentration of zearalenone at 1.25 mg/kg diet were fed to heifers (Moeser 2001).

To summarise, sheep may be rather sensitive to zearalenone, particularly during oestrus, and a reduced ovulation rate was shown at approximately 0.9 mg/kg feed (LOAEL). Heifers seem to be far less sensitive to adverse effects associated with exposure to zearalenone.

Horse

Mares were given daily oral doses of 7 mg purified zearalenone per animal (approximately 1 mg/kg total diet), starting 10 days after ovulation and continuing until the next ovulation (Juhasz et al. 2001). No effects on interovulatory intervals, luteal or follicular phases of the oestrus cycle, plasma progesterone, follicular morphology or uterine oedema were observed.

To summarise, no effects have been showed in horses exposed to approximately 1 mg/kg diet.

Rabbit

Female, 4 month-old rabbits, were given zearalenone in their diet for 18 days, at concentrations of 0, 0.5 or 1 mg/kg feed (Abdelhaid et al. 1992). The results showed a dose-related increase in body weight gain, food and water consumption, haemoglobin percentage, packed cell volume, and serum concentrations of calcium, phosphorus and vitamin C. Another study by the same authors with 8-month-old female rabbits fed 0, 1 or 4 mg/kg feed, provided rather contrasting results with these higher dosages, as dose-related decreases in these parameters were observed.

To summarise, effects in rabbits have been measured at 0.5 mg/kg diet (identified LOEL).

Dog and cat

Adult female dogs in anoestrus phase were orally treated with zearalenone at 0, 0.025 or 0.050 mg/kg bw per day for 100 days, and the ultrastructure of the ovaries was studied, together with blood biochemical and haematological variables (Gajecka et al. 2008). The dosage levels correspond to approximately 1.3 and 2.5 mg/kg diet. Ultrastructural changes in the granular layer of ovarian follicles were observed at highest dosage. The clinical-biochemical and haematological profiles showed some changes in both zearalenone groups, indicating metabolic disorder with anaemia and hyperbilirubinemia.

No studies on the effects of zearalenone on cats have been found.

To summarise for dogs, a LOAEL is identified at approximately 1.3 mg/kg diet, based on metabolic changes with anaemia and hyperbilirubinemia.

Fish

Zearalenone and its metabolites cause disruption of vitellogenin (Vtg) and zona radiate (Zr) proteins, and mRNA expression in *in vitro* and *in vivo* in rainbow trout studies (Arukwe et al. 1999; Celius et al. 1999, 2000; Wozny et al. 2008), as well as *in vivo* studies on Atlantic salmon with *i.p.* exposure (Arukwe et al. 1999). The zearalenone metabolite, α-zearalenol is the most potent oestrogenic disrupter in both rainbow trout and Atlantic salmon (Arukwe et al. 1999; Celius et al. 1999). Most studies on zearalenone in finfish species are based on *in vitro* studies or exposure via *i.p.* injection and hence cannot be directly used for assessment of the effect levels of dietary exposures. One oral study on Atlantic salmon did not show adverse effects on feed intake, specific growth rate, clinical plasma parameters, or histology in adult fish (405 g) fed diets containing up to 0.77 mg zearalenone/kg feed for 15 weeks. Possible oestrogenic disruption was not reported in this study (Döll et al. 2010).

To summarise, reports of oestrogenic effect of oral zearalenone exposure in fish are lacking, but no other health effects have been shown in Atlantic salmon fed up to 0.8 mg/kg feed.

Table 31. Critical effect (most sensitive endpoint) for zearalenone in feed at concentrations where NOAEL or other available effect levels are derived/identified for various species.

Animal	Critical Effect	Concentration in feed	Available effect level	Pivotal reference
Pig	Oestrogenic effect	0.2 mg/kg	NOAEL	Döll et al. (2003)
Poultry	No symptoms	>50 mg/kg	NOAEL	EFSA (2011b)
Sheep	Oestrogenic effect	Approximately 0.9 mg/kg	LOAEL	Smith et al. (1990)
Horse	Oestrogenic effect	Approximately 1 mg/kg	No effect (one dosage level)	Juhasz et al. (2001)
Rabbit	Anabolic and haematological effects	0.5 mg/kg	LOAEL	Abdelhamid et al. (1992)
Dog	Metabolic changes	1.3 mg/kg	LOAEL	Gajecka et al. (2008)
Atlantic salmon	Clinical effect	0.77 mg/kg	NOAEL	Döll et al. (2010)

9.2.4 Carry-over of zearalenone to humans through animal-derived food products

Very low levels of zearalenone have been found in edible tissues of farm animals (see 9.2.2) in experimental studies. Only very low levels of zearalenone and its metabolites were found in milk from cows given a very high dose of zearalenone (6 g for one day). EFSA summarised and evaluated the studies of the transfer of zearalenone from animals to humans in the food chain and concluded that there is only limited deposition of zearalenone in meat and that the transfer to milk and eggs is low. No information was found on the toxicokinetics and carry-over in farmed fish species.

VKM supports the conclusions reached by JECFA and EFSA, that human exposure to zearalenone through consumption of animal-derived food products is very limited (EFSA 2011b; JECFA 2001).

9.2.5 Effects of zearalenone in humans and animals relevant for human risk assessments

There is no substantial information about the effects of zearalenone in humans, although some suspected associations between zearalenone and hormonal effects, such as premature breast development, have been reported (reviewed in EFSA 2011b). The human risk characterisation is therefore based on the oestrogenic effects in female pigs, considered to be the most sensitive species and sex. This may be partly due to the relatively larger proportion of zearalenone that is metabolised to the more active α–zearalenol in pigs than in most other species.

9.2.6 Human hazard characterizations (TDI) of zearalenone

The human risk assessment is based on the oestrogenic effects of zearalenone in pigs, which is regarded as the most sensitive species. EFSA concluded that zearalenone is, at most, a weak carcinogen and that the TDI could be established based on the NOEL of 10 µg/kg bw/day for oestrogenic effects in female pigs. Using an uncertainty factor of 10 for inter-individual differences and 4 for inter-species differences in toxicokinetics, EFSA derived a TDI of 0.25 µg/kg bw/day (EFSA 2011b). This TDI is used in the current risk assessment.

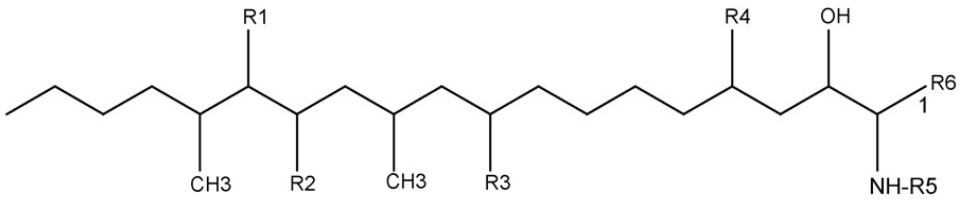
9.3 Fumonisins

Fumonisins were first identified and isolated in 1988 and 28 fumonisin analogues are known to date (Gelderblom et al. 1988; Nelson et al. 1993; Rheeder et al. 2002). Fumonisins are divided into four groups: serial A, B, C and G (Figure 15). In naturally contaminated food and feed maize, fumonisin B_1 (FB_1) accounts for 70-80 % of the total fumonisin load. The present regulation of fumonisins in Norway applies to the sum of fumonisins B_1 and B_2 and this risk assessment will focus on these two. Furthermore, fumonisins are only found in high levels in maize, a food commodity that is generally imported and only consumed in low amounts by Norwegians. Occurrence data from maize on the Norwegian market are very limited. The main focus in this assessment is therefore on fumonisins in imported feed materials.

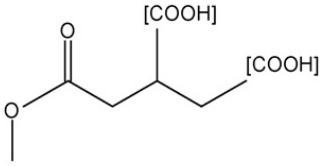
Fumonisins are very polar, water-soluble compounds. They have a long chain-aminopentol backbone (polyhydroxyl alkylamine), esterified with two carbon acids, i.e. tricarballylic acid (TCA).

The molecular formulae and weights of fumonisin B_1 are $C_{34}H_{59}NO_{15}$ and 721.8 g/mol, and for fumonisin B_2 they are $C_{34}H_{59}NO_{14}$ and 705.8 g/mol.

Fumonisins are possibly carcinogenic to humans and animals. The first assessment of maize products concluded that fumonisin intake by high consumers was more than three orders of magnitude lower than the NOEL in animal studies (Kuiper-Goodman et al. 1996). An assessment by the Nordic Council of Ministers came to similar conclusions (Eriksen and Alexander 1998). However, a risk assessment for a sensitive sub-population, patients with coeliac disease, revealed no margin of safety (De Nijs 1998). Depending on the end-point of the toxicity model chosen and the respective NOEL or LOEL, as well as the safety factor considered, ADI or TDI values in the range 0.16 to 8 µg/kg body weight were proposed (De Nijs 1998; Marasas 1997; Commission of the European Union 1998). A monograph on fumonisin B_1 was published in 2001 (JECFA 2001).



Tricarballylic acid (TCA)



Toxin	R1	R2	R3	R4	R5	R6
Group A						
FA1	TCA	TCA	OH	OH	COCH3	CH_3
FA2	TCA	TCA	H	H	COCH3	CH_3
FA3	TCA	TCA	OH	OH	COCH3	CH_3
Group B						
FB1	TCA	TCA	OH	OH	H	CH_3
FB2	TCA	TCA	H	OH	H	CH_3
FB3	TCA	TCA	OH	H	H	CH_3
FB4	TCA	TCA	H	H	H	CH_3
Group C						
FC1	TCA	TCA	OH	OH	H	H
FC3	TCA	TCA	OH	H	H	H
FC4	TCA	TCA	H	H	H	H

Figure 15. Chemical structures of main fumonisin analogues (Krska et al. 2007).

9.3.1 Mode of action of fumonisin

Fumonisin toxicity is based on the structural similarity to the sphingoid bases, sphingosine and sphinganine. Fumonisins are inhibitors of sphinganine (sphingosine) N-acyltransferase (ceramide synthase), a key enzyme in lipid metabolism, thereby resulting in a disruption of this pathway. This enzyme catalyzes the acylation of sphinganine in the biosynthesis of sphingolipids and the deacylation of dietary sphingosine and the sphingosine that is released by degradation of complex sphingolipids (ceramid, sphingomyelin and glycosphingolipide) (Wang et al. 1992). Sphingolipids are basically important for the membrane and lipoprotein structure and for cell regulation and communication (second messenger for growth factors). As a consequence of the disruption, the levels of many bioactive intermediates, including free sphingoid bases, are elevated, while the concentrations of complex sphingolipids, for example, are reduced. Free sphingoid bases are toxic to most cells, affecting cell proliferation and inducing apoptosis or necrotic cell death (Riley et al. 1996; Stevens et al. 1990). The accumulation of sphinganine is associated with hepatotoxic and nephrotoxic effects (Riley et al. 1996). Complex sphingolipids are important for cell growth regulation and cell-cell interactions. Accumulation of free sphingoid bases in serum and urine is a useful biomarker for exposure to fumonisins. In mutation assays, fumonisins have been found to be inactive, although they induce DNA-damage such as micronuclei *in vitro* and *in vivo*.

The toxicity of hydrolyzed fumonisin B_1 (HFB_1), which occurs as a result of some corn possessing procedures such as tortilla manufacturing, has not been clarified, but it may also be hepatotoxic and nephrotoxic (Voss et al. 1998).

9.3.2 Toxicokinetics of fumonisin

The oral bioavailability of fumonisin B_1 (FB_1) is lower than 5 %. In a peroral study in rats fed with 7.5 mg/kg radio-labelled FB_1, almost 100 % of the dose was recovered unmetabolised in the faeces (Shephard et al. 1992). FB_1 is rapidly distributed and eliminated after uptake (Martinez-Larranaga et al. 1999). Little FB_1 is detected in plasma and tissues after oral administration, and is distributed to most tissues, with highest levels in liver and kidney. HFB_1 is better absorbed than FB_1, although biliary excretion has been shown to be similar (Dantzer et al. 1999).

In vervet monkeys (*Cercopithecus aethiops*), FB_1 plasma half-life was 3-4 times longer than in other species, though bioavailability was low (Shephard et al. 1994). The compound was mainly excreted un-metabolised or hydrolyzed in the faeces (76 %). Similar results have been obtained from animals given FB_2 (Shephard and Snijman 1999).

FB_1 toxicokinetics have been assessed in several production animal species. In laying hens, plasma half-life was short and the bioavailability was only 0.5 %, with 98 % of the dose provided being recovered in the faeces after 24 h (Vudathala et al. 1994). In pigs and weaned piglets, the bioavailability was 4 %, the rate of excretion via faeces was 76 %, and an extensive enterohepatic circulation was detected (Prelusky et al. 1994; Dilkin et al. 2010). Feeding experiments showed that a withdrawal period of at least 2 weeks was required to eliminate the toxin from the organs (Martinez-Larranaga et al. 1999). Absorption increased through repeated FB_1 applications to 4 % in pigs (Szabo-Fodor et al. 2008). In dairy cows, FB_1 or known metabolites were not found in plasma after per os administration. Ruminal flora reduced FB_1 by up to 18 % in *in vitro* experiments, but hydrolysed metabolites such as aminopentol were not detected (Caloni et al. 2000). In goats, only 0.5 % of the ingested toxin was excreted unmetabolised in the faeces (Gurung et al. 1998).

In a feeding study with rainbow trout using the Oregon test diet, between 12 and 80 % of FB_1 was absorbed and biologically active, increasing free sphinganine in the liver, kidney and serum (Meredith et al. 1998).

Aminopentol (AP1) is the backbone and main hydrolysis product of FB_1 and is formed through complete hydrolysis of the ester-linked tricarballylic acids of FB_1. Aminopentol is more hydrophobic than FB_1 and is more effectively absorbed. A further derivative, N-palmitoyl-aminopentol, is tenfold more toxic than FB_1 (Humpf et al. 1998).

No metabolites were detected in *in vitro* phase I metabolism studies of FB_1 using rat hepatocytes (Cawood et al. 1994) or bovine liver microsomes (Spotti et al. 2001). However, FB_1 can act as substrate in phase II metabolism, and be glucuronidated and excreted via the bile (approximately 1.4 % of the dose) (Dantzer et al. 1999).

9.3.3 Effects of fumonisin in domestic animals

There are considerable species differences in sensitivity to fumonisins. Horses and pigs are considered to be the most sensitive species, developing species-specific clinical syndromes such as equine leukoencephalomalacia and porcine pulmonary oedema.

Pig

Fumonisin toxicosis in pigs is characterised by pulmonary, cardiovascular and hepatic symptoms. Moreover, hyperplastic oesaphagitis, gastric ulceration, hypertrophy of the heart and hypertrophy of the pulmonary arteries have been described (EFSA 2005). The pulmonary

effects seem to be critical at low doses of fumonisins. Lethal pulmonary oedema and hydrothorax have been observed in pigs exposed to feed containing more than 12 mg FB_1/kg (Haschek et al. 2001). Levels of FB_1 as low as 1 mg FB_1/kg feed over an 8-week period have produced proliferation of connective tissues in the lungs without being accompanied by clinical signs (1 of 4 pigs). These changes increased dose-dependently at 5 and 10 mg/kg (Zomborszky-Kovacs et al. 2002). An increase in the sphinganine:sphingosine (Sa:So) ratio in serum and tissues is used diagnostically for fumonisin exposure and for assessing the onset of adverse effects. Increased Sa:So ratios have been found in pigs fed 5 mg FB_1/kg feed and above (Riley et al. 1993; Zomborszky-Kovacs et al. 2002), but not in pigs fed 1 mg FB_1/kg (Rotter et al. 1996; Zomborszky-Kovacs et al. 2002). However, Rotter et al. (1996) fed male and female 6-week old piglets pure FB_1 at concentrations of 0, 0.1, 1 and 10 mg/kg diet for 8 weeks, and observed a significant reduction in the average daily gain at 1 and 10 mg FB_1/kg (8 and 11 %, respectively) of the male pigs during weeks 5-8. Furthermore, Rotter et al. (1997) fed growing-finishing pigs pure FB_1 at concentrations of 0, 0.11, 0.33 and 1.0 mg/kg feed for 11 weeks until market weight, and suggested increased variabilities in feed consumption and carcass quality at 1.0 mg/kg diet.

In sum, there are indications of lung connective tissue proliferation, performance changes and variable carcass quality in pigs fed 1 mg FB_1/kg feed, which indicate a NOEL at 0.33 mg/kg diet. However, more obvious adverse connective tissue proliferation in the lungs and an increased plasma Sa:So ratio have been shown at 5 mg/kg diet and above, corresponding to a NOAEL of 1 mg/kg diet.

Poultry

Broiler chicken, turkeys and ducklings have been regarded as relatively resistant to FB_1 compared with other animal species. However, young poultry are more sensitive to the toxin than adults. From the assessment of fumonisin effects in birds (EFSA 2005), a LOAEL for broiler chickens of 20 mg FB_1/kg feed was derived. This conclusion was based on the increased sphinganine concentration and liver Sa:So ratio reported by Henry et al. (2000), who fed broiler chickens purified FB_1 at 0, 20, 40 and 80 mg/kg feed for 21 days from day 1. More recent results may indicate a higher sensitivity, at least in ducks. In ducks force-fed with a diet containing 5, 10 and 20 mg FB_1 + FB_2 for 12 days signs of toxicity, including mortality, were obtained at 20 mg/kg, whereas an increased Sa/So ratio (serum, liver, kidney) was observed at 5 mg/kg (Tardieu et al. 2009). Based on these results, we identified a LOAEL of 5 mg/kg diet for ducks.

To conclude, for broiler chicken a LOAEL of 20 mg FB_1/kg diet is derived and for ducks we identify a LOAEL of 5 mg/kg diet.

Ruminants

In contrast with many other mycotoxins, FB_1 is incompletely degraded by the rumen flora, but signs of intoxications have only been reported in cattle, sheep and goats exposed to rather high concentrations of fumonisin in their feed (EFSA 2005). Feeder calves were fed diets containing 26 or 105 mg/FB_1/kg for 31 days (Osweiler et al. 1993). No effects were shown at the lowest dosage, but clinical chemical and histological changes indicative of toxicological liver effects were shown at the highest dosage level. Steer were fed a diet containing, on average, 94 mg FB_1/kg for 253 days. This dosage increased serum aspartate transaminase (AST) and gamma-GT activities, and induced histological evidence of mild hepatocellular injury and bilary epithelial hyperplasia (Baker and Rottinghouse 1999). In cows fed with a

diet containing 75 mg FB_1/kg for 14 days, a decrease in feed intake and milk production, as well as increased serum cholesterol, were observed (Richard et al. 1996). In goats, an increased Sa:So ratio in the liver was found following exposure to a concentration of 95 mg FB_1/kg feed (Gurung et al. 1998).

In sum, adverse effects occur in ruminants fed with FB_1 at 75 mg/kg in the feed or more. No effects were observed in feeder calves fed 26 mg/kg feed (identified NOAEL).

Horse

Equids (horses, donkeys and mules), are sensitive to fumonisins and can develop severe equine leukoencephalomalacia (ELEM). The evaluation of many cases of ELEM in the USA indicated that consumption of feed containing more than 10 mg FB_1/kg is associated with an increased risk for horses developing ELEM, whereas at concentrations below 6 mg/kg diet no increased risk is found (Constable et al. 2000; Ross et al. 1994). It has been hypothesised that ELEM is a result of cerebral oedema due to an inability to reduce the blood flow to the brain when the horse lowers its head to eat and drink.

Rabbit

Rabbits seem to be relatively sensitive to fumonisins. Pregnant rabbits were given FB_1 at 0.1-1.75 mg/kg bw per day orally by gavage during gestation days 3-19 (LaBorde et al. 1997). Maternal toxicity was observed at 0.25 mg/kg bw per day, and foetal toxicity occurred at the lowest dose tested (0.1 mg/kg bw, approximately 2 mg/kg diet, which we identify as LOAEL) but no evidence of teratogenic effects was shown.

Dog and cat

Data on effects of FB_1 and FB_2 in dogs and cats have not been identified.

Fish

The effects of dietary FB_1 have been most studied for finfish species such as common carp (*Cyprinus carpio L*), channel catfish (*Ictalurus punctatus*), and Nile tilapia (*Oreochromis niloticus*). In channel catfish, FB_1 was shown to cause liver damage, reduced weight and increased mortality (Lumlertdacha et al. 1995; Yildirim et al. 2000). FB_1 also had an immunosuppressive effect on catfish and reduced their resistance to bacterial infections (Lumlertdacha and Lovell 1995). FB_1 is a known neurotoxin, causing brain pathology (apoptosis) in common carp (*Cyprinus carpio L*) (Kovacic et al. 2009; Lumlertdacha et al. 1995). The EFSA opinion on fumonisins as undesirable substances in animal feed (EFSA 2005) addressed the adverse effects of FB_1 on Nile tilapia, channel catfish and carp. The EFSA opinion concluded that the data from catfish and Nile tilapia suggested a NOAEL of 20 mg/kg feed, whereas in carp a LOAEL of 10 mg/kg feed was described (EFSA 2005). Further dietary studies with carp also indicated that signs of FB_1-induced toxicity can be observed following exposure to 10 mg FB_1/kg feed (Kovacic et al. 2009).

Few studies have been performed on salmonid species. Although FB_1 has been associated with tumour formation, FB1 itself is not an initiator of tumours in rainbow trout (Carlson et al. 2001). Co-exposure to FB_1 with the tumor initiator aflatoxin B1 causes the promotion of liver tumours. FB_1-related promotional activity in aflatoxin B1-initiated liver tumours was

suggested to be mediated by FB_1-induced alterations in sphingolipid signalling pathways (Carlson et al. 2001). In an 8-week dietary study, juvenile rainbow trout (>1 g) were fed purified FB_1-enriched diets at levels of 0, 3.2, 23, and 104 mg FB_1/mg feed (feed also contained traces of FB_2; 1.1 ± 0.1 %). Liver tumour promotion and free sphingosine production after initiation with aflatoxin B1 were not observed at dietary levels of 3 mg/kg, but dietary levels of 23 mg/kg caused an increase in tumours and free sphingoid presence in the liver after aflatoxin B1-initiated hepatocarcinogenicity (Carlson et al. 2001). In two rainbow trout (15-25 g) studies, fish were fed purified FB_1-enriched diets at levels of 0, 5, 25, and 100 mg/kg for 7 days or 0, 100, 250, and 500 mg/kg for 5 days.

Dietary levels of 25 mg/kg for 1 week resulted in a significant increase in free sphinganine andsphingosine, as well as in increase in free spinganine/free sphingosine ration, in the liver, kidney and serum, and exposure levels of 100 mg/kg resulted in an increase of free sphinganine that was comparable in magnitude to that associated with liver toxicity in mammals (Meredith et al. 1998).

In sum, based on the disruption of sphingolipids, a LOEL for rainbow trout could be set to 25 mg/kg (with an assumed feeding rate of 3.5 %/bw/day, dose of 0.87 mg/kg bw/day), while levels of 100 mg/kg (with an assumed feeding rate of 3.5 % of bw/day, dose of 3.5 mg/kg bw/day) could be set as the LOAEL. Feed levels of 3 mg/kg (0.15 mg/kg bw/day) feed can be set as the NOEL.

Table 32. Critical effect (most sensitive endpoint) for fumonisins in feed at concentrations relevant for setting NOAEL or other available effect levels for various species.

Animal	Critical Effect	Concentration in feed	Available effect level	Pivotal references
Pig	Lung connective tissue proliferation, performance changes and variable carcass quality Increased plasma Sa:So ratio	0.33 mg/kg 1 mg/kg	NOAEL NOAEL	Zomborszky-Kovacs et al. 2002
Broiler chicken	Increased sphinganine concentration and liver Sa:So ratio	20 mg/kg	LOAEL	EFSA 2005
Ducks	Increased Sa/So ratio (serum, liver, kidney)	5 mg/kg	LOAEL	Tardieu et al. 2009
Cows	Decreased feed intake and milk production	75 mg/kg	Observed effect level (one dosage level)	Richard et al. 1996; EFSA 2005
Feeder calves	Liver effects	26 mg/kg	NOAEL	Osweiler et al. 1993
Horse	Equine leukoencephalomalacia (ELEM)	6 mg/kg	Threshold	Constable et al. 2000; Ross et al. 1994
Rabbit	Foetal toxicity	Approximately 2 mg/kg	LOAEL	LaBorde et al. 1997
Dog	No information found			
Cat	No information found			
Rainbow trout	Increased sphingosine production	25 mg/kg 3 mg/kg	LOEL NOEL	Meredith et al. 1998; Carlson et al. 2001

9.3.4 Carry-over of fumonisins to humans through animal-derived food products

Humans are not exposed to notable amounts of FB_1 via animal-derived food, except for livers and kidneys from pigs, cattle, and poultry (Aga 2005; Voss et al. 2007; Szabo-Fodor et al. 2008). The carry-over rate of FB_1 appears to be low, as shown by a study on suckling pigs from sows fed with fumonisin in the feed (Becker et al. 1995) and milk from cows fed with up to 125 mg toxin (Hammer et al. 1996). In US milk samples, a maximum concentration of 0.0013 mg/L was detected (Maragos and Richard 1994). Fumonisin transfer into chicken eggs has not been demonstrated (Vudathala et al. 1994; Prelusky et al. 1996). JECFA concluded that the contributions of other commodities to the intake of fumonisins are too low and too variable to have a significant contribution to overall long-term exposure (JECFA 2001). Little information exists on carry-over of fumonisins to the edible parts of fish.

VKM agrees with the conclusions from JECFA, and considers that human exposure to fumonisins from animal-derived food products is negligible.

9.3.5 Effects of fumonisins in humans and animals relevant for human risk assessments

The effects of fumonisins on human health are uncertain (Voss et al. 2007). However, fumonisins are suspected risk factors for oesophageal (Marasas 2001) and liver (Ueno et al. 1997) cancers, neural tube defects (Gelineau-van Waes et al. 2005; Missmer et al. 2006), and cardiovascular problems (Fincham et al. 1992) in populations consuming relatively large quantities of food made with contaminated maize. According to the IARC (International Agency for Research on Cancer), fumonisins are classified as possible human carcinogens.

In animals, fumonisins are known to be hepatotoxic and nephrotoxic in all animal species, although with considerable species differences regarding sensitivity to the toxin. Although FB_1 is poorly absorbed and metabolised in the intestine, there is considerable evidence that the toxin also causes intestinal disturbances like abdominal pain and diarrhoea (Bouhet and Oswald 2007). Feeding experiments in rats with FB_1 and FB_2 led to the production of neoplastic nodules, hepatocellular carcinomas, cholangiocellular carcinomas and forestomach papillomas and carcinomas (Howard et al. 2001). In non-human primates, administration of FB_1 has resulted in increased serum concentrations of sphingoid bases, cholesterol, and enzymes indicative of liver functions, and these remained elevated for several weeks after dosing (van der Westhuizen et al. 2001).

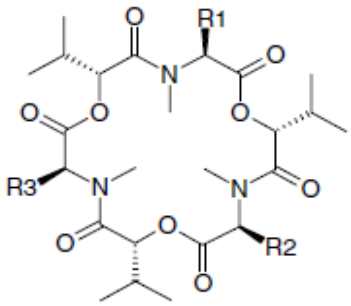
9.3.6 Human hazard characterizations of fumonisins

The JECFA committee allocated a group provisional maximum tolerable daily intake (PMTDI) for the fumonisins FB_1, FB_2, and FB_3, alone or in combination, of 2 µg/kg body weight per day. The PMTDI was obtained by model calculations on the basis of a NOAEL of 0.22 mg/kg body weight per day observed for the development of renal tumours (adenomas and carcinomas) in male Fischer 344 rats in a study conducted for the National Toxicology Program in the USA (1999) and the application of a safety factor of 100 (JECFA 2001).

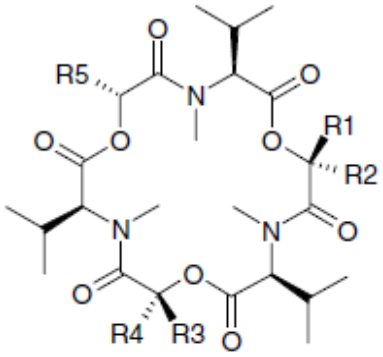
9.4 Enniatins

Enniatins are produced by *Fusarium avenaceum*, the most prevalent species of mycotoxin-producing fungus on grains in northern countries.

Enniatins are six-membered cyclic depsipeptides (Figure 16), which are commonly composed of three D-2-hydroxyisovaleric acid (Hiv) residues linked alternately to three N-methyl-L-amino acid residues (N-Me-R), which are used for distinguishing the individual enniatins. 28 homologues have been identified (Firakova et al. 2007), of which enniatin B, a $(N\text{-Me-Val-Hiv})_3$ – molecule with the molecular formula and weight $C_{36}H_{63}N_3O_9$ and 639.83 g/mol, is the most prevalent.



Enniatin	R1	R2	R3
A	$CH(CH_3)CH_2CH_3$	$CH(CH_3)CH_2CH_3$	$CH(CH_3)CH_2CH_3$
A1	$CH(CH_3)CH_2CH_3$	$CH(CH_3)CH_2CH_3$	$CH(CH_3)_2$
A2	$CH(CH_3)CH_2CH_3$	$CH(CH_3)CH_2CH_3$	$CH_2CH(CH_3)_2$
B	$CH(CH_3)_2$	$CH(CH_3)_2$	$CH(CH_3)_2$
B1	$CH(CH_3)_2$	$CH(CH_3)_2$	$CH(CH_3)CH_2CH_3$
B4	$CH(CH_3)_2$	$CH_2CH(CH_3)_2$	$CH(CH_3)_2$
C	$CH_2CH(CH_3)_2$	$CH_2CH(CH_3)_2$	$CH_2CH(CH_3)_2$
D	$CH(CH_3)_2$	$CH(CH_3)_2$	$CH_2CH(CH_3)_2$
E	$CH(CH_3)_2$	$CH_2CH(CH_3)_2$	$CH(CH_3)CH_2CH_3$
	$CH(CH_3)_2$	$CH(CH_3)CH_2CH_3$	$CH_2CH(CH_3)_2$
F	$CH_2CH(CH_3)_2$	$CH(CH_3)CH_2CH_3$	$CH(CH_3)CH_2CH_3$
G	$CH(CH_3)_2$	$CH_2CH(CH_3)_2$	$CH_2CH(CH_3)_2$



Enniatin	R1	R2	R3	R4	R5
H	H	$CH(CH_3)_2$	H	$CH(CH_3)_2$	$CH(CH_3)CH_2CH_3$
I	H	$CH(CH_3)_2$	$CH(CH_3)CH_2CH_3$	H	$CH(CH_3)CH_2CH_3$
L	$CH(CH_3)_2$	H	$CH(CH_3)_2$	H	$C(CH_3)OHCH_2CH_3$
M1	$CH(CH_3)_2$	H	$CH(CH_3)CH_2CH_3$	H	$C(CH_3)OHCH_2CH_3$
M2	$CH(CH_3)CH_2CH_3$	H	$CH(CH_3)_2$	H	$C(CH_3)OHCH_2CH_3$
N	$CH(CH_3)CH_2CH_3$	H	$CH(CH_3)CH_2CH_3$	H	$C(CH_3)OHCH_2CH_3$

Figure 16 Structures of cyclohexadepsipeptidic enniatin homologues (Firkova et al. 2007).

Enniatin B is considerably resistant to heat (melting point: 173-175 °C) (Altomare et al. 1995), acids, and digestion, and has been shown to reach substantial concentrations in breads and breakfast cereals (Noser et al. 2007; Jestoi et al. 2004; Queslati et al. 2011; Mahnine et al. 2011).

Improvements in analytical methods have resulted in better knowledge on the prevalence of enniatins in grains and grain-based foods. However, until now, occurrence data have been sparse and a risk assessment has not yet been performed.

9.4.1 Mode of action of enniatins

The cyclopeptidic enniatins form ionophores with hydrophobic groups on the outside and polar groups in the core, resembling a disc in the three-dimensional conformation (Jestoi 2008; Tedjiotsop Feudijo et al. 2010). They can transport monovalent and divalent cations, either in sandwiched complexes or by creating channels in biological membranes. Due to their hydrophilic and hydrophobic properties, enniatins can be extracted using different solvents.

The primary toxic action of the enniatins is considered to be related to their ionophoric properties. Their antibacterial, antihelmintic, antifungal, herbicidal and insecticidal potency has been demonstrated in *in vitro* toxicity studies (Jestoi 2008; Tedjiotsop Feudijo et al. 2010). Enniatin B in levels up to 100 µM did not show genotoxicity, but demonstrated cytotoxicity at low micromolar concentrations (Ivanova et al. 2006; Jestoi 2008; Behm et al. 2009). The observed activities included specific inhibition of acyl-coenzyme A cholesterol acyltransferase (ACAT), depolarization of mitochondria, inhibition of osteoclastic bone resorption, and induction of apoptosis in cancer cells, as well as interactions with ATP-binding cassette transporters like P-glycoprotein (Tedjiotsop Feudijo et al. 2010; Ivanova et al. 2010). The treatment of bacterial infections in the upper respiratory tract by a so-called fusafungin mixture of enniatins A, A1, B, B1 in a 1 % nasal inhalation solution is the only approved application for enniatins in humans (Lohmann 1988; Kroslak 2002).

Enniatins are considered to be emerging toxins, but are also potential drug candidates, especially in anti-cancer applications. In 2008, a patent on an ABC transporter inhibitor containing enniatins or its analogue as the active ingredient was filed as United States Patent Application (US 2008/0167444 A1).

9.4.2 Toxicokinetics of enniatins

Toxicokinetic data on enniatins from *in vivo* studies have not been published so far. The lack of correlation between *in vitro* and *in vivo* toxicity is presumably due to low bioavailability from per os administration, which seems to be caused by limited uptake from the gastrointestinal tract due to low water solubility and interaction with efflux pumps (Ivanova et al. 2010), and possibly also due to elimination from the systemic circulation by metabolism. In two recent papers, the hepatic metabolism of enniatin B was studied *in vitro* with microsomal assays, and biotransformation products, pathways, and preliminary toxicokinetic parameters have been characterized (Ivanova et al. 2011; Fæste et al. 2011).

9.4.3 Effects in domestic animals

No animal toxicity data that are suitable for use in risk assessment of enniatins in animal feed were found in the data. The low bioavailability of enniatins, primarily due to low absorption, indicates low animal toxicity.

9.4.4 Carry-over of enniatins to humans through animal-derived food products

A survey of Finnish eggs revealed a widespread occurrence of enniatins B and B1 in low concentrations (up to 3.8 µg/kg). This is probably due to the tendency of enniatins to

bioaccumulate in lipophilic media (Jestoi et al. 2009). The presence of enniatin residues in the tissues of poultry exposed to up to 3.8 mg/day enniatin B via the feed has also been reported (Jestoi et al. 2007). No other studies of carry-over are available.

9.4.5 Effects of enniatins in humans and animals relevant for human risk assessment of enniatins

There are no reports of natural cases of mycotoxicosis in humans or animals. Only one oral feeding study has been published, in which mice were fed 0.5 or 1 mg/kg bw, and toxic effects were not detected (Gäuman et al. 1950, review in Jestoi 2008). Acute toxicity and death were only shown after intraperitoneal application of up to 40 mg/kg bw over several days to HIV-infected immunodeficient (SCID) mice (McKee et al. 1997), whereas oral application of up to 1 g/kg bw in mice and 50 mg/kg bw in rats did not produce toxic effects (Gäumann et al. 1950; Lohmann 1988; Bosch et al. 1989).

9.4.6 Human hazard characterizations of enniatins

No TDI or similar levels for safe intake have been derived for humans. The available data are insufficient to establish safe intake levels for humans.

9.5 Beauvericin

Beauvericin is a depsipeptide and was first isolated from the fungus *Beauveria bassiana* (Hamill et al. 1969; Gupta et al. 1995), but it has since been identified as a secondary metabolite in a number of different *Fusarium* species (Logrieco et al. 1998; Moretti et al. 2007). It is structurally similar to the enniatins (Plattner and Nelson 1994).

The chemical structure of beauvericin (Figure 17) features a cyclic lactone trimer of the amide of N-methyl L-phenylalanine and D-α-hydroxyisovaleric acid. The molecular formula is $C_{45}H_{57}N_3O_9$ and the molecular weight 783 g/mol. The melting point is at 93-97 °C (Hamill et al. 1969).

Two beauvericin homologues have been identified so far, beauvericin A, with a molecular weight of 797 g/mol, and beauvericin B, weighing 811 g/mol (Gupta et al. 1995; Logrieco et al. 2002). The presence of one and two additional methylene groups in beauvericin A and B, respectively, account for the increased lipophilicity of these variants. The absence of any chargeable groups in beauvericin explains the poor water solubility and low chemical reactivity. There are, however, several non-ionic polar groups in the molecule allowing the formation of a disc with hydrophobic groups on the outside and polar groups in the core.

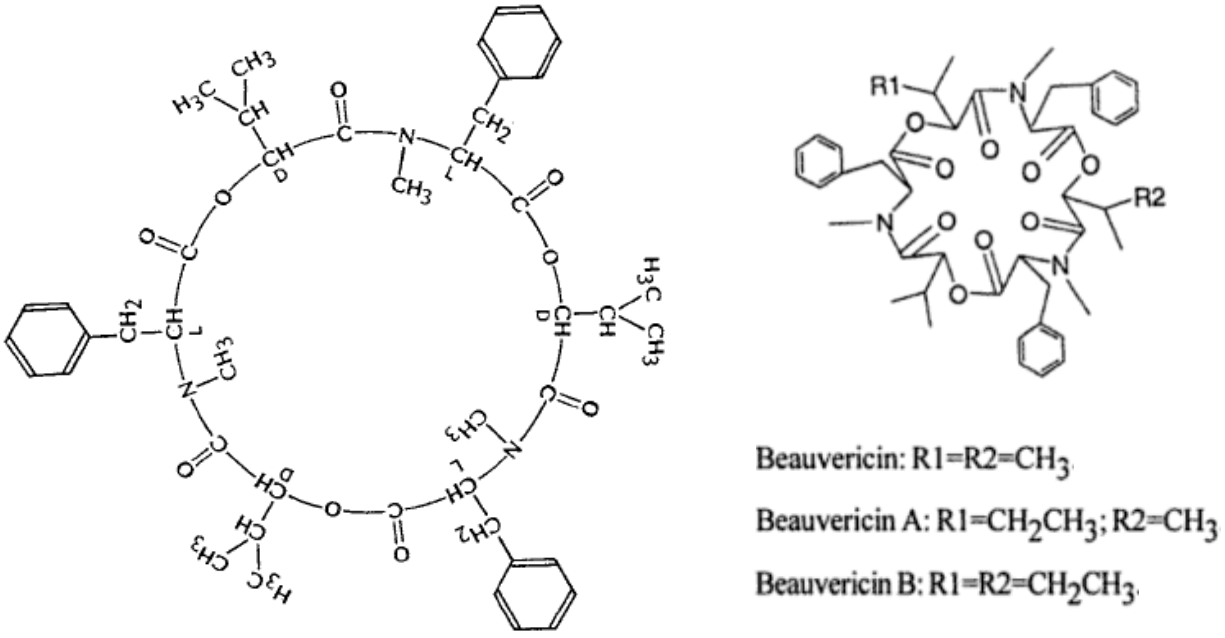


Figure17. Beauvericin structure (Peeters et al. 1988) and beauvericin homologues (Logrieco et al. 2002).

The considerable prevalence of beauvericin in grains, including maize and products thereof, and the resultant, presumably continuous, low-level exposure of humans and animals to beauvericin in food and feed have become evident with the improvement of analytical methods. However, until now, occurrence data have been sparse and a risk assessment has not yet been performed.

9.5.1 Mode of action of beauvericin

Beauvericin is an ionophoric molecule that can form stable and lipophilic complexes with cations and transport them into the lipophilic phase. One cation – one ionophore complexes (1:1) are usually formed, but sandwich-type multi-complexes, including two (1:2) or three (1:3) beauvericin molecules, have also been observed. The stable beauvericin-metal ion cluster can be formed into a channel pore in the cell membrane. Beauvericin complexes with monovalent and divalent cations (it binds Ca^{2+} with high efficiency) and activates their transport into mammalian mitochondria and through membranes (Lemmens-Gruber et al. 2000).

The primary toxic action of beauvericin is related to its ionophoric properties, as beauvericin interferes with the electrochemical gradient of the membranes, resulting in the disturbance of the normal physiological levels of cations in the cell. Apoptosis can be induced with 50 µM beauvericin, triggering an increase in the cytoplasmatic calcium concentration (Dombrink-Kurtzman 2003). Furthermore, beauvericin can interfere with DNA replication by intercalation of double-stranded DNA (Pocsfalvi et al. 1997) and leading to internucleosomal DNA fragmentation (Ojcious et al. 1991). A specific cholesterol acyltransferase inhibitor activity has been found for 3 µM beauvericin (Tomoda et al. 1992). Additionally, beauvericin has been found to be chemosensitizing, inhibiting the active efflux of other antibiotics and thereby increasing their efficacy (Lee et al. 2001).

The toxic potential of beauvericin has been elucidated in a considerable number of *in vitro* cytotoxicity and genotoxicity assays (Jestoi 2008). Beauvericin is antimicrobial (IC_{50} about 1.5 µg/ml), but not mutagenic in the Ames test. It has an insecticidal potential with a mean LD_{50} of 10 µg/ml (Gupta et al. 1991; Moretti et al. 2007). Cytotoxicity assays using mammalian cells showed that effect concentrations are in the range 2.5 to 50 µM (Logrieco et al. 2002; Jestoi 2008).

Beauvericin is considered to be an emerging toxin, but also has potential as a drug candidate that can lower cholesterol levels in blood. In 1991, a patent for a tablet containing 5 mg beauvericin was filed (Omura et al. 1991). In traditional Chinese medicine, beauvericin is used as a constituent in anticonvulsant and antineoplastic preparations (Mao 1985). Beauvericin has also been shown to inhibit the multidrug transport protein in human cancer cells, suggesting clinical use in combination with chemotherapeutic drugs (Sharom et al. 1998).

9.5.2 Toxicokinetics of beauvericin

Toxicokinetic data on beauvericin are sparse. There is one study on the metabolism and inhibition of beauvericin in human and rat liver microsomes (Mei et al. 2009). Furthermore, beauvericin toxicokinetics were studied after oral administration of rats with 0.5, 1.0, and 2.0 mg/kg. The *in vivo* half-life was about 3.0 h and the dose-normalized $C_{max,norm}$ = 6.4 kg/l. Beauvercin was found to be substrate and inhibitor to the cytochromes CYP 3A and CYP2C19 and effectively inhibited ketoconazole metabolism, a possible explanation for the observed synergistic effect after co-administration.

9.5.3 Effects of beauvericin in domestic animals

Very little information is available on the toxic effects of beauvericin that are suitable for use in risk assessment. No information is available regarding effectcs of beauvericin in Norwegian farmed finfish species.

Pig

A strain of *Fusarium proliferatum* isolated from maize that was associated with an incident of swine mycotoxicosis in the US was shown to produce beauvericin (Plattner and Nelson 1994). However, beauvericin usually co-occurs with other grain-contaminating mycotoxins like fumonisins, enniatins, moniliformin, DON, nivalenol, HT-2, T-2, and zearalenone (Jestoi 2008). Therefore, there is no evidence that beauvericin was the disease-causing agent in this incident.

Poultry

Strains of *Fusarium subglutinans* that were shown to be toxic to ducklings were able to produce beauvericin (Marasas et al. 1984). However, there was no evidence that beauvericin was responsible for the observed toxic effect.

No acute effects were seen in ducklings given a single dose of 100 mg/kg bw beauvericin by gavage (Vesonder et al. 1999).

Diets with up to 12 mg/kg of beauvericin and 2.7 mg/kg of moniliformin fed to broiler chickens for 37 days via maize inoculated in the field showed no effects on growth, carcass traits, or chemical composition of the carcass (Zollitsch et al. 2003).

No growth, slaughter performance, meat quality or chemical carcass composition effects were found in turkeys given 0 – 2.5 mg/kg feed (+ 2.4 mg moniliformin/kg feed) (Leitgeb et al. 2000).

Table 33. Critical effect (most sensitive endpoint) for beauvericin in feed at concentrations where NOAEL or other available effect levels are derived/identified for various species.

Animal	Critical Effect	Concentration in feed	Available effect level	Pivotal referece
Pig	No information found			
Broiler chicken	Growth, carcass traits or chemical composition of the carcass	12 mg/kg	No effect (one dosage level)	Zollitsch et al. (2003)
Laying hen	No information found			
Ruminants				
Horse				
Rabbit				
Dog				
Cat				
Fish				

9.5.4 Carry-over of beauvericin to humans through animal-derived food products

A survey of Finnish eggs revealed the occurrence of beauvericin at trace levels, with up to 1.3 µg/kg in egg yolks (Jestoi et al. 2009). The transmission rate of beauvericin from feed to eggs could not be calculated due to lack of data. Beauvericin was detected in 2 % of the turkey meat samples in a Finnish study in 2004, but in low concentrations under the limit of quantitation. (<1 µg/kg) (Jestoi et al. 2007). No residues of beauvericin were detected in carcasses, heart, gizzards, livers or intestinal tract of broilers fed diets with up to 12 mg/kg of beauvericin and 2.7 mg/kg of moniliformin for 37 days via maize inoculated in the field (Zollitsch et al. 2003).

No data on beauvericin have been found for ruminants, horse, rabbit, dog, cat and fish.

9.5.5 Effects of beauvericin in humans and animals relevant for human risk assessments

The biological activity of beauvericin has only been tested in a few *in vivo* studies and no acute effects have been observed. The LD_{50} in mice was found to be >100 mg/kg for per os administration and >10 mg/kg for intraperitoneal application (Omura et al. 1991). Feeding studies with turkeys and broilers, using up to 12 mg/kg beauvericin in the feed, did not show any effects.

9.5.6 Human hazard characterizations of beauvericin

No TDI or similar levels for a safe intake on beauvericin have been derived. The available data are insufficient to establish safe intake levels.

9.6 Moniliformin

International bodies such as EFSA or JECFA have not previously assessed moniliformin, but a review paper has been published (Peltonen et al. 2010). Moniliformin normally occur as the sodium or potassium salt of semisquaric acid (3 hydroxycyclobut-3 ene-1,2dione, see Figure 17). It is highly soluble in water due to its high polarity. Sensitive analysis of the molecule has been a demanding task, but methods are now available (review in Jestoi 2008).

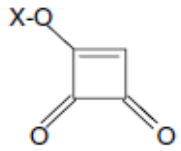


Figure 17. The chemicale structure of moniliformin. X=H (free acid), Na (sodium salt), or K (potassium salt).

9.6.1 Mode of action of moniliformin

Moniliformin is an inhibitor of those enzymes that depend on the cofactor thiamine pyrophosphate (TPP) (Pirrung et al. 1996). Several important enzymes involved in energy metabolism, such as pyruvate dehydrogenase, ketoglutarate dehydrogenase and pyruvate decarboxylase, are dependent on TPP and are susceptible to moniliformin. Moniliformin may also inhibit gluconeogenesis and aldose reductase (reviewed in Jestoi 2008; Peltonen et al. 2010). The *in vitro* toxicity of moniliforminis is relatively low compared with *in vivo* data, possibly due to low *in vitro* uptake of moniliformin into cells, as moniliformin is ionised at physiological pH (Wu et al. 1995; Peltonen et al. 2010).

9.6.2 Toxicokinetics of moniliformin

Moniliformin could no longer be detected in the urine of rats 12 hours after oral administration of single doses of 5-50 mg/kg bw (Jestoi et al. 2010). No other information on the toxicokinetics on moliniformin are currently available.

9.6.3 Effects of moniliformin in domestic animals

The effects of moniliformin in animals, including farm animals, have been reviewed (Peltonen et al. 2010). The available information is limited, but some feeding experiments have been published. Culture material from *Fusarium* spp. has been used in most studies.

Pig

Only studies with moniliformin-containing fungal cultures of *F. fujikuroi* are available. Weaned barrow piglets were fed 0, 25, 50, 100 or 200 mg/kg of moniliformin from the culture material for 28 days (Harvey et al. 2001). No effects were observed at 25 mg/kg. Haematological values were affected in barrows fed 50 mg/kg and above. At 100 mg/kg feed and above, reduced feed intake and weight gain were shown, and mortalities were also reported.

To summarise, a NOAEL at 25 mg/kg of moniliformin from culture material in the diet is identified for pigs.

Poultry

Several feeding studies have been published in which poultry have been fed moniliformin-containing fungal culture materials or pure moniliformin mixed into the feed (reviewed in Peltonen et al. 2010). The toxic lesions of moniliformin in poultry occur in various tissues, including liver, kidney, and heart. Clinical symptoms in the chickens fed diets containing moniliformin included muscle weakness, poor feathering, decreased feed and water intake, and reduced immune response. Dose-response relationships have been measured for the various effects. Broiler chicken fed purified moniliformin at 0, 27 or 154 mg/kg feed from age 1-14 days showed dose-response related lesions with mortalities (Javed et al. 2005). Pathological lesions were observed particularly in the liver, but other tissues were also affected.

Broomhead et al. (2002) fed broilers from 1 to 7 weeks of age and turkeys from 1 to 14 weeks of age with culture material, providing 0, 25 and 50 mg/kg diet to the broilers and 0, 12.5, 25, 37.5 and 50 mg/kg to the turkeys. In broilers, increased serum gamma glutamyltranserferase activity was shown at 25 mg/kg and above, and lower body weight gain and feed conversion, increased heart and proventriculus weights, as well as increased mortality were shown at 50 mg/kg. In turkeys, no significant effects were shown at 12.5 mg/kg, but heart weight was increased at 25 mg/kg and above, and increased liver weight and reduced weight gain were shown at 37.5 mg/kg and above.

Other researchers have not shown adverse effects in broiler chickens fed diets with up to 16 mg/kg of moniliformin from culture material (Allen et al. 1981a) or up to 2.7 mg/kg of moniliformin plus 12 mg/kg of beauvericin from maize inoculated in the field (Zollitsch et al. 2003). These results may indicate a higher tolerance for moniliformin when fed as an inoculum rather than as isolated toxin.

Reduced immune responses have been shown in broiler chickens fed a diet with moniliformin from culture material at 75 and 100 mg/kg feed, but no effects were shown at 50 mg/kg (Li et al. 2000). Chickens fed moniliformin at 100 mg/kg also had a reduction in feed intake and weight gain.

Laying hens fed moniliformin from culture material at 50 and 100 mg/kg feed for 420 days from 3 weeks of age showed dose-response effects, with reductions in liver weight at the lowest dose and also increased heart weight, reduced kidney weight, and impaired egg production at the highest dose (Kubena et al. 1999).

In sum, for broiler chickens, purified moniliformin elicited liver pathology at 27 mg/kg diet and, in another study, moniliformin from culture material increased gamma glutamyltranserferase at 25 mg/kg and produced more pronounced effects (on performance and pathology) at 50 mg/kg. On this basis we identified a LOAEL for moniliformin in broiler chickens at approximately 25 mg/kg diet. For turkeys, we identified a NOAEL at 12.5 mg/kg based on increased heart weight at 25 mg/kg and above when fed moniliformin from culture material.

Ruminants

Data on effects of moniliformin in ruminants have not been identified.

Horse

Data on effects of moniliformin in horses have not been identified.

Dog and cat

Although no data on the effects of moniliformin on dogs and cats were found, a study of moniliformin (0, 8 and 17 mg/kg diet) in another carnivore, mink, indicates reproductive toxicity (Morgan et al. 1998). Culture material mixed into commercial feed for female mink from 2 weeks before breeding season until the offspring were 8-weeks old. Neonatal mortality was increased and body weights at birth and 8-weeks of age were decreased in the highest dosage group. No other effects were revealed.

Rabbit

Data on effects of moniliformin in rabbits have not been identified.

Fish

In a study with Nile tilapia fingerlings, the fish (2.7 g) were fed diets containing 0, 10, 40, 70, 150 mg/kg of moniliformin obtained from *Fusarium moniliforme* culture materials for 8 weeks. Fish fed either 70 or 150 mg moniliformin/kg diet had significantly lower mean weight gains than control fish. Haematocrit was significantly reduced only in fish fed diets containing 150 mg moniliformin/kg diet. Serum pyruvate levels were significantly higher than in control fish for all tilapia fed moniliformin. No histopathological lesions were observed in tilapia fed diets containing moniliformin (Tuan 2003). Young channel catfish fed 0, 20, 40, 60, and 120 mg moniliformin/kg feed showed reduced weight gain at the lowest exposure level after 10 weeks. Fish fed diets with the lowest concentration of moniliformin (20 mg/kg diet) had significantly less weight gain than control fish. Increasing the level of moniliformin in the diets resulted in a linear decrease in weight gain. Mean serum pyruvate level was dose-dependently significantly higher in fish fed the diet containing 60 mg moniliformin/kg diet and higher, and smaller nuclei of liver cells were observed in fish fed diets containing the two highest levels of moniliformin (Yildirim 2000). No data on moniliformin were found for Norwegian farmed finfish species.

To summarise for fish, Nile tilapia showed increased serum puyruvate from 10 mg/kg (identified LOEL) and reduced weight gain from 70 mg/kg (40 mg/kg identified NOAEL). In channel catfish, increased serum pyruvate and smaller nuclei of liver cells were shown at 60 mg/kg and above, from which we identified a NOAEL at 40 mg/kg.

Table 34. Critical effect (most sensitive endpoint) for moniliformin in feed at concentrations where NOAEL or other available effect levels are derived/identified for various species.

Animal	Critical Effect	Concentration in feed	Available effect level	Pivotal references
Pig	Haematological values	25 mg/kg	NOAEL	Harvey et al. (2001)
Chicken	Reduced production, and pathological effects	Approximately 25 mg/kg	NOAEL	Broomhead et al. (2002); Javed et al. (2005)
Turkey	Increased heart weight	12.5 mg/kg	NOAEL	Broomhead et al. (2002)
Ruminants	*No information found*			
Horse				
Rabbit				
Dog				
Cat				
Salmon or trout				

9.6.4 Carry-over of moniliformin to humans through animal-derived food products

No information on moniliformin carry-over to humans through animal-derived products was identified.

9.6.5 Effects of moniliformin in humans and animals relevant for human risk assessments

There is very little information available regarding the oral toxicity of moniliforminin that is relevant for human risk assessment. Poultry seems to be more sensitive to moniliformin than other species with effects occurring from 60 mg/kg feed, the lowest dose used in the study (Abbas et al. 1990). One out of three rats given 1.0 or 1.5 g crystalline moniliformin/kg feed and two out of three given 2.0 or 2.5 g/kg feed died within 16 h. Haemorrhaging in the intestines was observed in rats given 2 g moniliformin/kg diet, while no lesions were observed in rats exposed to 0.75 g /kg feed or less, corresponding to 80 mg/kg bw (Abbas et al. 1990).

In a review paper (Peltonen et al. 2010), an unpublished 28-day oral study according to OECD guideline 407 was briefly described. Based on the pathological and histopathological observations from this experiment in Sprague Dawley rats (5/group) given oral daily doses of moniliformin (99.8 % pure), the authors estimated a NOAEL value of around 10 mg/kg bw/day.

9.6.6 Human hazard characterizations - of moniliformin

No TDI or similar levels for safe intake in humans have been derived. The available data are insufficient to establish a safe intake level. According to a review of the information available about moniliformin, a NOAEL of 10 mg/kg bw/day was derived from an unpublished sub-chronic study involving oral exposure of mice (Peltonen et al. 2010).

9.7 Ochratoxin A

Ochratoxin A (OTA) is a mycotoxin that occurs worldwide and is produced by several species in the *Penicillium* and *Aspergillus* genera. These fungi primarily infect and grow during

transport and storage. Ochratoxin A comprises a dihydrocoumarin linked to a b-phenylalanine via an amide bond (Figure 18) and has CAS No 303-47-9.

Ochratoxin A is heat-stable and does not decompose at normal food and feed processing temperatures, as the molecule only degrades at temperatures exceeding 250°C.

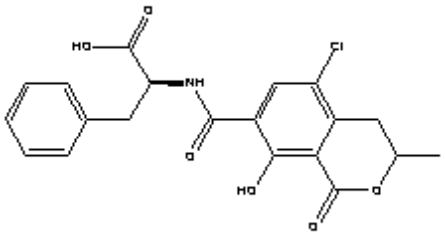


Figure 18. Chemical structure of ochratoxin A.

9.7.1 Mode of action of ochratoxin A

Ochratoxin A affects several different biochemical pathways. It is known to inhibit the enzyme phenylalanine–tRNAPhe synthetase, thereby blocking tacylation of amino acids and consequently peptide elongation in protein synthesis.

Ochratoxin A also reduces the activity of glycolytic enzymes and increases the activity of gluconeogenic enzymes. Ochratoxin A has also been shown to increase lipid peroxidation and formation of reactive oxygen species. The significance of the oxidative effects of ochratoxin A has particularly been studied in order to clarify the exact mode of action related to the kidney carcinogenic activity of ochratoxin A (JECFA 2001, 2007; EFSA 2006).

9.7.2 Toxicokinetics of ochratoxin A

Ochratoxin A is rapidly absorbed from the intestines in most species, with most absorption from the small intestine. The absorption has been estimated to range from 40 % in chickens to 66 % in pigs (reviewed in EFSA 2006; JECFA 2007). Ochratoxin A in plasma is largely bound to albumin and other plasma macromolecules and the unbound fraction has been estimated to be as low as 0.02 % (Hagelberg et al. 1989). Following absorption, the toxin is then rapidly distributed by the blood, mainly to the kidneys, but lower concentrations are also found in the liver, muscle, and fat. Specific transport proteins are probably involved in cellular uptake of ochratoxin A into kidneys, where it accumulates.

Ochratoxin A in plasma mainly occurs as the parent compound, but minor amounts of conjugates and hydroxylation products have also been reported. All metabolites are considered to be less toxic than ochratoxin A itself. In ruminants, microorganisms in the rumen efficiently hydrolyse ochratoxin A to phenylalanine and ochratoxin α, prior to absorption. Ochratoxin α is considered to be of low toxicity and secretion of this metabolite in milk is not considered to pose any significant health risk (EFSA 2006; JECFA 2001, 2007).

Ochratoxin A and metabolites are excreted in milk, urine, and faeces. In ruminants, the main proportion in milk is excreted as ochratoxin α, a metabolite of low toxicity. In monogastric animals and humans, ochratoxin A is secreted in the milk, and thus mothers' milk may be a significant route of exposure for infants when mothers are ingesting ochratoxin A (Skaug et al. 1998, 2001).

The proportion of ochratoxin A and its metabolites that are secreted in the urine and faeces is species-dependent, possibly determined by the extent of enterohepatic recycling and serum protein binding in each species (EFSA 2006). These factors are also considered to be the main factors explaining the large variation in plasma half-life between species. The serum half-life varies between species, from 1-1.5 days in mice, to 2-5 days in rats, 3-5 days in pigs, to around 20 days in macaque and vervet monkeys, and to 35 days in one human volunteer (EFSA 2006; JECFA 2001).

Preliminary results from a study of toxicokinetics of ochratoxin A in smoltified salmon fed up to 2.4 mg ochratoxin A/kg diet (start weight 50-70 g) for eight weeks indicate that ochratoxin A does not accumulate in tissue and that tissue concentrations declined in the following order, liver > kidney > muscle (Bernhoft et al., manuscript in preparation). The muscle level of ochratoxin A at the end of 8 weeks exposure to 2.4 mg/kg was at the quantification level of 0.09 µg/kg.

9.7.3 Effects of ochratoxin A in domestic animals

EFSA has reviewed the effects of ochratoxin A in animal feed (EFSA 2004d).

Pig

Pigs are considered to be the farm animal species that is most sensitive to ochratoxin A. The critical effect has been regarded to be nephrotoxicity. Nephropathy has been induced in pigs fed diet contaminated with 0.2, 1 or 4 mg/kg (Krogh et al. 1974, 1988; reviewed by EFSA 2004d). Female pigs fed ochratoxin A at 0.2 mg/kg diet (equivalent to 8 µg/kg bw/day) for 90 days showed reduced activity of the renal enzymes phosphoenolpyruvate carboxykinase and gamma-glutamyl transpeptidase, as well as reduced tubular secretion of p-aminohippurate and increased glucosuria, indicators of decreased kidney function. These results are the basis for the human PTWI (EFSA 2004d; Walker and Larsen 2005).

Pleadin et al. (2012) fed 70 kg pigs with 0.3 mg ochratoxin A/kg diet for 30 days and demonstrated changes in serum biochemical parameters indicative of impaired kidney and liver functions, including higher creatinine, urea, potassium and alkaline phosphatase concentrations, and lower concentrations of glucose and total protein than in control pigs. Other studies have observed mild kidney effects at higher feed concentrations (0.8-1.0 mg/kg) or lower feed concentrations (0.09-0.18 mg/kg) (EFSA 2004d; Stoev et al. 2001, 2002a, 2002b, 2011). However, at the lower levels other mycotoxins, such as penicillic acid, fumonisins, citrinin or others, may have influenced the response.

Concerning immune effects, growing pigs fed ochratoxin A at 2.5 mg/kg feed, demonstrated reductions in the phagocytic activity of macrophages and production of IL2 (Harvey et al. 1992). Stoev et al. (2000) demonstrated that growing pigs fed a diet contaminated with *A. ochraceus,* which contained 1 or 3 mg ochratoxin A/kg, for up to 3 weeks were more susceptible to *Salmonella* infection. In a further experiment in which growing pigs were fed purified ochratoxin A at 1 mg/kg feed and immunised against *Salmonella*, ochratoxin A-exposed pigs presented with haemorrhagic diarrhoea, whereas control pigs did not. These

results indicate immunosuppression and delayed immune response due to ochratoxin A, concomitant with a nephrotoxic effect.

A significant decrease in body weight, daily weight gain and feed efficiency were observed in growing pigs fed ochratoxin A at 0.025 mg/kg diet for 119 days until slaughtering (Malagutti et al. 2005).

Spermatological parameters were evalualted in boars given 0.020 mg ochratoxin A per day (0.08 µg/kg bw per day, estimated to be approximately 0.003 mg/kg diet) for 6 weeks (Biró et al. 2003). Significantly lower ejaculate volume, sperm viability and motility were observed in exposed boars than in controls after a lag period. No histological effects were seen in Leydig cells or epidydimal structures.

In sum for pigs, a LOAEL at 0.2 mg/kg diet based on nephrotoxic effects has been derived. However, effects on spermatological parameters and on slaughter weight / daily gain / feed efficiency allow us to identify LOAELs of 0.003 and 0.025 mg/kg diets, respectively.

Poultry

Nephrotoxicity, reduced feed intake, and, in particular, various immunosuppressive effects of ochratoxin A have been demonstrated in poultry. These have been measured by inhibition of humoral, cellular and innate immune responses, including cellular depletion of lymphoid organs, in broiler chicks at feed concentrations of 0.5-1 mg/kg (EFSA 2004d; Pfohl-Leszkowicz and Manderville 2007). Severe clinical signs of ochratoxicosis (weight loss, decreased egg production, increased water intake, diarrhoea, excessive urine excretion) occur in poultry fed above 2 mg/kg feed. Increased susceptibility to infectious agents has been confirmed by the observation that chicks infected with inclusion body hepatitis virus and concurrently exposed to ochratoxin A at 0.5 mg/kg feed for up to 35 days had more pronounced haematological, biochemical, histopathological changes than chicks exposed to the virus alone or ochratoxin A alone (Sandhu et al. 1995, 1998).

Recently, Ul-Hassan et al. (2012a) conducted a study on immunological responses of male white Leghorn chicks fed ochratoxin A contaminated feed at 0, 0.1, 0.5, 1.0 and 1.5 mg/kg for 21 days. The results showed dose-response related effects, with significant reductions in the relative weights of the bursa of Fabricius and the spleen, lower phagocytic index of the blood reticuloendothelial system, reduced macrophage function and lymphoproliferative response, and also decreased antibody titres at lowest dosage level.

Furthermore, Ul-Hassan et al. (2012b) demonstrated immune suppression in the progeny of breeder hens fed ochratoxin A contaminated feed (3 and 5 mg/kg).

Ochratoxin A in feed at 1.3, 2.6, or 5.2 mg/kg feed decreased egg production of laying hens in a dose-dependent manner (Scholtyssek et al. 1987). At the highest dose, egg production was as low as 28.4 % of that of the control group.

To summarise, a NOEL remains to be established for ochratoxin A in birds. A LOAEL in male chicks at 0.1 mg/kg diet was identified, based on a recent study on immunological effects. Previouly, a LOAEL at 0.5 mg/kg for broiler chicks based on immune function has been derived.

Ruminants

Ruminants are less sensitive to ochratoxin A due to extensive metabolism in the rumen prior to absorption (see above). In pre-ruminant calves given 0.1 - 2 mg ochratoxin A/kg bw per

day for 30 days, polyuria and mild enteritis and also mild tubular degeneration were shown at the end of the exposure (Pier et al. 1976, quoted by Krogh 1991). Cows given ochratoxin A orally for 4 days at 0.2 to 1.7 mg/kg bw per day remained clinically normal (Ribelin et al. 1978).

In sheep, ochratoxin A at 5 mg/kg diet was rapidly degraded to ochratoxin α by rumen microorganisms, primarily protozoa, and neither compound was detected in blood (Kiessling et al. 1984). However, this ability to degrade ochratoxin A decreased after the sheep were fed a high ration of compound feed.

Xiao et al. (1991) showed a 4.3-fold increase of the systemic availability of ochratoxin A in sheep fed a complete grain diet compared to sheep fed a complete hay diet. Blank et al. (2003) found considerable amounts of ochratoxin A absorbed by sheep when fed a diet with 70 % compound feed. About 10 – 20 % excretion of ochratoxin A compared to ochratoxin α in faeces and urine was found when the sheep were fed 0.01 to 0.03 mg/kg of ochratoxin A. Thus, the results show increased uptake of ochratoxin A in ruminants fed non physiological amounts of compound feed.

Horse

Only individual case reports of suspected cases of ochratoxin A intoxications were available to EFSA (2004d). No quantitative toxin exposure was presented.

Rabbit

Ochratoxin A from *A. ochraceus* was given via gastric intubation to rabbit does from 6 to 18 days of gestation, at levels of 0.025, 0.05, and 0.1 mg/kg bw per day (Dwiwedi et al. 2004). In the highest dosage group (corresponding to approximately 2 mg/kg diet), teratogenic effects were observed in the form of a significant increase in the incidence of gross, skeletal malformations as well as soft tissue anomalies. Furthermore, the number of live foetuses and mean foetal weight and length were lower at the highest dosage level.

Dog and cat

Beagle dogs were given ochratoxin A at doses of 0.1 or 0.2 mg/kg bw per day (approximately 5 and 10 mg/kg feed) for 14 days. Kidney tubular necrosis and ultrastructural changes in the proximal tubuli were seen at both doses (Kitchen et al. 1977a, b, c). Necrosis of lymphoid tissues was also seen in the thymus and tonsils of the dogs. A NOAEL could not be established. A LOAEL of 5 mg/kg feed was identified.

No dose-response studies of ochratoxin A that are appropriate for use in risk assessment were found for cats.

Fish

Channel catfish were fed ochratoxin A containing culture material at dietary levels of 0, 0.5, 1.0, 2.0, 4.0, and 8.0 mg /kg feed for 8 weeks. Fish fed 1.0 mg/kg feed and above had a significantly reduced weight gain and reduced number of exocrine pancreatic cells in the liver. Catfish fed 2.0 mg/kg and above had an increased incidence and severity of melanomacrophage centres in the hepatopancreatic tissue and posterior kidney. Feed conversion was significantly decreased, and hepatic and posterior kidney lesions increased in fish fed 4.0 mg/kg feed and above. Haematocrit was only significantly affected in the highest

(8.0 mg/kg feed) exposure group (Manning et al. 2003b). In another study, channel catfish exposed to ochratoxin A-enriched diets (2.0 and 4.0 mg/kg) for 6 weeks were challenged with *Edwarsiella ictaluri* to test disease resistance (Manning et al. 2005c). Fish fed at the dietary ochratoxin A level of 4.0 mg/kg had a significantly higher mortality (80.5 %) than control catfish (68.8 %) after bacterial challenge (Manning et al. 2005c).

The acute oral toxicity of ochratoxin A was assessed in sea bass (*Dicentrarchus labrax* L). Fish (40 g) were orally administered ochratoxin A by gavage, once a day for four days, at levels of 0, 50, 100, 150, 200, 250, 300, 350, and 400 µg/kg bw (El-Sayed et al. 2009). Acute mortality occurred at levels of 150 µg/kg bw and above, and acute oral 96 h LC_{50} levels were set at 277 µg/kg bw (El-Sayed et al. 2009).

Adult Atlantic salmon were fed ochratoxin A at 3 dietary concentrations (0.09-0.33 mg/kg) for 15 weeks. No significant differences were observed in growth, feed intake, feed conversion, or plasma enzymes (Döll 2010). Preliminary results from a study of administration of ochratoxin A at 0, 0.2, 0.4, 0.8, 1.6 and 2.4 mg/kg diet to smoltified Atlantic salmon (start weight 50-70 g) for eight weeks inidcate that adverse effects do not occur clinically, or can be measured clinical-chemically, or be detected pathologically (Bernhoft et al., manuscript in preparation).

To summarise on ochratoxin A in fish, studies on channel catfish have shown reduced weight gain and number of exocrine pancreatic cells in the liver at 1 mg/kg diet and a NOAEL was identified at 0.5 mg/kg. No effects were detected in a study on Atlantic salmon fed ochratoxin A at concentrations up to 2.4 mg/kg diet.

Table 35. Critical effect (most sensitive endpoint) for ochratoxin A in feed at concentrations where NOAEL or other available effect levels are derived/identifed for various species.

Animal	Critical Effect	Concentration in feed	Available effect level	Pivotal References
Pig	Nephrotoxic effects Spermatological parameters Reduced performance	0.2 mg/kg 0.003 mg/kg 0.025 mg/kg	LOAEL LOAEL Effect level (one dosage)	EFSA (2004d) Biro et al. (2003) Malagutti et al. (2005)
Broiler chicken male Broiler chicken	Immunological effect Reduced immunological function	0.1 mg/kg 0.5 mg/kg	LOAEL LOAEL	Ul-Hassan et al. (2012a, b) Sandhu et al. (1995, 1998)
Ruminants Horse	*No dose-response studies usable for risk assessment were found*			
Rabbit	Teratogenic effects	1 mg/kg	NOAEL	Dwiwedi et al. (2004)
Dog	Kidney tubular necrosis	5 mg/kg	LOAEL	Kitchen et al. (1977a, b, c)
Cat	*No dose-response studies usable for risk assessment were found*			
Atlantic salmon	Reduced performance	2.4 mg/kg	NOAEL	Bernhoft et al. (in preparation)

9.7.4 Carry-over of ochratoxin A to humans through animal-derived food products

A large fraction of the ochratoxin A in plasma is bound to albumin and other macromolecules and has a prolonged plasma half-life. Ochratoxin A will therefore occur in blood-containing tissues. In addition, ochratoxin A has been detected in food samples of animal origin, such as eggs, milk, offal, and meat.

In the exposure estimations made by EFSA(2004d), food of animal origin only had a minor contribution to the total dietary intake (generally < 3 %, and in populations with specific dietary preferences < 10%).

9.7.5 Effects of ochratoxin A in humans and animals relevant for human risk assessments

Human risk assessment is currently based on the nephrotoxic effects of ochratoxin A, and rats and pigs are particularly sensitive to this toxin. Ochratoxin A also causes tumours in the kidney. The mechanism for this carcinogenic effect is still a matter of debate.

In a series of feeding studies in which pigs were fed 0, 0.2, 1 or 5 mg ochratoxin A/kg feed, equivalent to 0, 8, 40 and 40 μg/kg bw/day, for periods of 5 days up to 2 years, dose-related decreases in renal enzyme activities and renal functions were found. Progressive nephropathy was observed in female pigs fed 40 μg/kg bw/day, but not in females fed 8 μg/kg bw/day for 2 years, but enzymatic alterations were observed even in the lowest dose group. Based on this, a LOAEL of 8 μg/kg bw/day was established.

9.7.6 Human hazard characterizations of ochratoxin A

Both JECFA (2001, 2007) and EFSA (2006) have concluded in their evaluations that effects are probably due to indirect genotoxic effects, possibly involving oxidative DNA damage. Both organisations based their evaluations on a NOAEL of 8 μg/kg bw/day from a 2-year pig study and based their evaluations on a threshold model. Both evaluations concluded that the accumulation of ochratoxin A in the kidneys is of importance and that a tolerable weekly intake would be more relevant than a TDI. JECFA established a PTWI of 100 ng/kg bw/day and EFSA a TWI of 120 ng/kg bw/day.

Human exposure to ochratoxin A in Norway has been estimated in blood donors (Thuvander et al. 2001). The exposure was estimated to be below the TDI set by SCF in 1996, which was used at that time (5 ng/kg bw/day), and well below the current PTWI of 100 or 120 ng/kg bw/day. However, Skaug et al. (2003) reported blood plasma concentrations higher than those reported by Thuvander et al. (mean values of 0.397 ng/mL versus 0.18 ng/mL plasma, respectively), indicating that Thuvander et al. may have under-estimated the exposure. In Europe, the contribution from cereals is estimated to be below 50 % of the total dietary intake, using an average concentration of 0.29 μg ochratoxin A/kg in the cereal products (EFSA 2006). This concentration is in the same range as the annual mean concentrations found in Norwegian cereals (Tables 8-10).

9.8 **Aflatoxins**

In grain, aflatoxins are mainly found in maize and maize gluten used for animal feed, and are seldom found in grains intended for human consumption in Norway. This is mainly because aflatoxin-producing fungal species are not normally adapted to the Norwegian climate. Consequently, in the present risk assessment for mycotoxins in grains in Norway, aflatoxins are only evaluated as imported contaminants in animal feed. Human exposure to aflatoxins will not be assessed since the main sources for aflatoxin uptake are non-grain foods, including dried fruits and nuts.

Among the 18 different types of aflatoxins identified, major members are aflatoxin B1, B2, G1 and G2 (Asao et al. 1965) and current legislation focusses on these four aflatoxins. Aflatoxin B1 (AFB1) is the most important compound with respect to both prevalence and toxicity for man and animals (Figure 19). Aflatoxins M1 and M2 are hydroxylated metabolites of aflatoxin B1 and B2 and are the main forms found in milk.

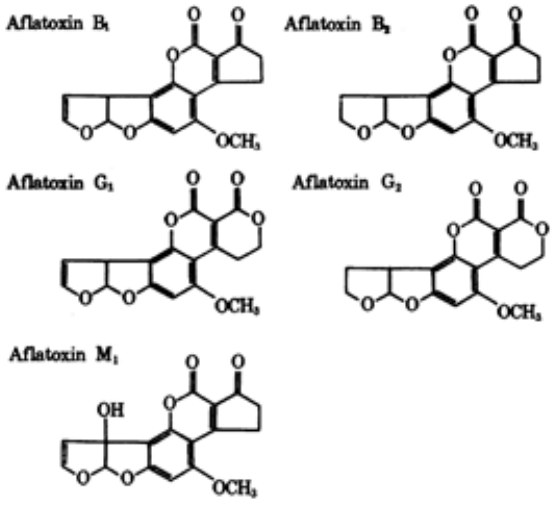


Figure 19. Chemical structures of important aflatoxins.

Aflatoxins are odourless, tasteless and colourless difuranocoumarins. They are classified in two groups according to their chemical structures: difuranocoumarocyclopentenones (including aflatoxin B1, aflatoxin B2, aflatoxin M1, aflatoxin M2) and difuranocoumarolactones (including aflatoxin G1, aflatoxin G2). The individual aflatoxins can be distinguished by the colour of their fluorescence under ultraviolet light. Aflatoxin B1 and aflatoxin B2 produce a blue fluorescence, whereas aflatoxin G1 and aflatoxin G2 have a green fluorescence. Aflatoxins are soluble in methanol, chloroform, acetone, and acetonitrile, and in the dry state are very stable up to the melting point.

Several risk assessments of aflatoxins have been published by JECFA (JECFA 1987, 1997, 1998, 2001, 2007), EFSA (2007, 2009), and others. Furthermore, the genotoxic and carcinogenic effects of aflatoxins have been evaluated by IARC (IARC 1993). Although there are reports of acute/sub-acute human (Krishnamachari et al. 1975; Lewis et al. 2005) and animal (Blount 1961; McKenzie et al. 1981) aflatoxicosis, which may lead to lethal hepatotoxicity, the critical effect in human risk assessments is the carcinogenic effect.

IARC concluded that “naturally occurring aflatoxins are carcinogenic to humans (Group 1)”. The evaluations generally consider aflatoxins to be genotoxic carcinogens, such that no threshold dose can be defined below which tumour formation would not occur.

Aflatoxins are assessed as a group, based on the data for aflatoxin Bl since the toxicological profiles of the most important naturally occurring aflatoxins, Bl, B2, Gl, and G2, appear to be similar (IARC 1993; JECFA 1998; EFSA 2009). Additionally, the aflatoxins G1, B2, and G2 are generally not detected in the absence of aflatoxin B1 and occur at lower quantities in food. The genotoxic carcinogenicity of aflatoxin M1, the main metabolite found in milk, has been found to be approximately 10 times lower than that of aflatoxin B1, and it was concluded that “AFMl is possibly carcinogenic to humans (Group 2B)” (IARC 1993; JECFA 2001).

EFSA’s Scientific Panel on Contaminants in the Food chain (CONTAM) (EFSA 2007, 2009) used a Margin of Exposure (MOE) in their risk assessment. A MOE of 10,000 or higher was calculated. It was based on the lowest benchmark dose level ($BMDL_{10}$) (10 % extra cancer risk) value of 170 ng/kg bw per day.

A risk assessment for aflatoxin in animal feed, with a focus on ensuring low levels of aflatoxin M1 in milk, was conducted by the EFSA CONTAM panel in 2004 (EFSA 2004e). The maximum aflatoxin content for a number of feeds have been defined (EU 2003).

The current permissible aflatoxin levels (0.005 mg/kg) in feeds for dairy cattle (EU 2003) ensure that the aflatoxin M1 level in the bulk milk supply is below 0.05 µg/kg, the maximum permissible level for aflatoxin M1 in milk in the EU (EU 2001).

Aflatoxins are also of considerable importance for warm-water farmed fish species. Aflatoxin contamination in feeds for farmed fish is a wide-spread problem in tropical regions due to the use of by-products that are frequently contaminated with aflatoxins, as well as to improper feed milling and storage procedures (Santacroce et al. 2008). Aflatoxin B1 (AFB1) was not specifically assessed for farmed finfish in the EFSA opinion from 2004 on aflatoxin B1 as an undesirable substance in animal feed (EFSA 2004a). The metabolism and toxicity of aflatoxins in aquatic species have been reviewed by Santacroce et al. (2008).

9.8.1 Mode of action of aflatoxins

In the liver, aflatoxin is transformed to its DNA-reactive form, aflatoxin-8,9-epoxide. This molecule can bind to liver proteins and inhibit their functionalities, potentially resulting in acute aflatoxicosis. Alternatively, it can bind to DNA, leading to aflatoxin-induced hepatocellular carcinoma. Aflatoxin B1 is mutagenic in bacterial systems and in eukaryotes. The G>T transversion mutation is observed predominantly (Groopman et al. 1996). The aflatoxin–DNA adduct is unstable and undergoes depurination, leading to its urinary excretion. Aflatoxins also bind to proteins such as albumin (AF-alb) via the formation of aflatoxin B1-8,9-dihydrodiol (Sabbioni 1990). There is a high correlation between the presence of aflatoxin-DNA adducts in the liver, their urinary excretion, and the formation of the serum albumin adduct.

9.8.2 Toxicokinetics of aflatoxins

Aflatoxins are rapidly absorbed in the rat small intestine (Ramos and Hernández 1996). Ingested aflatoxin B1 is metabolised to various metabolites, including the endo- and exo-epoxides of aflatoxin B1, the hydroxy-metabolites aflatoxin M1, aflatoxin M2 and aflatoxin M4, and aflatoxin P1, as well as glutathione-conjugated metabolites. The most important

metabolite in humans and animals is aflatoxin M1, the 4-hydroxy metabolite of aflatoxin B1 (Figure 19).

The liver is the major site of aflatoxin metabolism. Aflatoxin B1-exo-8,9-epoxide is further metabolised via multiple biochemical pathways, including conjugation to glutathione (aflatoxin B1-N7-Gua) and hydrolysis to 8,9-dihydrodiol (Essigmann et al. 1982), which is unstable and rearranges to a dialdehyde that reacts with proteins, such as albumin (Guengerich 2005). Aflatoxin B1-N7-Gua also undergoes sequential metabolism and is excreted as aflatoxin–N-acetylcysteine in urine (Kensler et al. 2005). There are significant species differences regarding the catalytic GST-activity, with mice having the highest conjugation rate (Kirby et al. 1993). For this reason, mice are highly resistant to aflatoxin B1-induced hepatocarcinogenesis, while turkeys, with a low GST activity, are sensitive to aflatoxins. Rainbow trout are highly sensitive to aflatoxin B1-induced carcinogenesis. The sensitivity of rainbow trout to aflatoxin B1-induced carcinogenesis has been related to their limited capacity for DNA repair, especially removal of epoxy DNA adducts, together with a high capacity for P450 activation, combined low glutathione transferase-mediated detoxification of the epoxide metabolites (Bailey et al. 1996; Santacroce et al. 2008).

Only aflatoxin B1, aflatoxin G1 and aflatoxin M1 can be bioactivated by CYPs to form reactive, carcinogenic 8,9-epoxides. Aflatoxin B1 metabolites can be useful biomarkers of human exposure to aflatoxins (Groopman et al. 1985).

In ruminants, a considerable proportion of ingested aflatoxin B1 is degraded in the rumen prior to absorption and does not reach the systemic circulation. The absorbed fraction of aflatoxin B1 is extensively metabolised in the liver, resulting predominantly in aflatoxin M1, which can be excreted in urine or milk. Aflatoxin M2 and aflatoxin M4 have also been detected in milk.

Toxicokinetic parameters have been determined in rhesus monkeys, rats, and mice showing tissue clearances, volumes of distribution, and half-lifes for aflatoxinB1 (Wong and Hsieh 1980). Toxicokinetic parameters in humans have been established using a micro-dosing study, with oral application in capsules of 30 ng ^{14}C-labelled aflatoxin B1 per person (average weight 95 kg) (Jubert et al. 2009). Uptake was fast and the terminal half-life long ($t_{1/2\beta}$ = 64.4 h). However, as total radioactivity was measured in this experiment, no distinction was made between aflatoxin B1 and its metabolites or conjugates.

Toxicokinetic studies in channel catfish showed incomplete absorption of aflatoxin B1 and preferential elimination via the bile (Plakas et al. 1991; Santacroce et al. 2008). In a study in channel catfish, 95 % of orally administered aflatoxin B1 became bound to plasma proteins (Plakas et al. 1991). Species differences in toxicokinetics of orally-administered ^{3}H-AFB1 appeared in comparisons of rainbow trout (*Oncorhynchus mykiss*) and tilapia (*Oreochromis niloticus*) (Ngethe et al. 1993). The toxicokinetics of dietary aflatoxin B1 were studied in rainbow trout kept in metabolic chambers and force-fed tritium (^{3}H)-labelled aflatoxin B1 daily for 7 days (Ellis et al. 2000). Urine and bile were the main excretion pathways (14 % and 44.8 % of the total dose, respectively). Comparison of intravenous and oral kinetics indicated a high degree of gastrointestinal absorption (Ngethe et al. 1992). The highest concentrations were observed in the bile, liver, kidney, pyloric caeca, uveal tract of the eye, and the olfactory rosette, partly as a major fraction of this covalently bound metabolites (Ngethe et al. 1992). Retention and metabolism in extrahepatic tissues might be of importance regarding the toxicologic potential of aflatoxin B1 in rainbow trout (Larsson et al. 1992).

9.8.3 Effects of aflatoxins in domestic animals

Aflatoxin B1 is a strong acute toxin in various animal species (Eaton and Groopman 1994). Differences in susceptibility are related to epoxide formation rates and detoxication ratios. Clinical signs include anorexia, icterus, depression, weight loss, nasal discharge, gastrointestinal effects, haemorrhages, ascites, and pulmonary oedema (Newberne and Butler 1969). Bovine species are generally less sensitive than non-ruminants due to microbial degradation in the rumen. Sheep appear to be especially resistant to aflatoxin, and cases of acute aflatoxicosis have not been reported in sheep. Tumour formation due to aflatoxin B1-contaminated feed has not been reported from animals kept under European farm conditions.

Table 36. Comperative pathology in animals fed aflatoxin-contaminated feed, table is modified from Wogan (1966).

Liver lesions	Calves	Cattle	Swine	Sheep	Duckling	Adult duck	Turkey poult	Chick
Acute necrosis and haemorrhage	-	-	+	-	+	-	+	-
Chronic fibrosis	+	+	+	0	-	+	-	-
Regeneration nodules	-	+	+	0	±	+	+	-
Bile duct hyperplasia	+	+	+	0	+	+	+	±
Veno-occlusive disease	+	+	-	0	-	-	-	-
Enlarged hepatic cells	+	+	+	0	+	+	+	-
Liver tumors	0	0	0	0	-	+	0	0

+ reaction, ± week reaction, - no reaction, 0 not tested

Pig

Barrows fed 0.25 or 0.5 mg aflatoxin/kg feed (as a rice culture of *A. parasiticus* mixed into the feed), had increased levels of blood urea nitrogen (BUN) and aspartate amino transeferase (AST) as well as a decreased performance (Rustemeyer et al. 2010). These results are in agreement with the conclusions of previous reviews that a threshold for moderate effects on performance in pigs is slightly above 0.2 mg aflatoxin B1/kg feed (Osweiler and Ensley 2012). Possible immunosuppression has been reported from pigs at these levels. Effects on clinical parameters, such as obvious immune suppression, liver lesions, and cholangiohepatitis, are evident at concentrations from 0.4 mg/kg feed.

In sum, no effects have been reported in pigs fed diets of aflatoxin below 0.2 mg/kg feed, which may be considered as a NOEL.

Poultry

Aflatoxins were first discovered when they were shown to cause “Turkey X disease” that caused widespread deaths of turkeys and other poultry in Europe in the 1960s (Smith 1960). There are species differences in sensitivity to aflatoxins among poultry, with ducklings being most sensitive and broiler chicken considerably less sensitive (Plumlee 1994; Hoerr 2008; Rawal et al. 2010). Ducks were shown to develop hepatic lesions following dietary exposure to 0.030 mg aflatoxin/kg feed, turkeys at exposure to 0.3 mg/kg feed, and broiler chickens at

0.5 mg/kg feed (Coker 1979). Arafa et al. (1981) showed that 0.7 mg aflatoxins/kg feed reduced growth rate in turkeys, but not in broiler chickens, and Rauber et al. (1987) showed a decreased feed consumption in turkey poults given from 0.1 mg aflatoxins/kg feed. No lower doses were used. A mixture of aflatoxins B1, B2, G1 and G2 also affected the kidneys and increased the urinary secretion of salts from 0.5 mg/kg feed. No lower doses were used (Martinez-de Anda et al. 2010).

Aflatoxins cause delayed reduction in egg production and hatchability, but these effects occur at higher concentrations than those that have effects on feed intake, growth, and the liver (review in Bryden 2012).

In sum, aflatoxicosis in poultry, characterised by hepatotoxic effects, has been demonstrated in ducks following dietary exposure to 0.030 mg aflatoxin/kg feed, turkeys at exposure to 0.3 mg/kg feed, and broiler chickens at 0.5 mg/kg feed (LOAELs). Decreased feed consumption occurs in turkeys exposed to from 0.1 mg aflatoxins/kg feed.

Ruminants

EFSA evaluated aflatoxins in feed with special emphasis on their potential for transfer to milk. EFSA concluded that at the current limit of 0.005 mg/kg feed for dairy cattle, animal health will be protected and also the concentrations of aflatoxin M1 in bulk samples of milk will be generally ensured to be below the current maximum limit of 0.05 µg aflatoxin/kg consumable milk (EFSA 2004e). Effects in cattle occur at levels from 1.5 – 2.2 mg/kg feed and in small ruminants after exposure to > 50 mg/kg feed (Miller and Wilson 1994).

Horse

Some case reports of suspected aflatoxin poisonings of horses have been published in the literature, with clinical and pathological findings similar to the hepatotoxic effects of aflatoxins in other species (Vesonder et al. 1991; Angsubhakorn et al. 1981; Hasso 2003). Aflatoxins or aflatoxin-producing *A. flavus* have been found in the feed related to these cases, but the reports do not provide any dose-reponse information.

In addition, a few experimental studies on horses have been reported (Bortelli et al. 1983; Cysewski et al. 1982). Serum activity of the enzyme gamma-glutamyl transpeptidase (GGT) was increased in ponies given single doses of 0.5 mg/kg bw og higher, while alanine aminotranserferase (ALT) increased in ponies exposed to 4 mg/kg bw. Ponies receiving single doses of 4 mg/kg bw died, while 50 % of ponies fed 2 mg/kg bw died within 48 h (Bortelli et al. 1983).

In sum, alterations in clinical chemistry related to hepatic damage have been reported from horses given a single dose from 0.5 mg/kg bw (approximately 0.01 mg/kg single dose).

Rabbit

The effects of aflatoxins in rabbits were reviewed by Mézes and Balogh (2009). Like most other species, rabbits are sensitive to aflatoxins, and the liver is the primary target. The focus of recent feeding studies has been on the protective effects of a range of additives, such as antioxidants or binders (e.g. Salem et al. 2001; Sur et al. 2012) and feed treatments such as hydroxyperoxide or irradiation (e.g. Soliman et al. 2001). In these studies, only one feed concentration of aflatoxins has been used and effects on the studied parameters have been shown at these concentrations. No effect levels can be established from these studies.

Karakilcik et al. (2004) showed indications of liver damage with increased activity of several liver enzymes in rabbits given 0.1 mg aflatoxin B1 in the diet for 10 weeks,. The effects were partially mitigated by addition of vitamins C and E. Similarly, aflatoxins also caused liver damage, such as degenerated hepatocytes, bile duct epithelial hyperplasia, and hypertrophy, as well as ultrastructural alterations in the hepatocytes (Prabu et al. 2013). Significant effects on feed intake and growth, increased mortality (25 %), increased serum enzyme activity (alanine amino transferase and aspartate amino transferase) and histopathological damage were reported from growing New Zealand white rabbits given 0.83 mg aflatoxins/kg feed for 6 weeks (Soliman et al. 2001).

Other studies in which rabbits were given daily oral doses from 0.05 mg/kg bw confirm that liver damage is induced by aflatoxins in rabbits (Prabu et al. 2013; Guerre et al. 1996).

In sum, the available studies do not allow establishment of a safe level of aflatoxins in feed, but effects on the liver have been reported from rabbits given 0.1 mg aflatoxin B1/kg feed for 10 weeks.

Dog and cat

The LD_{50} for aflatoxins in dogs is low (0.5-1.8 mg/kg bw). There have been several reports of intoxications in dogs from consumption of aflatoxin-contaminated feed (e.g. Bruchim et al. 2012; Newman et al. 2007; Stenske et al. 2006). Levels of 0.2 – 0.6 mg/kg feed have been found in these cases.

Although it is rarely possible to know the precise aflatoxin exposure resulting from consumption of contaminated feed, previous reviews of such cases conclude that dog food containing levels of aflatoxin as low as 0.06 mg/kg feed has been implicated in cases of aflatoxicosis in dogs (reviews in Bischoff and Garland 2009; Adams and Bischoff 2011), while feed concentrations below this level have not been associated with aflatoxicosis in dogs.

Young adult beagle dogs, 2 females and 1 male per group, were given 1, 5, or 20 µg aflatoxin/kg bw (estimated to be approximately 0.03, 0.2 and 0.7 mg/kg diet) with an aflatoxin mixture (in %: B_1, 37.5; B_2, 5.4; G_1, 17.1; G_2, 1.0) for 5 days per week for 10 weeks. Clinical signs attributable to aflatoxicosis were generally absent among the dogs given 1 and 5 µg/kg bw. Icterus, inappetence, yellow-orange urine, and an increase in prothrombin time were noted in dogs fed 20 µg/kg bw. Moderate bile duct proliferation, bile pigment accumulation in the portal areas, and multiple vascular channels around the central and portal veins were shown in the livers from this group (Armbrecht et al. 1971).

In sum for dog, a NOEL is identified at 0.2 mg/kg diet based on liver toxicity at higher dosage levels.

There is very little available information from cats, and no natural cases of aflatoxicosis in cats are known. The LD_{50} for cats is low and at the same levels as for dogs (0.55 mg/kg bw).

Fish

There is a species-specific sensitivity to alfatoxin-initiated tumour formation among finfish. Rainbow trout are more sensitive to aflatoxin exposure than channel catfish (Abd-Allah et al. 1999) or other salmonid species such as coho salmon (*Oncorhynchus kisutch)* (Coulombe et al. 1984). Hepatocytes from rainbow trout that were activated by aflatoxin B1 showed a higher rate of mutagenesis in a *Samonella typhimurium* culture compared with aflatoxin B1-activated hepatocytes from coho salmon (Coulombe et al. 1984). Similarly, a single i.p. injection of 0.5 mg aflatoxin B1/kg caused DNA damage in the blood, kidney and liver of

rainbow trout, but not in channel catfish (Abd-Allah et al. 1999). Based on the tumour and general responses, species-specific sensitivity of fish to aflatoxins has been suggested, with rainbow trout the most sensitive species followed, in descending order, by coho salmon, Nile tilapia, and channel catfish (Han et al. 2010; Tuan et al. 2002; Deng et al. 2010; Chavez Sanchez et al. 1994; Jantrarotai and Lovell 1990; Manning et al. 2005b). Information on the relative sensitivity to dietary aflatoxin exposure is lacking in the cold-water species, Atlantic salmon. The liver is the primary organ for aflatoxin-induced carcinogenesis in rainbow trout, independent of the route of exposure (Bailey et al. 1996). Liver tumour formation associated with aflatoxins was reported in a hatchery in the USA producing 2-3 year old brood-stock trout (Ellis et al. 2000), and outbreaks of rainbow trout aflatoxicosis have been reported in Germany (Wunder and Korn 1982) and Denmark (Rasmussen et al. 1986; Santacroce et al. 2008). In rainbow trout, tumours can take a year or longer to develop, with initiation at 20 µg/kg feed, even after temporary feeding during early life stages (Coulombe et al. 1984). Also exposure of developing trout eggs to aflatoxin by simple immersion in solution (0.5 ppm for 0.5 hours) resulted in the development of a significant number of hepatocellular carcinoma (HCC) in rainbow trout fingerlings 9 months after exposure.

Dietary concentrations of aflatoxins in the low (~1-20 µg/kg) range, fed continuously for 9 - 18 months or higher doses (~10-80 µg/kg) fed continuously for shorter periods of time (1-30 days) have been shown to be carcinogenic for rainbow trout (Bailey et al. 1996). The lowest reported dietary concentration that causes hepatoma is 0.4 µg aflatoxin B1/kg, resulting in HCC development at a rate of 14 % after exposure to the contaminated feed for 15 months (Lee et al. 1968).

The possible immunosuppressive effect of aflatoxin B1 is of great importance for farmed animals (EFSA 2004a). Aflatoxins are known to impair the cellular and humoral immune systems, rendering animals more susceptible to bacterial, viral, fungal and parasitic infections. This immunosuppressive effect also impairs acquired resistance following vaccination, and may occur at sub-clinical levels of intoxication (EFSA 2004a). Similarly, embryo-exposed rainbow trout experienced long-term immune dysfunction after waterborne exposure (0.5 mg L^{-1}) (Ottinger and Kaattari 2000), and early *in vitro* and *in vivo* aflatoxin B1 exposures caused antibody responses in rainbow trout (Arkoosh and Kaattari 1987). No dietary NOEL could be defined regarding the immunosuppressive effects for farmed Atlantic salmon.

In sum, the lowest reported dietary concentration that causes hepatoma in rainbow trout is 0.4 µg aflatoxin B1/kg when fed contaminated feed for 15 months.

Table 37 Critical effect (most sensitive endpoint) for aflatoxin in feed at concentrations where NOAEL or other available effect limits are derived/identified for various species

Animal	Critical Effect	Concentration in feed	Available effect limit	Pivotal references
Pig	Decreased performance	0.2 mg/kg	NOEL	Osweiler and Ensley (2012)
Duck Turkeys Broiler chicken	Hepatotoxicity Hepatotoxicity Feed consumption Hepatotoxicity	0.03 mg/kg 0.3 mg/kg 0.1 mg/kg 0.5 mg/kg	LOAEL LOAEL LOAEL LOAEL	Coker (1979); Arafa et al. (1987); Rauber et al. (1987)
Ruminants	Transfer to milk	0.005 mg/kg	NOEL	EFSA (2004e)
Horse	Biomarker on hepatic reaction	Approximately 0.01 mg/kg (single dose)	Effective dose (one dosage level	Bortelli et al. (1982)
Rabbit	Hepatotoxicity	0.1 mg/kg	Effective dose (one dosage level)	Karakilcik et al. (2004)
Dog	Hepatotoxicity	Approximately 0.2 mg/kg	NOAEL	Armbrecht et al. (1971)
Rainbow trout	Hepatoma	0.0004 mg/kg	LOEL	Lee et al. (1968)

9.8.4 Carry-over of aflatoxins to humans through animal-derived food products

The exposure of lactating animals to aflatoxin B1 leads to the appearance of the hydroxy-metabolite aflatoxin M1 in the milk. A carry-over rate of 1-2 % has been estimated (Van Egmond 1989), depending on external factors such as feeding regime, health status, and actual milk production. For high-yielding dairy cows consuming large quantitities of feed concentrates, carry-over rates as high as 6.2 % have been reported (Veldman et al. 1992).

Chickens fed aflatoxin B1-containing feed had elevated toxin levels in breast muscles and livers (Madden and Stahr 1995; Hussain et al. 2010). Clearance from the tissue after withdrawal of aflatoxin B1 from the feed was slow. Eggs from laying hens fed with 3310 µg aflatoxin B1 and 1680 µg aflatoxin B2 /kg feed for 28 days contained aflatoxin. A maximum level in the eggs of less than 0.5 µg total aflatoxins/kg were reached after 5 days of feeding, and this remained relatively constant throughout the remaining feed period, but declined rapidly with toxin-free feed (Wolzak et al. 1985). Aflatoxin M1 was found in the eggs in addition to the administered aflatoxins.

Aflatoxin and metabolites are transferred to eggs. In experimental studies with laying hens fed 8 mg aflatoxins B1/kg feed for 7 days, the levels of aflatoxin B1 and the metabolite aflatoxicol increased for 4-5 days, and then levelled out at 0.02-0.2 µg/kg. After withdrawal of the contaminated feed, the levels decreased at the same rate at which they had increased, and only traces of aflatoxins B1 and aflatoxicol were found in eggs after 7 days on an aflatoxin-free diet (Truckness et al. 1983). As aflatoxins are regarded as potent genotoxic compounds, human exposure should be minimised. The levels of aflatoxins in the feed of laying hens should also therefore be kept as low as possible. VKM noted, however, that high levels of aflatoxins were used in the feed in this study; such high levels are unlikely to occur in Norwegian feed under normal circumstances. Despite the high concentrations used in the experimental feed, the levels in the eggs were well below the maximum limit for other food items such as grains and nuts.

Sea bass (*Dicentrarchus labrax* L.) administered with 18 µg aflatoxin B1/kg BW orally for 42 days had residue levels of around 5 µg/kg in fish muscle (El-Sayed and Khalil 2009). In aquacultured channel catfish at least 400 µg/kg residues of aflatoxin were recovered (Santacroce et al. 2008). In rainbow trout, the carry-over from feed to carcass was estimated to be approximately1.3 µg/kg (w/w) when fish were force-fed a diet of 20 µg/kg feed daily (Ellis et al. 2000). The estimate is based on the ^{3}H levels in feed and carcass, where the carcass consists of the whole fish minus body fluids and internal organs. In a study on channel catfish, a very low potential was found for the accumulation of aflatoxin B1 and its metabolites in the edible flesh through the consumption of aflatoxin B1-contaminated feed (Plakas et al. 1991).

9.8.5 Effects of aflatoxins in humans and animals relevant for human risk assessments

Aflatoxin B1 has been shown to be both a potent genotoxin and carcinogen in humans and experimental animals such as mice, rats, hamsters, fish, ducks, and monkeys. The primary target is the liver, but tumours have also been found in other organs, including the kidneys, gall bladder, and colon. Experimental evidence is also available regarding the carcinogenicity of naturally occurring mixtures of aflatoxins, and of aflatoxin G1 and aflatoxin M1, whereas there is only limited evidence for aflatoxin B2 and inadequate evidence for the carcinogenicity aflatoxin G2 (JECFA 1998; IARC 1993). A linear dose-response relationship has been demonstrated for aflatoxin B1 in at least two animal species, down to doses of less than 0.1 pg/kg bw/day.

The immunotoxicity of the aflatoxins, probably caused by disturbances of the intestinal integrity or modulation of cytokine expression (Gong et al. 2002), may be one explanation for stunted growth in children (Denning et al. 1995). Impairment of the cellular and humoral immune system has also been shown to render animals more susceptible to bacterial, viral, fungal and parasitic infections.

9.8.6 Human hazard characterizations of aflatoxins

No TDI or similar levels for safe intake have been established, as a NOAEL cannot be determined for the carcinogenic potential of aflatoxins. EFSA's Scientific Panel on Contaminants in the Food Chain (CONTAM) (EFSA 2007, 2009) used a Margin of Exposure (MOE) in their risk assessment and a lowest benchmark dose level ($BMDL_{10}$) (10 % extra cancer risk) value of 170 ng/kg bw per day as a reference point for the calculation of the MOE. EFSA calculated a MOE of 10,000 or higher.

9.9 Examples of other toxic fungal metabolites present in cereal grain

A range of other fungal metabolites are probably present in cereal grain. A large variety of *Fusarium* toxin metabolites have been described, but both the occurrence and the toxicity of these metabolites have been the subjects of very few studies and cannot be evaluated.

9.9.1 Alternaria toxins

Fungi of the genus *Alternaria* are also widespread in Norwegian grain (Kosiak et al. 2003). A large variety of *Alternaria* toxin metabolites have been described, but knowledge about their

occurrence and toxicity is very limited. EFSA evaluated *Alternaria* toxins in 2011 and concluded that knowledge on the toxic effects of *Alternaria* toxins on animals was insufficient to establish safe intake levels. Furthermore, EFSA concluded that there are indications that alternariol and tenuasonic acid may be of concern for poultry health (EFSA 2011c). Data on their occurrence in cereals in Norway is lacking. EFSA used a "Threshold of toxicological concern" (TTC) in their assessment of human health risk. For the genotoxic *Alternaria* toxins, alternariol, and alternariol monomethyl ether, it was concluded that the estimated chronic dietary exposure exceeded the relevant TTC value, indicating a need for additional toxicity and occurrence data. The estimated dietary intake of the non-genotoxic tentoxin and tenuazonic acid were lower than the relevant TTC value and were considered by EFSA to be unlikely to be of human health concern.

9.9.2 Ergot alkaloids

Only low frequencies of ergot are generally thought to occur in harvested cereals in Norway because of the effective removal of ergots from the grain during the harvest and cleaning of the crop. In Europe *Claviceps purpurea*, known to infect 400 plant species, is the dominating species of Claviceps. Rye is the most exposed small cereal grain but also wheat, barley and oats can be infected. The fungi produce a range of bioactive compounds, ergot alkaloids. EFSA (2012) has concluded that chemical analysis should cover the most common compounds: ergometrine, ergotamine, ergosine, ergocristine, ergocryptine (α- and β-isomers) and ergocornine, as well as their corresponding –inine epimers which are regarded inactive but easily transformed to the active forms.

Ergot alkaloids show moderate acute toxicity. Sublethal acute exposure produce neurotoxicity including restlessness, muscular weakness, tremor and rigidity. Repeated exposure inhibits blood circulation in particular in extremities, reduces body weight, and changes som hormone levels resulting in reproductive effects as interfered implantation, embryo toxicity and reduced lactation. Studies in rats indicate a similar potency of examined toxins (ergotamine, ergometrine, α-ergocryptine) with a NOAEL at 0.22-0.60 mg/kg bw per day. Vasoconstrition in the tail is the shown critical effect in rat with a benchmark dose level (BMDL10) at 0.33 mg/kg bw per day for ergotamine. EFSA (2012) has estimated a human acute reference dose (ARfD) at 1 µg/kg and a TDI at 0.6 µg/kg bw per day. As no significant difference in potency is revealed the ARfD and TDI given for the sum of ergot alkaloids.

As ergot and ergot toxins occur primarily in rye with low use as animal feed, the risk for effects in domestic animals is restricted. There are som effect data of ergot alkaloids in domestic animals. In piglets are shown reduced feed intake (critical effect) which starts at 3.6-11 mg/kg feed in different studies (EFSA 2012). In broiler chicken are shown intestinal changes at 2.8 mg/kg feed and above, whereas older poultry are considered to be less susceptible.

9.10 Effects of exposure to mixtures of mycotoxins

Fungi are usually able to produce more than one toxin and grain is contaminated with several fungal species simultaneously. Humans and animals are therefore exposed to combinations of mycotoxins. Combined effects can be synergistic, additive, less than additive, antagonistic, or one substance may potentiate the effects of other substances. It is known that some types of interaction may be dose-dependent (VKM 2008). In food and feed, maximum limits are set to ensure that the level of each toxic substance is below the threshold of toxicological effects, or

at a level of acceptable risk for substances for which no threshold for effect can be determined. The interactions of interest for a feed and food safety aspect are therefore those occurring at doses below the threshold of toxicological effects. Therefore, to be of relevance for risk assessment of mycotoxins in feed and food, it is crucial that studies of combined mycotoxin effects are conducted using levels of each component in the mixtures at, or below, their respective threshold values for toxicological effect.

A range of studies on the combined effects of mycotoxins *in vitro* have been published, including by Bernhoft et al. (2004), Thuvander et al. (1999), Bouaziz et al. (2012), and Ficheaux et al. (2012). These studies revealed several types of interactions, ranging from antagonistic to synergistic. It is, however, difficult to extrapolate from these *in vitro* studies to an *in vivo* situation.

A range of feeding studies with combinations of different mycotoxins have also been published, including combinations of DON and T-2 in pigs (e.g. Friend et al. 1992) and hens (Kubena et al. 1989), T-2 and ochratoxin A in pigs (Harvey et al. 1994), T-2 and diacetoxyscirpenol in hens (Hoerr et al. 1981; Diaz et al. 1994), DON and zearalenone in pigs (Trenholm et al. 1988) and mice (Forsell et al. 1986), DON and fusaric acid (Smith et al. 1997), and nivalenol and aflatoxins B1 in rats (Ueno et al. 1992).

A meta-analysis of studies of *in vivo* interactions of combinations of mycotoxins has been published (Grenier and Oswald 2011). All types of interactions were found, ranging from synergistic to antagonistic. Most studies showed a synergistic or additive outcome on animal performance parameters such as feed consumption, weight gain, and egg production, whereas the measurements of other factors, including biochemical ones, led to other types of interactions, ranging from synergistic to antagonistic, for the same combination of toxins, and even in the same study. Even for animal performance, less than additive and antagonistic responses were reported from some studies. Unfortunately, high doses with clear effects from each single toxin in the combination dose were used in most studies. Very few studies have been published that investigate the effects of combinations of low doses of the toxins, representative for field situations.

It is therefore currently not possible to reach a conclusion on the potential effects from exposure to combinations of mycotoxins.

9.11 Summary of human hazard characterizations

The tolerable intakes and critical effects of the toxins for which the dietary intake has been estimated are given in Table 38.

Table 38. Summary of mycotoxins for which human exposure estimates or exposure scenarios have been made, their NOAEL/LOAEL values in animal studies, critical effects in animals and the tolerable daily intakes (TDIs) given.

Mycotoxin	Critical effect	NOAEL/LOAEL[1]	Tolerable intake[2]	Reference
DON	Reduced feed intake and weight gain	0.1 mg/kg bw/day (NOAEL)	1.0 µg/kg bw	JECFA 2011; SCF 1999
T-2 + HT-2	Immunotoxicity/haemato toxicity	0.03 mg/kg bw/day (LOAEL)	0.1 µg/kg bw/day	EFSA 2011a
Nivalenol	Reduced growth, leucopenia	0.7 mg/kg bw/day (LOAEL)	0.7 µg/kg bw/day	SCF 2000b
Zearalenone	Oestrogenic activity	10.0 µg/kg bw/day (NOAEL)	0.25 µg/kg bw/day	EFSA 2011b

[1] NOAEL – no observed adverse effect level, LOAEL – lowest observed adverse effect level.
[2] Tolerable daily intake, TDI, is a term used by EFSA, while JECFA uses the term provisional maximal daily intake (PMTDI)

Ochratoxin A was not included in the human risk characterisation because the majority of dietary intake is from non-grain food. Evaluation of the total ochratoxin A dietary intake is not within the terms of reference for the present risk assessment. Furthermore, the available data on ochratoxin A occurrence in Norway are limited to crude grains (Tables 8, 9, and 10), and no data are available for food products (Tables 17, 18, and 19).

Aflatoxins were not included in the present risk assessment, for the same reasons to those given for ochratoxin A.

The dietary intake of fumonisins could not be estimated due to few occurrence data. The existing data (Table 19) confirm that fumonisins are of little importance in Norway. Addionally, fumonisins occur almost exclusively in maize, and human consumption of maize in Norway is very limited.

No hazard characterisation could be made for moniliformin, enniatins, or beauvericin due to the lack of occurrence data in foods and insufficient toxicological data.

10 Risk characterisation of mycotoxins

10.1 Risk characterisation of mycotoxins in animal feed

The risk characterisation of the mycotoxins in feed combines the data from the hazard characterisation for each type of animal with the corresponding data for exposure. The results are characterisation of risk for the various mycotoxins and the various animal species or production groups, and which may be negligible, low, moderate or high. DON in pig feed is the only combination which is described as **high risk**, where the mean and median DON concentrations are at levels that may reduce animal performance and welfare. For several combinations, the term 'negligible risk' is used, indicating a lack of risk, or close to no risk, for mycotoxin effects. Low risk implies a certain, but quite unlikely, risk, while moderate risk implies that the mycotoxin exposure sporadically may occur above the level considered to result in effects. The available effect levels, NOAELs, NOELs, LOAELs, LOELs, benchmark doses or other thresholds, are derived from other reviews/assessments or are identified by the authors of the present assessment based on the collected effect data from original studies as referenced.

Particular focus on DON, T-2 and HT-2

For DON and T-2 and HT-2, which constitute the main mycotoxin problem in Norwegian cereals, a relatively extensive database is available, both on effects and on occurrence in feed cereals, and there are also some data on prepared compound feed. Thus, for these trichothecenes it is possible to characterise the risk relatively precisely for several animal species.

The occurrences of DON and T-2 and HT-2 in compound feed for the various species, are based on predicted data from measured concentrations in cereals combined with data on normal feed compositions, as well as based on measured (observed) data on these toxins in prepared compound feed when available (see Chapter 6).Various available levels for effect (NOAELs, NOELs, LOAELs, LOELs, effect thresholds as benchmark dose) are taken from the hazard characterisation (see Chapter 9). The predicted or observed percentages, respectively, of feed above these levels are presented in Tables 39-42. For DON in pigs, NOAELs and LOAELs have been identified within broad ranges. The lowest LOAEL is 0.35 mg/kg diet and the lowest NOAEL is 0.6 mg/kg diet, based on reduced feed intake. For risk assessment, it is reasonable to use the available effect levels at the lower ends of these ranges, provided that the studies are well conducted and the results are reliable, as is the case here. Based on the LOAEL for DON at 0.35 mg/kg diet, the predicted frequencies of compound feed for pigs above this level varies from 4 - 13 % (Table 39). The observed frequency above this level, based on the measured concentrations in all samples of feed for pigs, is 53 %. Based on the other available effect levels for pigs, correspondingly lower frequencies of feed will be above the effect level. For other animal species, the rates of exceeding the available effect levels are lower than for pigs.

For T-2 and HT-2, the available effect levels for pigs are more similar to corresponding effect levels for other species. Thus, sensitivity to these toxins seems more similar among different animal species. Based on the benchmark dose of T-2 and HT-2 in pigs at 0.17 mg/kg diet, the predicted frequencies of compound feed exceeding this level vary from 0.3 – 1 %. The observed frequency of exceeding this level, based on the measured concentrations in compound feed for pigs, is 3 – 4 %. The available effect level in ruminants at 0.1 mg/kg diet

indicates that the general tolerance of ruminants for several mycotoxins does not include T-2 and HT-2.

Table 39. Predicted frequency of feed with DON concentrations above available effect levels (LOAELs, LOELs, NOELs,) derived from prevous risk assessements or identified for the present assessment based on typical recipes for cereals and observed toxin levels in wheat, barley and oats.

	LOAELs		NOAELs	
	mg/kg	% above	mg/kg	% above
Piglet A	LOAELs 0.35-2	13-0.6	NOAELs 0.6-0.9	6-3
Piglet B	LOAELs 0.35-2	9-0	NOAELs 0.6-0.9	4-2
Pig growth A	LOAELs 0.35-2	4-0	NOAELs 0.6-0.9	1-0.3
Pig growth B	LOAELs 0.35-2	9-0.4	NOAELs 0.6-0.9	4-2
Sow A	LOAELs 0.35-2	7-0	NOAELs 0.6-0.9	3-1
Sow B	LOaELs 0.35-2	8-0	NOAELs 0.6-0.9	3-1
Broiler chicken	LOAEL 1.7	0.6		
Laying hen	LOAEL 12	0		
Ruminant			NOEL 6	0
Horse	LOAEL 5	0.5		
Dog	LOAEL 2.7	0		

Table 40. Observed frequency of samples with DON concentrations above available effect levels (LOAELs, LOELs, NOELs), derived from previous risk assessements or identified for the present assessment in feed analysed at the Norwegian Veterinary Institute and, samples of pig feed analysed by the industry (all results grouped in feed for pig (all)).

Feed type	N	LOELs/LOAELs		NOAELs	
		mg/kg	% above	mg/kg	% above
Feed for pig (all)	309	LOAELs 0.35-2	53 -0	NOAELs 0.6-0.9	22 -10
Feed for pig	131	LOAELs 0.35-2	36 -0	NOAELs 0.6-0.9	15 -7
Feed for poultry	53	LOAEL Broiler chicken 1.7 LOAEL Laying hen 12	2 0		
Feed for horse	16	LOAEL 5	0		
Feed for dog	24	LOAEL 2.7	0		
Feed for ruminants	3			NOEL 6	0

Table 41. Predicted frequency of feed with T2 + HT2 concentrations above available effect levels (BMDLs, LOAELs, NOAEL) derived from prevous risk assessements or identified for the present assessment based on typical recipes for cereals and observed toxin levels in wheat, barley and oats.

	BMDL		LOAELs		NOAEL	
	mg/kg	% above	mg/kg	% above	mg/kg	% above
Piglet A	0.17	1	0.5	0.2		
Piglet B	0.17	0.6	0.5	0		
Pig growth A	0.17	0.1	0.5	0		
Pig growth B	0.17	1	0.5	0.1		
Sow A	0.17	0.3	0.5	0		
Sow B	0.17	0.5	0.5	0		
Duck			0.2	0.5		
Broiler chicken			0.5	0.1		
Laying hen			2	0.5		
Sheep					0.1	3
Horse			0.7	0.5		

Table 42. Observed frequency of samples with T2 + HT2 concentration above available effect levels (BMDLs, LOAELs, NOAEL) derived from prevous risk assessements or identified for the present assessment in feed analysed at the Norwegian Veterinary Institute and also, samples of pig feed analysed by the industry (all results grouped in feed for pig (all)).

Feed type	N	BMDL		LOAELs		NOAEL	
		mg/kg	% above	mg/kg	% above	mg/kg	% above
Feed for pig (all)	175	0.17	3	0.5	0		
Feed for pig	131	0.17	4	0.5	0		
Feed for poultry Duck Broiler chicken Laying hen	53			 0.2 0.5 2	 9 0 0		
Feed for horse	16			0.7	0		
Feed for ruminants	3					0.1	33

10.1.1 Risk characterisation of mycotoxins in feed for each animal species (terrestrial and aquatic)

10.1.1.1 *Pig*

Deoxynivalenol (DON)

The range of estimated DON concentrations in pig feed, based on measured concentrations in grain combined with the amounts of grain used in the feed recipes, as well as the measured concentrations in prepared pig feed, shows that pigs may be exposed to DON above those levels often shown to reduce their feed intake. However, the lowest effective level varies considerably (we identified LOAELs from 0.35-2 mg/kg feed) in the various studies, largely due to unknown factors. These factors may include other non-measured mycotoxins and other chemical compounds in the feed, the pigs' environment, and their breed. Reduced feed intake and secondary effects, such as reduced weight gain and stress, are the critical effects of DON in pigs. Other effects, such as reduced immunity, occur at much higher exposure levels.

From the estimates of DON in pig feed, based on concentrations in grain and the grain content in recipes, mean DON exposure is below the lower end of the range for critical effects in all recipes. Most of the estimated 95-percentiles for DON in the recipes are, however, within the

range of observed effects. It appears that exclusion of oats from the recipes reduces the risk of producing pig feed with the potential for causing DON-related health problems. Chemical analysis of DON in prepared pig feed has demonstrated that even the mean and median DON concentrations may impact on pig feeding behaviour.

That higher DON levels are found by chemically analysing compound pig feed than by estimating the levels from the analysed ingredients of grains may be due to the fact that the analysis results from compound feed are from recent years only, whereas those from grains are from the last 15 years. Other reasons for this diffence could be due to DON occurring in ingredients other than cereal grains in the chemically analysed samples of compound feed. It is known that maize may be a source of *Fusarium* toxins such as DON. However, maize is usually not included in Norwegian compound feed for pigs, or only small amounts (up to 5 %) are used.

In sum, the lowest effect level that results in reduced feed intake varies considerably between studies (identified LOAELs range from 0.35 to 2 mg/kg feed). There is a high risk of performance and welfare effects in pigs due to exposure to DON via feed produced by commonly used recipes from cereals. The risk increases with the amount of oats in the recipes.

T-2 and HT-2

For T-2 and HT-2, the estimated levels in compound feed for pigs, based on measured concentrations in the crude cereal grains and the measured concentrations in prepared compound feed for pig, are more similar to each other than was found for DON. These exposure data, combined with the effect data for T-2 and HT-2, indicate that the risk for effects in pigs due to these toxins is lower than for DON. Most of the pig feed seems to be below the level at which adverse effects are expected. A LOAEL for T-2 is derived at 0.5 mg/kg diet for the critical haematological and immunological effects, and, via benchmark modelling performed by EFSA, this corresponds to a $BMDL_{05}$ at 0.17 mg/kg diet. This estimated benchmark threshold for adverse effects is at approximately the 99-percentile for estimated and measured HT-2 + T-2 concentrations in compound feed for pig concentrate. As for DON, the risk of T-2 and HT-2 induced health problems in pigs increases with the amount of oats in the pig formulations.

In sum, most of the currently available pig feed seems to be below the derived LOAEL at 0.5 mg/kg diet for critical haematological and immunological effects, and the benchmark dose level ($BMDL_{05}$) of 0.17 mg/kg diet. The risk for adverse effects in pigs is considered low. The risk related to T-2 and HT-2 exposure increases with the amount of oats in the pig formulations.

Nivalenol

In Norway, only trace amounts of nivalenol occur in cereal grains, slightly above the quantification limit. The levels are far below those levels associated with effects in pigs (2.5 mg/kg diet). The risk for adverse effects is considered negligible.

Zearalenone

A NOAEL of zearalenone in female pigs at 0.2 mg/kg diet is derived. Zearalenone is produced by the same *Fusarium* species that produces DON, and may occur in small grain cereals. Levels are usually highest in oats, with yearly means up to 0.18 mg/kg measured

during 2005-2009. Zearalenone has been suspected to cause reproductive disorders in sows in Norway. The current restricted use of oats reduces the risk of zearalenone effects in pigs. Maize may also be a source of *Fusarium* toxins such as zearalenone. However, maize is usually not used, or only small amounts (up to 5 %), in Norwegian compound feed for pigs. Norwegian data on zearalenone in maize are lacking.

In sum, the risk for adverse effects of zearalenone in pigs is considered low, primarily due to the currently limited use of oats and maize in pig feed.

Fumonisins

Adverse lung effects due to fumonisin B1 have been shown at doses of 5 mg/kg feed and above, with a derived NOAEL of 1 mg/kg diet. There are indications of connective tissue proliferation in the lungs and reduced performance and carcass quality in pigs fed 1 mg fumonisin B1/kg diet, which identifies a NOAEL at 0.33 mg/kg diet. Fumonisins are contaminants of concern in maize. Fumonisin concentrations in maize may reach several mg/kg and the risk for adverse effects is associated with an increased use of maize in pig feed. In Norway, maize is not used, or only in small amounts (< 5 %), in pig feed formulations.

In sum, the risk for adverse effects in pigs at current exposure levels is considered low to negligible.

Enniatins and beauvericin

No effect data are available. Enniatins and beauvericin seem to have low absorption following oral exposure and very low bioavailability in animals. Enniatins certainly occur in cereal grains, but their restricted bioavailability indicates a low risk of adverse effects.

Beauvericin occurs at far lower levels in cereal grains and the effect risk is suggested to be negligible.

Moniliformin

A NOAEL at 25 mg/kg of moniliformin in the diet is derived for pigs.

This level is far higher than those measured in cereal grains in Norway. The maximum level measured is up to 1 mg/kg in wheat.

In sum, the risk for adverse effects of moniliformin in pigs is negligible.

Ochratoxin A

For ochratoxin A, a LOAEL for nephrotoxicity in pigs has been derived by EFSA corresponding to 0.2 mg/kg feed.There are also more recent results on spermatological parameters and on slaughter weight/daily gain/feed efficiency, indicating LOAELs of 0.003 and 0.025 mg/kg diets, respectively. The derived LOAEL at 0.2 mg/kg feed is about 1000 times higher than the mean level of ochratoxin A in crude cereal grains in Norway, and 5 times higher than the highest concentration measured (0.04 mg/kg in a sample of crude barley). Similarly low concentrations are usually also measured in grains in other European countries. However, considerably higher concentrations (> 1 mg/kg) have also been shown in maize and small cereal grains in European surveys (EFSA 2004d). The LOAELs, based on spermatological and performance effect levels, are considerably closer to realistic exposure levels in Norway.

In sum, the risk based on a nephrotoxic concentration of ochratoxin A in Norwegian feed for pig must be considered to be low. However, the risk for ochratoxin A concentrations shown to influence performance or sperm quality may be considered as moderate to high.

Aflatoxins

A NOEL has been derived for pigs at 0.2 mg/kg feed. Aflatoxins are only a risk in imported feed ingredients, particularly maize. The yearly mean levels found in maize are 0.0002-0.002 mg/kg. These data, combined with the low use of maize in pig compound feed, mean that the risk for aflatoxin health effects in pigs in Norway is negligible.

10.1.1.2 *Poultry*

Deoxynivalenol (DON)

In broiler chickens, DON at 1.7 mg/kg feed has been demonstrated to reduce the size of gut villi and influence immune responses to vaccinations, indicating a LOAEL at 1.7 mg/kg feed for broiler chickens. Prior to that, EFSA had concluded that the available data do not allow a maximum tolerance level to be estimated. Laying hens are more tolerant, with a derived LOAEL at 12 mg/kg feed, except for in breeder hens due to possible foetal effects at 2.5 mg/kg feed.

The estimated DON levels in a typical broiler chicken formulation, based on DON data in crude grains, result in a 99-percentile at 1.3 mg/kg, which is slightly below the LOAEL identified for broilers. However, broiler chicken feed may also contain a considerable amount of maize, which is usually also contaminated with DON. Thus, taken in total, there may be a certain (moderate) risk that DON concentrations in broiler chicken feed reach a level of health concern. The analytical results of DON in poultry feed showed a 99-percentile at 1.6 mg/kg, without specification of whether this was in broiler chicken feed or laying hen feed. For laying hens, the estimated DON levels, based on the measured DON concentrations in crude grains in a typical formulation, are about 3 times higher than in broiler chicken feed due to more oats being included in laying hen feed. Feed for laying hens may also contain some maize that typically further increases the DON concentration of the compound feed. According to the hazard characterisation, laying hens tolerate such DON levels and the risk for health or performance effects of DON must be regarded as low. However, particular care should be taken for breeder hens and the risk for foetal effects if they are exposed to feed with DON levels as estimated in feed for laying hens.

In sum, the risk for health effects in broiler chickens may be characterised as moderate, based on effects on intestinal villus size and immune defence. For laying hens, the risk for health or performance effects due to DON must be considered as low. Particular care should be taken for breeder hens as there may be a moderate risk of foetal effects if they receive feed with DON levels as estimated in the feed for laying hens.

T-2 and HT-2

No NOAELs are identified for poultry, but EFSA (2011a) reviewed a range of studies from which LOAELs may be identified. For broiler chickens and turkeys, 0.5 mg/kg of T-2 is identified as LOAEL based on oral lesions. For ducks, a LOAEL is identified at 0.2 mg/kg feed based on reduced weight gain. For laying hens, a LOAEL at 2 mg/kg feed is identified based on reduced egg production and hatchability. As a substitute for more data on poultry, EFSA (2011a) proposed using the $BMDL_{05}$ of 0.010 mg/kg bw, originally estimated for pigs,

for poultry. Assuming a normal feed consumption for poultry, this would correspond to 0.10 mg/kg of T-2+HT-2 in broiler chicken feed and 0.17 mg/kg in laying hen feed. This estimate of effect threshold may be appropriate for broiler chickens, but seems to be too low (highly conservative) for adult birds (layers).

The estimated levels in typical formualtions for broiler chicken and laying hens, based on analyses for these toxins in crude grains, show that the effect threshold for broiler chickens is exceeded at the 99-percentile. The analytical data on T-2 and HT-2 in unspecified poultry feed confirm that these toxins may reach the estimated threshold for health effects in poultry.

In sum, the risk for health effects in broiler chickens, turkeys, and ducks from T-2 and HT-2 in the feed is considered as moderate and the risk may be somewhat lower for adult hens.

Nivalenol

A limit for observed effects in poultry is 1 mg/kg, where intestinal bleeding was observed in laying hens. As nivalenol is usually undetectable or at levels only slightly above the detection limit in cereal grain in Norway, the risk for adverse effects in poultry is regarded as low.

Zearalenone

Poultry seem to be relatively tolerant of zearalenone. Symptoms of hormonal effects have been only observed at very high doses that are unlikely to occur under normal feeding conditions.

Fumonisins

For broiler chickens, a LOAEL at 20 mg FB1/kg diet has been derived based on an increased sphinganine:sphingosine ratio. From more recent results on ducks, we identified a LOAEL at 5 mg/kg diet based on the same effect biomarker. Young poultry are more sensitive to the toxin than adults. Fumonisins are contaminants of concern in maize, and poultry used to be fed on relatively large amounts of maize (up to about 20 %). However, the risk of reaching levels that produce adverse effects is considered as low.

Enniatins and beavericin

No effect data are available for enniatins. No effects were observed in broiler chickens fed diets containing up to 12 mg/kg of beauvericin, given via maize inoculated in the field.

The adverse effect risk for enniatins is probably low due to low bioavailability and for beavericin the risk is negligible.

Moniliformin

We identified a LOAEL for moniliformin in broiler chickens at approximately 25 mg/kg diet based on liver toxicity, and a NOAEL for moniliformin in turkeys at 12.5 mg/kg diet based on increased heart weight at 25 mg/kg.

These levels are considerably higher than those measured in cereal grains in Norway, where the maximum level measured is below 1 mg/kg in wheat.

In sum, the risk for moniliformin effects in poultry is considered to be negligible.

Ochratoxin A

Knowledge regarding the dose-response relationships of ochratoxin A in poultry is scarce. A NOEL has yet to be established for ochratoxin A in birds. A LOAEL is identified from a recent study on male broiler chicken at 0.1 mg/kg diet based on immune effects, and previously was derived a LOAEL at 0.5 mg/kg based on immune function in broiler chicken . Ochratoxin A levels measured in cereal grains in Norway are low and below those expected to be of health concern for poultry. However, ochratoxin A occurs at higher concentrations in feed ingredients available at the international market.

In sum, the risk for ochratoxin A effects in poultry in Norway must be considered as low.

Aflatoxins

There are species differences in sensitivity towards aflatoxins among poultry, with ducklings being most sensitive and broiler chickens considerably less sensitive. It has been shown that ducks developed hepatic lesions following dietary exposure to 0.030 mg aflatoxin/kg feed, turkeys at exposure to 0.3 mg/kg feed, and chickens at 0.5 mg/kg feed. Effects on egg production and hatchability in laying hens occur at higher exposure levels than liver effects. Aflatoxins are only a risk in imported feed ingredients, particularly maize. The yearly mean levels shown in maize are 0.0002-0.002 mg/kg.

In sum, there may be a certain, but low, risk for effects in ducks fed large amounts of aflatoxin-contaminated maize. For broiler chickens, laying hens, and turkeys the risk is negligible.

10.1.1.3 *Ruminants*

Deoxynivalenol (DON)

It is difficult to reach conclusions regarding the tolerance of ruminants for DON-contaminated feedstuff. Under normal circumstances, DON is inactivated by rumen microorganisms and feed containing several ppm of DON has been fed to ruminants without resulting in any health or production problems. However, ruminants in forced production, generating stress and with increased ruminal turnover, or ruminants with any kind of metabolic disorder, such as chronical ruminal acidosis, will have a reduced capacity to inactivate the mycotoxin in the rumen. Thus, DON in total ration up to at least 6 mg/kg is suggested to be safe under normal rumen-physiological circumstances, but intensive feeding with compound feed may increase susceptibility.

In sum, the risk for adverse effects of DON in healthy ruminants fed normal amounts of compound feed (below about 50 % dry weight) is considered negligible. An increased risk of DON effects is observed if ruminants are fed higher amounts of compound feed.

T-2 and HT-2

The effect data for ruminants are scarce, but there are indications of reproductive adverse effects in sheep exposed to T-2 at approximately 0.3 mg/kg total diet, and also at approximately 0.1 mg/kg total diet in sheep with induced rumen acidosis. A case report describes health and production effects in dairy cows exposed to 0.3-0.6 mg/kg total ration. Thus, there are indications that ruminants can be rather susceptible to T-2. On this basis, care should be taken when including oats in ruminants' feed. However, by using oats at a normal level, as in the example of formualtion in this evaluation (10 %), the risk for T-2 and HT-2

effects in ruminants is regarded as low (the estimated 99-percentile showed 0.17 mg/kg of T-2 and HT-2 in compound feed). Furthermore, the fact that ruminants eat considerable quantities of roughage (max 50 % compound feed) improves the safety margin.

In sum, by using oats at the normal restricted level in compound feed and with a considerable ratio of roughage (above 50% of dry matter), the risk for T-2 and HT-2 effects in ruminants is low.

Nivalenol

There are no effect data for nivalenol in ruminants. However, the toxic potency of nivalenol in ruminants is generally at a similar level as that of DON. Based on the low occurrence of nivalenol in cereals in Norway, a negligible risk is suggested.

Zearalenone

Sheep may be rather sensitive to zearalenone, particularly during oestrus. A reduced ovulation rate has been shown at approximately 0.9 mg/kg feed, which is a derived LOAEL. Heifers seem to be far less sensitive to adverse zearalenone effects. Data for zearalenone in grains in Norway are all below the effect level in sheep. However, zearalenone may occur at rather high levels in maize and ruminants' feed may contain up to about 20 % maize.

In sum, the risk for health effects in sheep is regarded as low, and in other ruminants negligible.

Fumonisins

Ruminants are not sensitive to fumonisins. Adverse effects are observed in ruminants fed fumonisin B1 at 75 mg/kg feed and above, and no effects were observed in feeder calves fed 26 mg/kg feed. The effect level is far above the levels that occur in maize.

In sum, the risk for adverse effects in ruminants is negligible.

Enniatins and beauvericin

There are no effect data available. Nevertheless, effects are unlikely due to the low bioavailability. The risk is assumed to be negligible.

Moniliformin

There are no effect data available. However, moniliformin has been shown to be of rather low toxic potency generally and occurs in cereals at a fairly low level. Thus, effects in ruminants from exposure to moniliformin are not suggested to constitute a particular risk. The risk is assumed to be negligible.

Ochratoxin A

Ruminants are tolerant of ochratoxin A. Immature ruminants are less tolerant, but the occurrence data show that the levels are orders of magnitude below an effect level. The risk is negligible.

Aflatoxins

The problems associated with aflatoxins in ruminants are not animal health effects, but ruminal biotransformation and excretion of metabolites in milk that may then be used for human consumption. Aflatoxin carry-over is discussed in Chapter 8.8.4. The present maximum limit for aflatoxins in feed for ruminants has been set in order to prevent the maximum limit for aflatoxins in milk being exceeded. Annual monitoring shows an absence of, or only minor traces of, aflatoxin M1 in milk.

10.1.1.4 *Horse*

Deoxynivalenol (DON)

Reduced feed intake, lower body weight gain or biochemical effects have been shown in studies of horses fed levels of DON that correspond to approximately 5 mg /kg total diet. A NOEL is not established. The estimated DON levels in compound feed for horses are based on the DON concentrations measured in crude cereal grains and a typical formulation (with 30 % oats) and showed a 99-percentile at 3.8 mg DON/kg. Maize is not normally used in horse feed and thus does not increase the DON level any further.

In sum, based on a normal feed recipe and the fact that cereals constitute a restricted part of the total diet, the risk for adverse effects of DON in horses may be regarded as low. Higher amounts of oats in the diet will increase the risk for adverse effects.

T-2 and HT-2

In their evaluation of T-2 and HT-2, EFSA (2011a) concluded that the available data did not provide a basis for establishment of safe levels in horse feed. In the absence of NOAELs or LOAELs for horses, EFSA proposed to use the same reference point (0.010 mg/kg bw/day) as that derived for pigs, as the toxicokinetics of T-2 and HT-2 toxins in horses are not substantially different from those in pigs. This reference point would provide an indication on the possible risk, and such a daily dose would correspond to approximately 0.6 mg/kg feed per day. From a study of T-2 in horses, feed refusal and skin lesions were observed at approximately 0.7 mg T-2/kg total diet. This result may indicate that the threshold for effects in horses is lower than 0.6 mg/kg total feed. The estimated T-2 and HT-2 concentrations in compound feed for horses, based on the HT-2+T-2 concentrations measured in crude cereal grains and a typical formulation of compound feed for horses (with 30 % oats), showed a 99-percentile at 0.48 mg/kg of T-2 and HT-2.

In sum, based on a normal compound feed formulation and the fact that cereals constitute a restricted ratio of the total diet, the risk of effects from T-2 and HT-2 in horses may be regarded as low. Horses fed on higher amounts of oats have an increased risk of suffering adverse effects.

Nivalenol

There are no effect data for nivalenol in horses. However, the toxic potency of nivalenol is generally at a similar level as that of DON. Based on the low occurrence of nivalenol in cereals in Norway, a negligible risk is anticipated.

Zearalenone

No effects were shown in a study of horses fed zearalenone at approximately 1 mg/kg diet. Based on the Norwegian data of relatively low zearalenone levels in cereal grains, and the fact

that horses receive only very small quantities of maize (which may be particularly contaminated with zearalenone), the risk for adverse effects from zearaleonone in horses is regarded as negligible.

Fumonisins

Some data indicate that consumption of feed containing more than 10 mg fumonisin B1/kg is associated with an increased risk of horses developing encephalomalacia (ELEM), whereas at concentrations below 6 mg/kg diet no increased risk is found. Maize is the commodity of greatest concern as fumonisin concentrations in maize may reach several mg/kg. However, horses do not normally receive considerably amounts of maize.

In sum, the risk for adverse effects for fumonisins in horses in Norway is regarded as negligible.

Enniatins and beauvericin

There are no effect data available. Nevertheless, due to the suggested low bioavailability effects are unlikely. The risk is considered to be negligible.

Moniliformin

There are no effect data available. However moniliformin is shown to be of rather low toxic potency generally and occurs in cereals at fairly low levels. Thus, the effects of moniliformin in horse are not suggested to constitute a particular risk. The risk is considered negligible.

Ochratoxin A

Horses' sensitivity to ochratoxin is broadly unknown, but cases of intoxication indicate relatively low sensitivity. The measured occurence levels in cereals are low and the risk of adverse effcts in horses from ochratoxin A is assumed to be negligible.

Aflatoxins

Although the dose-response information for horses is very limited, as maize is seldom included in horse feed, the risk to horses from aflatoxin effects is negligible.

10.1.1.5 *Rabbit*

Deoxynivalenol (DON)

The available data for rabbits are too scarce to establish a safe limit for DON in feed. Although no effects have been reported from rabbits fed up to 3 mg/kg feed, a reduced maternal body weight was reported from pregnant female rabbits fed a diet containing from 7.5 mg/kg diet. Considering the DON levels occurring in crude grain in Norway, there might be a certain (low to moderate) risk for adverse effects from DON in rabbits.

T-2 and HT-2

A NOAEL of 0.1 mg T-2 toxin/kg bw per day (estimated to be approximately 2 mg/kg diet) is derived for rabbits. Considering the T-2 and HT-2 levels occurring in grain in Norway, there may be a low risk for adverse effects from T-2 and HT-2 in rabbits.

Nivalenol

There are no effect data for nivalenol in rabbits. However, the toxic potency of nivalenol is generally at a similar level as that of DON. Based on the low occurrence of nivalenol in cereals in Norway, a negligible risk is expected.

Zearalenone

Effects in rabbits have been measured at 0.5 mg/kg of zearalenone in the diet, which we identified as a LOEL for anabolic effects. The levels of zearalenone in cereals in Norway show ranges above this LOEL and rabbit feed may also contain some maize (which may also contain some zearalenone). Therefore, the risk for effects from zearalenone in rabbits is expected to be moderate.

Fumonisins

Rabbits seem to be relatively sensitive to fumonisin. Maternal toxicity was observed at 0.25 mg/kg bw per day, and foetal toxicity occurred at the lowest dose tested (0.1 mg/kg bw, approximately 2 mg/kg diet), but no evidence for teratogenic effects was shown. This level was identified as LOAEL. If maize constitutes a considerable part of the diet, the risk for adverse effects from fumonisins in rabbits may be moderate.

Enniatins and beauvericin

There are no effect data available. Nevertheless, due to the suggested low bioavailability, effects are unlikely. The risk is supposed to be negligible.

Moniliformin

There are no effect data available. However, moniliformin is shown to have rather low toxic potency generally and occurs in cereals at fairly low levels. Thus, the effect of moniliformin in rabbits is not suggested to constitute a particular risk. The risk is supposed to be negligible.

Ochratoxin A

The effect data of ochratoxin A in rabbit are scarce. Approximately 2 mg/kg diet has been shown to be teratogenic and reduce the number of live foetuses, as well as the mean foetal weight and length. No effects were observed at approximately 1 mg/kg feed. This effect level is far higher than the levels measured in cereals in Norway and the risk is supposed to be negligible.

Aflatoxins

Liver effects have been shown in rabbits fed 0.1 mg aflatoxin B1/kg feed. Data on the concentrations of aflatoxins in rabbit feed in Norway are lacking. Analysis results of aflatoxins in maize and in feed for ruminants show levels that are far lower, and which may indicate that the levels in rabbit feed are also low. The risk of aflatoxin effects in rabbits in Norway is supposed to be negligible.

10.1.1.6 *Dog and cat*

Deoxynivalenol (DON)

In dogs, 2.7 mg/kg of DON in the diet (only one dosage level) reduced their feed intake and body weight, but the effect might have been influenced by co-occurring toxins. Another study estimated a threshold for reduced feed intake at 4.5 mg/kg diet. In cats, a threshold for reduced feed intake was estimated at 7.7 mg/kg diet.

The estimated DON concentration in compound feed for dogs, based on DON concentrations in crude cereal grains and a typical formulation of compound feed, showed low DON levels (mean DON level at 0.04 mg/kg). However, analysis of 24 samples of commercial compound dog feed showed mean DON levels that were ten times higher and a 99-percentile at 1.2 mg/kg. This discrepancy is probably due to the fact that most dog feed on the Norwegian market is imported, and its cereal grain content does not reflect the DON levels in Norwegian grain. Furthermore, other ingredients, such as maize, are included in most dog feed (usually 10-20 %), which probably contributes significantly to the DON level in the feed.

In sum, the measured DON levels in some samples of compound feed for dogs, as an indicator of DON levels in commercial dog feed on the Norwegian market, demonstrate that the levels are below the estimated adverse effect level for dogs,. This suggests that there is a relatively low risk for adverse effects. However, both effect data and occurrence data are scarce. The sparse effect data for cats indicate that cats are somewhat more tolerant to DON than dogs. As the amounts of cereals included in compound feed for cats are lower than in feed for dogs, the risk for adverse DON effects in cats should be negligible.

T-2 and HT-2

There are no effect data on dogs available. In the absence of effect data for dogs, EFSA (2011a) proposed to use the same reference point (0.010 mg/kg bw/day) as that derived for pigs as the toxicokinetics of T-2 and HT-2 toxins in dogs do not differ substantially to those in pigs. Such a daily dose would correspond to approximately 0.3-0.5 mg/kg diet for dogs. This level is more than 10 times higher than the mean concentration measured in dog feed. For cats, the data show high sensistivity but are not suitable for estimation of NOAEL or LOAEL. Occurrence data of the toxins in cat feed are not available, but are suggested to be low due to the lower cereal content of cat feed.

In sum, the risk for health effects in dogs is supposed to be negligible, but both effect data and occurrence data are scarce. Cats seem to be particularly susceptible to T-2. As cat feed contains lower amounts of cereal grains, the risk for adverse effects in cats is considered to be low.

Nivalenol

There are no effect data for nivalenol in dogs and cats. However, the toxic potency of nivalenol is generally at a similar level as that of DON. The occurrence of nivalenol in cereals in Norway is low. Similarly low occurrence is also shown in most surveys of cereals from other countries. This is important as most compound feeds for dogs and cats are imported. A negligible risk is expected.

Zearalenone

For dogs, a LOAEL is identified at approximately 1.3 mg/kg diet, based on metabolic changes with anaemia and hyperbiliruminemia. The few analyses for zearalenone in cereal grains in Norway indicate relatively low levels in Norwegian cereals. However, dog feed may contain considerable amounts (10-20 %) of maize, which is an ingredient that may be particularly contaminated with zearalenone. The mean measured levels of zearalenone in commercial compound feed for dog in Norway (2011) was about 1:10 of the LOAEL, with a maximum at 0.25 mg/kg. For cats, no effect data are found.

In sum, the risk for adverse effects of zearalenone in dogs is considered low. As cat feed contains lower amounts of cereal grains, the risk of adverse effects from zearalenone in cats is considered negligible.

Fumonisins

There are no effect data found for dogs and cats. Maize is the commodity of concern, and the fumonisin concentration in maize may reach several mg/kg. Fumonisin analyses in samples of commercial compound feed for dogs (2011), measured up to 0.24 mg/kg of B1+B2. If dogs are as sensitive as pigs for fumonisins, the risk is supposed to be low. However, this conclusion is uncertain due to few data on occurrence and the lack of effect data. As cat feed contains lower amounts of cereal grains, the risk of adverse effects from fumonisins in cats is supposed to be negligible.

Enniatins and beauvericin

There are no effect data available. Nevertheless, due to the suggested low bioavailability effects are unlikely. The risk is supposed to be negligible.

Moniliformin

There are no effect data available for dogs and cats. However, in another carnivore, mink, reproductive effects (increased neonatal mortality and reduced body weight of the offspring) were observed at exposure to 17 mg/kg of moniliformin in the diet during the breeding and nursing period. No significant effects were measured at 8 mg/kg. Moniliformin is generally of rather low toxic potency and seems to occur in cereals at levels far below the NOEL in mink. Thus, effects of moniliformin in dogs and cats are not suggested to constitute a particular risk. The risk is considered to be negligible.

Ochratoxin A

Kidney tubular necrosis and ultrastructural changes in the proximal tubuli were shown in dogs fed ochratoxin A at the lowest dosage level tested, approximately 5 mg/kg feed and above. The measured occurrence levels in cereals in Norway and the levels measured in dog feed (2011) are 4-5 orders below this level. The risk for adverse effcts in dogs from ochratoxin A is regarded as negligible.

For cats, no dose-response effect data have been found. The risk is supposed to be negligible based on the effect data for dogs, the few occurrence data in dog feed, and the large margin of difference between effect and occurrence in dog feed.

Aflatoxins

Only trace amounts, slightly above the detection level, occur in maize samples in Norway, and most samples of compound feed for dogs examined in 2010 were negative, except for some detectable trace concentrations in some samples. The risk for aflatoxin effects in dogs in Norway is expected to be low or negligible. An identified NOAEL for aflatoxins in dogs is 0.2 mg/kg feed. For cat, no data are available, but the risk is considered to be lower than for dog, as there are fewer sources of aflatoxins in cat feed.

10.1.1.7 *Fish*

Relative feed intake (% per bw/day) depends on fish size and water temperature, with highest feed intake in juvenile-start feeders at high water temperatures, and lowest feed intake for adult salmon at low temperatures. In order to estimate the exposures between dietary concentration and consumed mycotoxin per kg body burden per day, a high feed intake of 3 % bw/day was used for juvenile fish (<70 g) and 0.5 % bw/day for adult (>2 kg) Atlantic salmon.

DON

A NOAEL for DON in Atlantic salmon at 2 mg/kg diet is identified based on reduced performance as critical effects. In rainbow trout the LOAEL is below 2.6 mg/kg diet. The theoretical highest level in salmon feed is 0.1 mg/kg diet (0.003 mg/kg bw/day for juvenile

Atlantic salmon and 0.0005 mg/kg bw/day for adult salmon). In sum, the risk for DON effects in Atlantic salmon is negligible, while for rainbow trout no such feed risk assessment could be made as no NOEL was established.

T-2 and HT-2

A NOAEL for T-2 in rainbow trout at 1.0 mg/kg diet is derived, based on reduced performance and haematocrit level. In channel catfish, a LOAEL at 0.63 mg/kg feed is derived based on reduced performance. The theoretical highest level of T-2 and HT-2 in Atlantic salmon feed is 0.006 mg/kg feed (0.17 µg/kg bw/day for juveniles and 0.03 µg/kg bw/day for adults). In sum, the risk for T-2 and HT-2 effects in rainbow trout is negligible and the same is considered for Atlantic salmon.

Nivalenol

There are no effect data for nivalenol in fish. However, the toxic potency of nivalenol is generally at a similar level as that of DON. Nivalenol shows low occurrence in cereals in Norway. Similarly low occurrence is also shown in most surveys of cereals from other countries. A negligible risk for adverse effects in Atlantic salmon is expected.

Zearalenone

Reports of oestrogenic effects from oral exposure to zearalenone in fish are lacking, but no other health effects have been shown in Atlantic salmon fed up to 0.77 mg/kg feed. The theoretical highest level in feeds for Atlantic salmon is 0.1 mg/kg (0.003 mg/kg bw/day for juveniles and 0.0005 mg/kg bw/day for adults). A negligible risk of adverse effects from zearalenone in Atlantic salmon is considered.

Fumonisins

From data on rainbow trout feed levels of fumonisin B1 at 3 mg/kg diet (0.15 mg/kg bw per day), a NOEL is identified as based on spingolipid disruption. The highest theoretical levels of fumonisin B1 in salmonid feeds were estimated to be 0.24 mg/kg (0.007 mg/kg bw/day for juvenile and 0.0012 mg/kg bw/day for adult Atlantic salmon), which is about 10 times lower than the NOEL for rainbow trout. The risk of adverse effects in salmonid fish is considered as negligible. No information exists on the effects of fumonisin B2 and B3.

Enniatins and beauvericin

There are no effect data available. Nevertheless, due to the suggested low bioavailability effects are unlikely. The risk is expected to be negligible.

Moniliformin

There are no effect data available for fish. Moniliformin is shown to be of rather low toxic potency and seems to occur at low levels in cereals. Thus, the effects of moniliformin in fish are not suggested to constitute a particular risk. The risk is expected to be negligible.

Ochratoxin A

Studies on channel catfish have shown reduced weight gain and fewer exocrine pancreatic cells in liver at 1 mg/kg diet and a NOAEL was identified at 0.5 mg/kg. The preliminary results from a study on Atlantic salmon are that no effects are demonstrated when fed up to 2.4 mg/kg diet. The theoretical highest levels in salmon feeds are 0.001 mg/kg (0.0003 mg/kg bw/day for juvenile Atlantic salmon and 0.00005 mg/kg bw/day for adults). In sum, the risk for DON effects in salmonid fish is expected to be negligible.

Aflatoxins

Based on the development of tumours in later life, after temporary feeding during early life stages, the lowest observed adverse effect levels (LOAEL) could be set at 0.0004 mg/kg feed. For aflatoxin B1, the highest theoretical levels in salmonid feeds are 0.0004 mg/kg, suggesting that long-term (> 1 year) low exposures to a theoretical high level (mostly derived from maize) could cause the formation of hepatocellular carcinoma (HHC) in rainbow trout. It should be noted that aflatoxins may be degraded during the heat extrusion process in aquafeed production. This potential degradation is not included in this risk assessment. Rainbow trout are highly sensitive to dietary aflatoxin B1-induced HHC, while other species, such as coho salmon, are less sensitive. No information are available for Atlantic salmon.

10.2 **Human risk characterisation of mycotoxins in food**

The safe intake levels for mycotoxins applied in the present risk assessment have been established by different international Food Safety Authorities (e.g. EFSA, JECFA, and SCF) on the basis of toxicity studies in animals. Safe intake levels are expressed as Tolerable Daily Intake (TDI), Provisional Maximum Tolerable Daily Intake (PMTDI) or as an Acute Reference Dose (ARfD) for a single meal. The TDIs represent the daily levels that a person, including vulnerable individuals like infants and small children, can be exposed to throughout life without appreciable health risk.

TDIs are derived from the toxicological effects triggered at the lowest exposure dose in the most sensitive experimental species. Due to the integrated uncertainty factors and the conservative way in which the tolerable intake levels are derived, exceeding the TDI will initially only represent a reduced safety margin. Thus, the TDIs are not a threshold for toxicity with onset of adverse effects, and it is therefore difficult to quantify the risk caused by intakes above the TDI.

The exposure via grain-products is compared to the TDI or PMTDI for the mycotoxin in question, and in the characterization of risk VKM makes use of the terms "of no concern", or "of concern" depending on whether the calculated exposure is below and/or close to, or exceeding, the respective TDI. Human exposure below or close to TDI is considered to be of no concern whereas exposure exceeding the TDI is considered to be of concern. Correspondingly, for characterization of risk for acute dietary exposure the calculated exposure via one single meal is compared to ARfD. Acute exposure similar to or below the ARfD is considered to be of no concern.

10.2.1 Risk characterisation for mycotoxin propagation in the food chain (animal products)

Trichothecenes do not accumulate in any tissue or organ in exposed food-producing animals. No residues or only small traces of the trichothecene mycotoxins DON, nivalenol, T-2 and HT-2 are found in animal derived food products such as meat, milk or eggs. Any potential human exposure to these toxins through consumption of animal-derived food products are considered to be very low compared to the exposure from cereals.

There is only limited deposition of zearalenone in meat and the transfer to milk and eggs is low. The human exposure to zearalenone through consumption of animal-derived food products is very limited.

Ochratoxin A has a longer half-life in animals due to protein binding in plasma. Previous human exposure assessments has shown that the human exposure to ochratoxin A through consumption of meat, milk and milk products is low compared to the exposure to from consumption of plant products. EFSA estimated that that food of animal origin only had a minor contribution to the total dietary intake (generally < 3 %, and in populations with specific dietary preferences < 10%) (EFSA 2006).

10.2.2 Human risk characterisation from chronic dietary exposure to mycotoxins

10.2.2.1 *Chronic dietary exposure to deoxynivalenol (DON)*

DON found in practically all samples of wheat flour, wheat bran and oats. The TDI set by JECFA is a group TDI, including DON and its acetylated forms. Due to the lack of occurrence data for the acetylated forms in food, the exposure estimations in this risk assessment only includes DON. Studies of the occurrence of the acetylated forms in Norwegian grain have, however, shown that 3-acetyl-DON would add an approximately 10% to the DON levels (Langseth and Rundberget 1999). In addition, the uncertainty related to the levels and bioavailability of DON-glucosides and other bound forms should be kept in mind when considering the risks related to the dietary exposure to DON.

Risk for DON exposure in 1-year-old infants and 2- to 4-year-old children

The estimated mean total dietary intakes of DON (see Table 43) are in the range of, or exceeding, the TDI of 1 µg/kg bw/day (set by SCF 1999 and JECFA 2011) for the 1-, 2- and 4-year-olds in all years. The 2-year-olds have the highest exposure, probably due to a high consumption of grain-based food relative to their body weight. In a year with low DON concentration in the flour, the mean intake for the 2-year-olds was slightly above the TDI (1.1 versus 1 µg/kg bw/day), while it was 2-fold the TDI in years with high mean concentrations. The estimated high (95-percentile) exposures in these age groups exceed the TDI 2-3.5 times depending on age and year with low or high mean DON-concentrations.

In years of low DON concentration, 95-percentile exposure in 1- and 2-year-olds will exceed the TDI and is of concern.

In years of high DON concentration exceeding TDI for mean exposures of DON in 2-year-olds is of concern. Furthermore, in years of high DON concentration exceeding TDI for high exposures of DON in 1-, 2- and 4-year-olds is of concern.

A main contribution to the total estimated dietary intake for 1-year-olds is powder-based porridge. The occurrence data for commercial powder-based porridge are limited to one year, 2008. In this year, the mean DON-concentrations in wheat-based and oat-based powder porridges were 13 µg/kg and 34 µg/kg, respectively (see Appendix B). The use of data from powder-based oat porridge only, is a worst-case estimation since it is likely that 1-year-old infants also eat other types of powder-based porridges with lower levels of DON than oat-based. On the other hand, data for powder-based oat porridge were only available for one year so that the annual variation is not known. Powder-based porridge is, however, not a considerable food source for 2- and 4-year-olds.

Risk for DON exposure in 9- and 13-year-olds

The estimated mean exposure to DON in 9- and 13-year-old children is equal to or below the TDI in all years. In years with low DON levels in the flour, the high intake of DON (95-percentile) is in the range of the TDI in both age groups (Table 43). In years with high mean DON concentrations in the flour, the high intake of DON (95-percentile) exceeds the TDI up to 1.6-fold for the 9-years-olds.

In years of high DON concentration, exceeding TDI for high exposure of DON in 9-year-olds is of concern. For 13-year-olds the estimated DON exposure is of no concern.

Risk for DON exposure in adults

The estimated mean and high intakes in adults are all below the TDI. The estimated dietary intake of DON in the adult population is therefore not a health concern.

In summary

It should be remarked that even though the DON exposure in children may be of concern, the exposure is maximally 3.5-fold the TDI. Exceeding the TDI will initially only represent a reduced safety margin. There is, however, an uncertainty related to the exposure to and toxicological significance of the other forms of DON known to be present in cereal grains that not are included in the exposure estimates.

10.2.2.2 *Chronic dietary exposure to zearalenone*

The mean estimated intake as well as the estimated 95-percentile intake of zearalenone is below the TDI for all age groups.

The dietary intake of zearalenone is of no concern.

Table 43 Estimated exposure of deoxynivalenol and zearalenone (µg/kg bw/day) in years with low and high mycotoxin concentration in flour (see Table 23) in different age groups and the respective tolerable daily intake.

Mycotoxin	Mean low[1] (95-perc)[2] µg/kg bw/day	Mean high[3] (95-perc)[2] µg/kg bw/day	Tolerable daily intake µg/kg bw/day
1-year-olds (n=1635)			
DON	0.89 (1.8)	1.4 (3.1)	1[4]
Zearalenone	0.022 (0.044)	0.078 (0.16)	0.25[5]
2-year-olds (n=1674)			
DON	1.1 (1.9)	2.0 (3.5)	1[4]
Zearalenone	0.018 (0.032)	0.11 (0.18)	0.25[5]
4-year-olds (n=391)			
DON	0.73 (1.2)	1.1 (2.0)	1[4]
Zearalenone	0.009 (0.016)	0.096 (0.16)	0.255[5]
9-year-olds (n=810)			
DON	0.56 (1.02)	0.90 (1.6)	1[4]
Zearalenone	0.007 (0.012)	0.076 (0.13)	0.25[5]
13-year-olds (n=1005)			
DON	0.38 (0.72)	0.60 (1.1)	1[4]
Zearalenone	0.005 (0.009)	0.054 (0.10)	0.25[5]
Adults (n=1787)			
DON	0.27 (0.55)	0.45 (0.93)	1[4]
Zearalenone	0.004 (0.008)	0.031 (0.067)	0.25[5]

[1] Mean exposure in years with the lowest mean mycotoxin concentrations.
[2] High exposure (95-percentile).
[3] Mean exposure in years with the highest mean mycotoxin concentrations.
[4] Provisional maximum tolerable daily intake (PMTDI) (JECFA 2011).
[5] Tolerable daily intake (TDI) (EFSA 2011b).

10.2.3 Human risk characterisation based on chronic exposure scenarios

The dietary intake of the sum of T-2 and HT-2 toxins as well as of nivalenol could not be estimated due to the high number of samples below the LOD (see Chapter 8.4). Human risk was therefore characterized by preparing exposure scenarios based on toxin concentrations equal to half of the analytical LODs in wheat flour and equal to the LOD in wheat bran and oats.

10.2.3.1 *Chronic exposure scenario to the sum of T-2 and HT-2 toxins*

An overview of the exposure scenarios for the sum of T-2 and HT-2 toxins as well as the group TDI (EFSA 2011a) are given in Table 44. The exposures are highest for the youngest age groups, and in particularly for the 1-year-olds. This is due to the high consumption of grain-based products in relation to body weight.

Exposure to the sum of T-2 and HT-2 toxins in 1- to 4-year-olds

The exposure scenarios show that the mean exposures are in the range of the TDI for the 1-year-olds, and lower than the TDI for the 2- and 4-year-olds (Table 44). For the high consumption groups (95-percentile), the scenarios indicate exposures exceeding the TDI 2-fold for the 1-year-olds and 1.5-fold for the 2-year-olds. For the 4-year-olds, the intake in the high exposed group (95-percentile) is in the range of the TDI.

These scenarios indicate that 1- and 2-year-old children may exceed the TDI, which might be potentially of concern.

Exposure to the sum of T-2 and HT-2 toxins in 9- and 13-year-olds

The exposure scenarios for the sum of T-2 and HT-2 toxins in 9- and 13-year-olds show that the mean exposures as well as the high exposed groups are below the TDI (Table 44).

These scenarios indicate that exposure to the sum of T2 and H-T2 toxins in 9- and 13-year-olds is of no concern.

Risk for exposure to sum of T-2 and HT-2 toxins in adults

The exposure scenario indicates that the intake of T-2 and HT-2 toxins in adults is below the TDI (Table 44).

The scenario indicates that exposure to sum of T-2 and HT-2 toxins in adults is of no concern.

10.2.3.2 *Chronic exposure scenario to nivalenol*

An overview of the exposure scenario for nivalenol as well as the TDI (SCF 2000c) is given in Table 44. The exposures are highest for the youngest age groups, and in particularly for the 2-year-olds. This is due to the high consumption of grain-based products in relation to the body weight. However, the scenarios show that all consumption groups are exposed below the TDI for nivalenol.

The incidence of nivalenol in Norwegian grain is low, and levels in positive samples are normally low compared to DON.

VKM considers nivalenol to be of no concern for all age groups.

Table 44 Exposure scenarios for the sum T-2 and HT-2 toxins and nivalenol and their tolerable daily intakes (TDI)

	Scenario[1] Mean (95-percentile) µg/kg bw/day	Tolerable daily intake (TDI) (µg/kg bw/day)
1-year-olds (n=1635)		
Sum T-2 + HT-2	0.091 (0.21)	0.1[2]
Nivalenol	0.046 (0.092)	0.7[3]
2-year-olds (n=1674)		
Sum T-2 + HT-2	0.063 (0.15)	0.1[2]
Nivalenol	0.034 (0.063)	0.7[3]
4-year-olds (n=391)		
Sum T-2 + HT-2	0.033 (0.093)	0.1[2]
Nivalenol	0.016 (0.031)	0.7[3]
9-year-olds (n=810)		
Sum T-2 + HT-2	0.023 (0.077)	0.1[2]
Nivalenol	0.012 (0.023)	0.7[3]
13-year-olds (n=1005)		
Sum T-2 + HT-2	0.013 (0.041)	0.1[2]
Nivalenol	0.008 (0.016)	0.7[3]
Adults (n=1787)		
Sum T-2 + HT-2	0.017 (0.036)	0.1[2]
Nivalenol	0.007 (0.014)	0.7[3]

[1] The scenarios are based on mycotoxin concentrations given in Table 22. [2] TDI established by EFSA (2011a). [3] TDI established by SCF (2000).

10.2.4 Human risk characterisation of other mycotoxins in grains

Ochratoxin A was not included in the human risk characterization because a major part of the dietary intake is from non-grain food. The dietary intake of ochratoxin A, which is produced by *Aspergillus* and *Penicillium* species, has many sources including wine, coffee, tea, dried fruits, in particular raisins and dried apricots, etc., in addition to grain. The evaluation of the total ochratoxin A dietary intake is not within the terms of references for the present risk assessment. Ochratoxin A is the only mycotoxin assessed in this report with a known potential of transfer from feed to humans through meat consumption due to a long plasma half-life. Even so, other intake estimates have shown that a maximum of 10% of the total dietary intake of ochratoxin A is from consumption of meat, milk and eggs. VKM therefore considers that with the present levels of ochratoxin A in Norwegian grain, the transfer of ochratoxin A from animals to humans is low.

The occurrence of ochratoxin A in the crude grain samples analysed showed that the levels in stored samples of Norwegian grain were low, with most samples below the LOD (Norwegian Food Safety Authority, annual monitoring programme, Tables 8, 9, and 10). Data for food products were not available. Furthermore, ochratoxin A has earlier been analysed in plasma from blood donors and the food intake was estimated (Thuvander et al. 2001). The intake was below the PTWI used at that time. Since then, the TWI has been increased considerably (approximately 5-fold).

VKM considers it therefore as unlikely that ochratoxin A in grain is of any concern for human health in Norway.

Aflatoxins are normally not produced under the climatic conditions in Norway. Occurrence data for aflatoxins in Norwegian grain-based food products were not available. Comparable with the situation for ochratoxin A, aflatoxins occur mostly in non-grain foods so that the

evaluation of the total aflatoxin dietary intake would not be within the terms of references for the present risk assessment.

VKM considers that cereal grain and their products available in Norway probably do not significantly contribute to the total aflatoxin exposure in humans. The increasing maize consumption may, however, constitute a source to aflatoxins exposure, but this could not be estimated.

Aflatoxin M1 may be excreted in milk from dairy cows receiving feed contaminated with aflatoxins B1 (see Chapter 9.8.4). This is, however, not a risk to human health with the current levels of aflatoxins in cattle feed.

Fumonisins occur almost exclusively in maize, and the human consumption of maize in Norway is very limited. The dietary intake of fumonisins could not be estimated due to few occurrence data (Table 19).

Moniliformin, enniatins, or beauvericin are considered as emerging mycotoxins in grains. Occurrence data for Norwegian grain are scarce, but the available data (Tables 8, 9 and 10) confirm the presence of these mycotoxins. However, due to the lack of occurrence data in foods and insufficient toxicological data, a risk assessment could not be performed.

VKM recognizes the presence of moniliformin, enniatins and beauvericin in Norwegian grains, which potentially might be of risk for human health.

10.2.5 Human risk characterisation of acute exposure

DON is the only mycotoxin for which an acute reference dose (ARfD) is established (JECFA 2011). The estimated exposures of 1.5 µg/kg bw for 2-year-olds and 0.76 µg/kg bw for adults from one single meal (see Chapter 8.5, Table 27) are well below the ARfD of 8 µg/kg bw.

VKM therefore concludes that acute effects of DON by consuming grains or grain-products in Norway are of no concern.

10.2.6 Summary human risk characterisation

DON is the main mycotoxin of concern in Norwegian grain. It is the most prevalent mycotoxin in Norwegian grain and is present in almost all samples of flour and oat flakes. The estimated mean and high (95-percentile) exposures for years with low mean DON-concentration, respectively, were in the range of, or exceeded, the TDI in children (1- to 2-year-olds). The high exposure (95-percentile) exceeded the TDI for 1-, 2-, 4- and 9-year-olds in years with high mean DON-concentration.

VKM concludes that exceeding the TDI at mean or high exposures to DON in infants and children is of concern although the TDI is not a threshold for toxicity. The estimated dietary intakes of DON in adolescence and in the adult population are equal to or below the TDI and is therefore not a health concern. Acute exposure to DON is of no concern.

The estimated mean intake of zearalenone was below the TDI for all age groups, while the estimated 95-percentile for the 2- and 4-year-olds, the age groups with the highest intake, was close to the TDI. For older children and adults, all estimated intakes of zearalenone were below the TDI. VKM concludes that the dietary intake of zearalenone is considered to be of no concern for all age groups.

The dietary intake of the sum of T-2 and HT-2 toxins could not be estimated due to the high number of samples below the LOD. Human risk was therefore characterized by preparing

exposure scenarios based on toxin concentrations equal to half of the analytical LODs in wheat flour and equal to the LOD in wheat bran and oats. It was noted that this might constitute an overestimation. These scenarios indicate that the dietary intake of the sum of T-2 and HT-2 toxins in 1- and 2- year-olds may exceed the TDI, while the 4-year-olds with high exposure have an intake in the range of the TDI. According to the exposure scenarios, the exposures to the sum of T-2 and HT-2 toxins in 9- and 13-year-olds are below the TDI. Furthermore, both the mean and high exposures in adults are below the TDI. VKM concludes that according to the performed exposure scenarios, the dietary intake of the sum of T-2 and HT-2 is potentially of concern for the youngest age groups.

The dietary intake of nivalenol could not be estimated due to the large number of samples (> 90%) below the LOD. Human risk was therefore characterized by preparing exposure scenarios based on toxin concentrations equal to the analytical LODs. It was noted that this might constitute an overestimation. The exposures are highest for the youngest age group, and in particularly for the 2-year-olds. This is due to the high consumption of grain-products in relation to the body weight. However, the scenarios show that all consumption groups are exposed below the TDI for nivalenol. VKM therefore concludes that the risk from the dietary intake of nivalenol is of no concern for all age groups.

Ochratoxin A was not included in the human risk characterization because a major part of the dietary intake is from non-grain food and the evaluation of the total dietary ochratoxin A-intake is not included in the terms of references for the present risk assessment. The occurrence of ochratoxin A in the analysed crude grain samples showed that the levels in stored samples of grain on the Norwegian market were low, with most samples below the LOD, whereas data for grain-based food products were not available. Therefore, VKM considers it unlikely that ochratoxin A in grain alone is of concern for human health in Norway.

Aflatoxins are normally not produced under Norwegian climatic conditions. Occurrence data for aflatoxins in grain on the Norwegian market were not available. Comparable with the situation for ochratoxin A, non-grain foods contribute significantly to the dietary intake and the evaluation of the total aflatoxin dietary intake is not included in the terms of references for the present risk assessment. VKM considers that grain and grain-based products available in Norway do not significantly contribute to the total aflatoxin exposure in humans, but an increasing consumption of maize could be a potential source to dietary intake of aflatoxin.

Fumonisins occur almost exclusively in maize, and the human consumption of maize in Norway is very limited. The dietary intake of fumonisins could not be estimated due to few occurrence data.

Moniliformin, enniatins, or beauvericin are considered as emerging mycotoxins in grains. Occurrence data for Norwegian grain are scarce, but the available data confirm the presence of these mycotoxins. However, due to the lack of occurrence data in food and insufficient toxicological data, a risk assessment could not be performed. VKM recognizes the presence of moniliformin, enniatins and beauvericin in Norwegian grains, which potentially might be of risk for human health.

Uncertainties

Uncertainties exposed in this opinion are divided into three disciplines corresponding to terms of reference: plant health, animal health and human health.

Plant health

Mycotoxin contamination of cereals grains in the field requires the presence of mycotoxin-producing fungi in the field, environmental conditions suitable for infection and development and susceptible cereal varieties. There are uncertainties concerning all three factors.

Presence of mycotoxing-producing fungi in the field

Recent evidence for changes in the prevalence of mycotoxing-producing *Fusarium* fungi in Norwegian cereals indicates an uncertainty regarding the future composition of the fungal flora. While *F. graminearum* has been known in Norway for 70 years, there are indications of a recent introduction of a more toxigenic type of the fungus. Also, there has been an increase in median DON concentration in cereal grain. The increase in prevalence of *F. graminearum* in Norway is similar to the situation in other European countries. To understand the shift in *Fusarium* species prevalence and to be able to develop effective management strategies it is important to get insight in to the interactions among FHB species and the effects on disease development and mycotoxin accumulation. Also, there have been recent changes in *Fusarium* nomenclature, like the recent description of *F. langsethiae* and the discovery in *F. graminearum* of chemotypes with different potential for mycotoxin production.

The biology and epidemiology of *F. langsethiae* are not well known, and there are uncertainties regarding the importance of the sexual stage in the epidemiology of *F. graminearum* in Norway. Spores discharged from fruiting bodies in spring and during the summer may be carried by air currents over long distances. However, the importance of sexual spores in spreading the pathogen within Norway is uncertain and remains to be investigated.

There are many domestic and international reports on within-field spatial variations and regional differences in the occurrence of *Fusarium* head blight. In addition there are variations from year to year in the prevalence of mycotoxin-producing fungi. Therefore, sampling to provide a true picture of prevalence in each field is complicated, especially at low infection levels. This uncertainty influences epidemiological studies of mycotoxin-producing fungi.

The ability to survive in plant residues for the relatively weak plant pathogens *F. langsethiae, F. sporotrichioides* and *F. poae* is not well known. There are also some uncertainties regarding the survival ability and the host range for *F. avenaceum* and *F. graminearum.*

Fungicides for control of *F. graminearum* or *F. culmorum* has been available for some years, and in Norwegian field trials the DON level has been halved, while *F. langsethiae* has not been affected. There is uncertainty regarding the durability of the fungicidal effect, because of the risk that *Fusarium* spp. develop fungicide resistance.

Environmental conditions suitable for infection and development

Residues from previous crops are important for the survival of *Fusarium* species in the field. Ploughing buries leaf and straw remains and reduces the disease pressure in the crop. In Norway reduced tillage is encouraged in order to minimize soil erosion and water pollution. However, this practice increases the risk of *Fusarium* infection and mycotoxin contamination of cereal grain. There is uncertainty on the Government policy for environment protection in the cereal growing areas of Norway.

Precipitation in the infection period from flowering to harvest is critical for infection and growth of mycotoxin-producing fungi in cereals. During the last five growing seasons there has been more precipitation than normal during cereal flowering period in Norway. It is not certain whether these humid summer months are representative of the future climate. There is also uncertainty how the different Fusarium species might respond on future climatic changes.

Also, uncertainties remain on the relative importance of various factors contributing to mycotoxin contamination in cereals. Factors that should be considered are host plants, crop rotation, tillage, fertilization, use of fungicides, regional and local topography, land use heterogeneity, distances to lakes/rivers, soil types and local weather conditions.

Difference in susceptibility among cereal varieties

Breeding for resistance to mycotoxin-producing fungi has high priority in Europe. Wheat varieties with moderate levels of resistance adapted to the Central European climate have been developed, but no resistant wheat varieties for Norwegian climate are available. More recently resistance breeding in barley and oats has been initiated.

The Norwegian cereal-breeding programme gives priority to *Fusarium*-resistance. Resistant cereal varieties developed in other Nordic countries may also be grown in Norway. Breeding for *Fusarium*-resistance has been difficult, and the number of *Fusarium*-species producing mycotoxins in cereals complicates the situation. Moreover, distribution of masked (glycosylated) mycotxins in the different varieties is unknown. Among current cereal varieties, there are some barley and oat varieties which seem to represent lower risk for DON contamination. There are uncertainties related to when resistant varieties will be available for Norwegian farmers and the effect of the resistance on each of the important mycotoxin-producing *Fusarium* species.

Animal health

Occurrence data

There may be uncertainties related to the representativeness of the sampling. The main part of the cereal grain samples originates from the national monitoring programs for mycotoxins in feed and food organised by the Norwegian Food Safety Authority. The sampling has been carried out according to the guidelines prepared by NSFA based on the EU guidelines to be representative of the levels in feed and food. Additional data from samples of cereal grains from specific studies analysed at NVI have been included, when the samples are considered to represent the levels in cereals for feed and food production. Feed samples are collected as random representative samples during production or are collected randomly from stores by NFSA or by the feed industry. As mycotoxins are unevenly distributed in batches the sampling is demanding to ensure representative samples. However with NFSA or the trade industry as sampling responsible, the uncertainty must be considered as low.

For uncertainties related to the analytical measurements, see Human health (below).

The quantitative estimation of occurrence of the mycotoxins DON and HT-2 + T-2 in animal compound feed is based on the occurrence in crude cereal grain during the last 15 years. This is a huge amount of data which ensure a relatively low uncertainty. The expected mycotoxin concentrations in various animal feed is based on typical feed recipes and estimated from Monte Carlo simulation. However, there are several factors influencing these mycotoxin estimates. The feed may contain other ingredients than those put in the model such as bran and maize, or some of the feed grain may be dehusked. Analysis results of these toxins in compound feed for most animal species are sparse or available primarily from the last couple of years. The occurrence of other mycotoxins than DON and HT-2 + T-2 in feed or feed incredients is far less known and the uncertainty is large.

Toxicological data

Enough data on toxicological effects, suitable to verify the critical effects after adequate time of exposure, the dose-response relationship, and an effect level or effect threshold are not the common case concerning the various mycotoxins and animal species. In fact, there are relatively few mycotoxins and animal species/productions where we can be satisfied with the quality of these data. That means, the uncertainty is large, and the levels/thresholds of critical effect is often a quite approximate number. Furthermore, effects of other mycotoxins or other bioactive compounds than whose studied may probably also have influenced the total effect. In some study reports such other compounds are measured and reported. With our knowledge on production, distribution and occurrence of mycotoxins and other bioactive compounds there is reason to assume that mixed exposure are common in studies which have used naturally contaminated feed, that means not a pure toxin added to the feed.

Mycotoxin exposure

In this risk assessment the animal exposure to mycotoxins is expressed as their occurrence concentrations in the cereal feed and its ratio in the total diet. As most reports from effect studies of mycotoxins in domestic animals present feed concentrations and stakeholders need the effect data based on feed concentrations there is no need to calculate doses based on body weight. This simplification contributes to reduce the animal exposure uncertainty.

Human health

This risk assessment is composed of several different parts, including the occurrence data, the food consumption data, the toxicology data and the overall assessment of the different parts. There are uncertainties associated with all parts of the risk assessment and this will be explained below.

Occurrence data

There are uncertainties related to the *representativeness of the sampling*. The food samples are taken during production and are considered to represent one day production at the mill. The samples for feed is, however, considerably smaller and may not be representative for the whole lot.

There are uncertainties related to the *analytical measurements*. The uncertainty is estimated to 40% by the laboratory. This uncertainty will, however, be to both sides of the true value. In a relatively large number of samples this uncertainty is likely to be lower. The uncertainties related to the analytical methods are probably of less importance in this risk assessment compared eith the uncertainties related to the large number of samples below the LOD.There is a large uncertainty related to the *occurrence* of nivalenol and T-2 and HT-2 toxins. The levels of these mycotoxins are below thier LOD values in all or the majority of the food samples. Calculating the intake of particularly the sum of T-2 and HT-2 toxins using either the LOD or 0.5 x LOD for calculations of the mean or median concentrations would result in an estimated intake exceeding the TDI.

The *human exposure estimates* are based on levels in flour. There may be a certain reduction during food processing, but these changes are considered to be small due to the high stability of the toxins.

It is well known that several mycotoxins exist in a bound form that is not detected by the current analytical procedures and therefore frequently labelled "masked mycotoxins". The occurrence of these bound forms is not included in the exposure assessment.

Dietary assessment

Every dietary assessment is connected with uncertainty. A description of the most important uncertainties and assumptions in the dietary exposure calculations is described below.

Three concepts are fundamental to understanding the limitations of dietary assessment: habitual consumption, validity and precision (Livingstone and Black 2003).

The habitual consumption of an individual is the person's consumption averaged over a prolonged period of time, such as weeks and months rather than days. However, this is a largely hypothetical concept; the consumption period covered in a dietary assessment is a compromise between desired goal and feasibility. In the Norwegian dietary surveys the time period covered are 14-days among the 1- and 2-year-olds (Sped- and Småbarnskost 2006/2007), four consecutive days among the 4-, 9- and 13-year-olds (UNGKOST 2000) and two none-consecutive days among the adults (Norkost 3).

In the present dietary exposure assessment the mycotoxin analyses are from 2008-2011, and based on the two mycotoxins DON and zearalenone. These mycotoxins have been analysed in different flours. The dietary intake of sum T-2 and HT-2 could not be estimated due to the high number of samples below the LOD. Scenarios were therefore made, based on assumptions of toxin concentrations equal to half the LOD in wheat flour and equal to LOD in wheat bran and oats.

When evaluating high consumers, the uncertainty associated with the 95th percentile is higher than for the mean value, especially among the age groups with a low number of participants. In addition to the uncertainty of the 95-percentile, the use of lowest and highest mycotoxin values of the four years 2008-2011 adds a level of uncertainty. For example, the 95-percentile based on the year with the highest toxin value indicates that the high consumers have always eaten food with the highest mycotoxin value.

The validity of a dietary assessment method refers to the degree to which the method actually measures the aspect of diet that it was designed to measure (Nelson and Margetts 1997). Lack of validity is strongly associated with systematic errors (Burema et al. 1988). With systematic errors all respondents in a dietary study or each subgroup in a population produce the same type of error, like systematic under- or overestimation of intake. All the three different dietary

assessment methods used in this risk assessment have limitations when it comes to validity. Results from validation studies among 9- and 13-year-olds indicate an underestimation of energy intake around 20% when the precoded food diary, used in UNGKOST 2000, is compared with energy expenditure (Andersen et al. 2005; Lillegaard and Andersen 2005). The validation studies among 1- and 2-year-olds were performed on a previously established questionnaire, but the results showed a significantly higher energy intake with the FFQ than with the weighed record reference method (Andersen et al. 2003; Andersen et al. 2004; Andersen et al. 2009). The Norwegian 24-hour recall method used among adults in Norkost 3 has not been validated. However, other similar 24-hour recall methods have been validated and show an underestimation in energy intake of around 15% (Subar et al. 2003; Poslusna et al. 2009). Underestimation of energy intake indicates that not all foods eaten are reported, but not which foods are underreported. It has been shown that foods perceived as unhealthy such as fats, sweets, desserts and snacks tend to be underreported to a larger degree than foods perceived as healthy (Olafsdottir et al. 2006). However, among children and adolescents there have been studies were this selective underreporting was not shown (Sjøberg et al. 2003; Lillegaard and Andersen 2005). As the different flours are found in foods perceived both as unhealthy and healthy, it is not likely that the misreporting would strongly bias the estimated mycotoxin exposure. However, if underreporting of mycotoxin containing foods is of the same magnitude as for total energy, the estimates for mycotoxin exposure are more likely to be underreported than overreported.

The precision of a technique is one that gives the same answer on repeated administrations (Livingstone and Black 2003). Poor precision derives from large random errors in the techniques of dietary assessment. The effect of random errors can be reduced by increasing the number of observations, but cannot be entirely eliminated (Rothman 2002).

The data collections of the different dietary surveys were performed from 2000 till 2011. Only the Norkost 3 data were collected within the time frame where the mycotoxin values were derived from (2008-2011). Dietary patterns are constantly changing. It is unclear to which extent a low participation rate will influence the assessment of mycotoxin exposure. It has been shown that health-conscious people are more likely to participate in a dietary survey. This can indicate a somewhat different dietary pattern among the participants than among the whole population. The direction of the uncertainty is difficult to estimate. Bread and flour-based ingredients are important staple foods for the majority of the population. Health-conscious people tend to choose more whole grain products, but this is will not influence the exposure estimates as the mycotoxin concentrations did not differ much between milled and sieved wheat flour (Table 19). However, higher concentrations were found for wheat bran, but bran contributed very little to the total exposure (Figure 11).

Individual consumption data reported in the dietary surveys have been paired with person specific self-reported body weights for the same individuals. However, where no bodyweight were given the mean bodyweight from the study were imputed.

Toxicological data

The toxicological databases are limited for some of the toxins having a *tolerable daily intake (TDI)*, for example nivalenol. For other toxins, such as enniatins, beauvericin and moniliformin no TDI have been established. The available toxicological information about these toxins is very limited and not sufficient to establish any safe intake levels.

Combined mycotoxin exposure in animal and human diets implies an uncertainty to the risk characterisation of the individual mycotoxins. Humans (and animals) are exposed to a variety of mycotoxins through the diet. The effects of such combinations remain uncertain.

The bioavailability, biotransformation and toxicological effects of the bound mycotoxins are not known and therefore the toxicological significance of these forms in food can currently not be evaluated.

Summary of uncertainties

Evaluations of the overall effect of identified uncertainties are presented in Table 45, highlighting the main sources of uncertainty and indicating whether the respective source of uncertainty might have led to an over- or underestimation of the exposure and/or the resulting risk (EFSA 2006b).

Table 45. Qualitative evaluation of influences of uncertainties on the assessment of mycotoxin exposure

Source of uncertainty	Direction and magnitude
Dietary exposure assessment	
Different dietary assessment methods	+/-
Measurement uncertainty in the mycotoxin concentrations analysed	+/-
Use of recipes (calculation from mixed food to flour)	+/-
Bias due to missed reporting/underreporting	+/-
Masked mycotoxins are not included in exposure assessment	-
Sped- and småbarnskost 2006/2007	
Use of 95-percentile	+/-
FFQ time span is 14 days	+/-
Infant porridge was only analysed for one year	+/-
Oat values were used for all infant porridges	+
Ungkost 2000	
Study conducted in 2000-2001 Possible changes in the food patterns can have occurred	+/-
Use of 95-percentile - The number of participants among 4-year-olds is only 391	+/-
- Low participation rate among 4-year-olds	+/-
Four registration days	+/-
Norkost 3, Adults	
Low participation rate	+/-
Two registration days	+/-
Occurrence of mycotoxins	
Annual variation (data for four years only)	+/-
Norwegian vs. imported grain	+/-
Sampling	+/-
Majority of samples below LOD (sum T-2 and HT-2 toxins, and NIV)	+
Analytical methods	+/-

+: uncertainty likely to cause over-estimation of exposure
- : uncertainty likely to cause under-estimation of exposure

In this risk assessment exposure estimates were presented as a range based on highest and lowest occurrence values of DON and zearalenone in different flours analysed in the years 2008-2011. Exposure scenarios for sum T2- HT2 and nivalenol has been performed.

Despite of the limitations in assessing the food consumption and the uncertainties related to estimating the dietary exposure outlined above, VKM concludes that the mycotoxin exposures presented in this opinion are within realistic ranges for each age group.

Data gaps

Data gaps identified in this opinion are divided into three disciplines corresponding to terms of reference: plant health, animal health and human health.

Plant health

The data for temporal and spatial variations in mycotoxin contamination contain considerable uncertainties. Therefore, a good overview of the range and scale of the variations in *Fusarium* epidemics and the production of mycotoxins in Norwegian cereals has yet to be established.

In particular, the potential effects of regional and local topography, land use heterogeneity, distances to lakes/rivers, and soil types are all plausible factors where new information could improve cropping practices. New robust insights into the role of such factors, supplementing existing knowledge, could assist in determining the relative importance of the various factors contributing to mycotoxin contamination in cereals.

The Norwegian early warning and prognosis models for DON contamination have not been able to provide sufficiently reliable predictions. As cereals are more susceptible to *Fusarium* and mycotoxin contamination at specific phenological developmental stages (flowering period), more focus should be put into developing matemathical models for prediction of phenological development in cereals. Large data sets and more knowledge on *Fusarium* epidemiology are needed to further develop mathematical models to predict the risk of *Fusarium* and mycotoxin development in cereals.

The role of long-distance spread in the *Fusarium* epidemics in Norway remains to be understood. If this is found to be an important factor, then it will call for coordinated management efforts, both in crop rotation and in tillage practices, in order to reduce inoculum levels. The inoculum potential of crop debris from different plant species commonly used in the rotations need to be further evaluated.

The opposing requirements, for decreasing erosion risks by reducing tillage on fields and decreasing mycotoxin-producing fungi by using ploughing, should be studied.

Sampling and analysis for fungal contamination and mycotoxin content are challenging operations. Fungal growth is not uniform in the field, and it is difficult to obtain representative samples at delivery. It is also difficult to sample grain transporters and silos as they contain a mixture of grain from producers with different contamination levels.

Resistance to *Fusarium* has previously not been included in official cereal variety trials. Therefore, resistance to mycotoxin-producing fungi should also be evaluated in field trials with barley, oats and wheat. In breeding for enhanced resistance to *Fusarium* head blight, focus has been mainly directed onto reduction of DON. It needs to be further clarified whether resistance to DON producers corresponds with resistance to T-2 and HT-2 producers.

Today, all grain lots of oats and also wheat lots intended for food consumption are analysed for DON at delivery, this to reduce the risk of DON contamination of food and feed. As there is a non-linear negative association between DON and T-2 and HT-2 contamination in oats, some grain lots accepted for food and feed purposes due to a low DON contamination might contain T-2 and HT-2 values above suggested legislation limits. More emphasis should be put into understanding the effect of climate and agricultural practices on development of T-2 and HT-2.

In fungicide trials, more emphasis should be given to efficacy in *Fusarium* control. Focus should also be directed into control of T-2 and HT-2 contamination in cereals, as no fungicide treatment has yet proven to reduce these toxins in field trials.

The influence on mechanical, chemical, or biological treatments of cereal crop residues on survival and development of *Fusarium* inoculum should also be further studied.

Animal health

There is lack of knowledge on the factors influencing the broad range of levels for critical effect of DON in growing pig. Are these factors caused by animal breed differences in myctoxin sensitivity, or other mycotoxins/bioactive compounds in the feed or in the environment, or is it due to other environmental factors?

There is lack of knowledge on effects of DON, zearalenone and other mycotoxins on sows during gestation and lactation. Several cases from the field report on problems where mycotoxins are suspected as at least part of the problem.

Ochratoxin A has been shown to inhibit performance of growing pigs as well as reduce semen quality of boars at far lower feed concentrations than where the more well known nephrotoxicity are observed. More effect data of ochratoxin A on pig performance and semen quality are needed.

There is need for more knowledge on effects of mycotoxin binders/inactivators used as feed additives.

HT-2 is supposed to be of similar toxicological potency as T-2, as T-2 is rapidly deacetylated to HT-2 *in vivo*. However, effect studies of HT-2 in animals are lacking.

There are indications of increased susceptibility to DON and T-2 and HT-2 and possibly also other mycotoxins in intensively fed ruminants. However, the data are few and fragmentary, and more studies are necessary on this topic.

Effect data on T-2 and HT-2 in horse are lacking. These toxins are abundantly present in oats which traditionally is an important feed article for horses due to high nutritional and energetic value. Data on critical effects and dose-response relationships of these toksins in horses are needed.

Fumonisin effect data in dogs are lacking. Fumonisins are contaminants of concern in maize, and feed for dogs may contain considerable amounts of maize. Thus, data on critical effects and dose-response relationships of fumonsins in dogs are needed.

Enniatins and beauvericin are mycotoxins of low knowledge in spite of abundant occurrence in cereals and rather high *in vitro* toxicity. Their bioavailability after oral exposure may be restricted but examination of possible local intestinal effects would be important.

Effect data of co-exposure of mycotoxins at subclinical/critical effect levels for each of them from animal model studies are lacking, and strongly needed.

There are lack and thus need for more occurrence data on mycotoxins in feed ingredients. Maize is a feed ingredient of particular mycotoxin concern. Data on occurrence of DON, T-2 and HT-2, zearalenone, fumonisins, ochratoxin A and aflatoxins in feed maize are lacking in Norway.

There are lack and thus need for more occurrence data on various mycotoxins (DON, T-2 and HT-2, zearalenone, fumonisins, ochratoxin A, aflatoxins) in compound feed for most animal

species. DON and T-2 and HT-2 in feed for pigs are now followed closely but there are few data on compound feed for most other species. More knowledge on mycotoxins in stored feed for terrestrial and aquatic animals is also necessary.

Occurrence data on mycotoxins of emerging risk as *Alternaria* toxins, ergot alkaloids and enniatins in feed and food are lacking and needed.

Human health

There is a lack of both occurrence and toxicity data for several of the mycotoxins, particularly for the emerging mycotoxins enniatins, beauvericin and moniliformin. The growth conditions for the *F. avenaceum* and *F. poae*, the main producers of these toxins, have improved because of milder springs and wetter summers in Norway in the last years. Therefore, an increase of toxic metabolites in grains and grain-based products can be expected.

The occurrence data for the most important mycotoxins, DON and the sum of T-2 and HT-2 toxins, in Norwegian food products are scarce. A more systematic surveillance should be performed, especially focussing on products with high wheat and oat contents.

More sensitive analytical methods for T-2 and HT-2 toxins and nivalenol should be established ascertaining that the used LODs are low enough to measure low toxin concentrations. This will generate better data sets and will improve exposure assessment.

Naturally *Fusarium*-infected grain is more toxic than grain with the corresponding amount of pure toxin added. It is therefore likely that some unknown factors in the naturally infected grain considerably enhance and add to the toxic effects. There is a need to identify these factors. Currently, there is a focus on the role of bound forms of the mycotoxins, the so-called masked mycotoxins. The occurrence and toxicological significance of these derivatives of mycotoxins, such as DON and T-2 toxins is presently unknown.

Fungi produce more than one mycotoxin. Furthermore, grain is commonly infected with many species simultaneously. Humans are consequently exposed to mixtures of mycotoxins. The effects of such mixtures remain unknown. Additionally, the grain may contain other biological and chemical contaminants with biological activities, such as bacteria, pesticides etc. Potential interactions between these contaminants and mycotoxins are currently unknown.

The consumption of maize-based products and rice in Norway has increased in recent years, substituting potatoes that are less used (according to statistics from The Federation of Norwegian Agricultural Co-operatives). In 2010, the net import to Norway of maize including maize flour, maize starch, maize flakes (cornflakes), and blended products was 7 million kg (Helsedirektoratet 2011. Utviklingen i norsk kosthold. Matforsyningsstatistikk og Forbruksundersøkelser). Thus, Norwegian consumers might be exposed to maize-specific mycotoxins at higher extent than before. The monitoring of maize-based products for mycotoxins such as aflatoxin, zearalenone and fuminosin, but also for DON, appears to be urgently needed.

Conclusions

The conclusions of this opinion are divided into three disciplines corresponding to terms of reference: plant health, animal health and human health.

Plant health

Mycotoxin-producing fungi

- The most important mycotoxin-producing fungi infecting cereals during the growing season in Norway belong to the genus *Fusarium*. The most important storage fungi are species of *Aspergillus* and *Penicillium.* All three genera contain several species with different potentials for mycotoxin production.
- *Fusarium* species can cause seedling blight, root rot and *Fusarium* head blight. Seedling blight and infection on roots and lower parts of the straw may reduce the plant density and cause yield losses. *Fusarium* infection and growth in the head from flowering and up to harvest may, in addition to causing yield losses, result in mycotoxin production and accumulation.
- Seed infection is a pathway for long-distance dispersal and introduction of pathogens to disease-free fields. Between seasons, the *Fusarium* spp. survive as saprophytes in plant debris on the soil surface and in the upper soil layer. *Fusarium* spp. can produce large quantities of spores that are disseminated to cereal plants by water splash or air currents. The spores germinate under humid conditions on floral parts. Infection of cereal grains starts at flowering, when the plants are most susceptible, and continues towards harvest. In the initial stage, anthers and pollen may serve as a food-base for the pathogen. Sexual spores of *F. graminearum* can be dispersed over long distances by air currents.
- In Norway, the most common *Fusarium* species is *Fusarium avenaceum*, while *F. graminearum* has increased in prevalence during the last 10 years. *Fusarium langsethiae, F. crookwellense, F. culmorum, F. equiseti, F. poae, F. sporotrichoides*, *and F. tricinctum* are other mycotoxin-producing species that are often identified in Norwegian cereals.
- Deoxynivalenol (DON) is the most commonly produced mycotoxin in the field in Norway and other temperate areas, and *F. graminearum* and *F. culmorum* are the most important DON producers. Also, enniatins, produced by *F. avenaceum,* are common in Norwegian cereals. *Fusarium langsethiae* is the most important producer of the mycotoxins T-2 and HT-2 in the field. Other *Fusarium* spp. produce lower levels of the same mycotoxin. Several *Fusarium* spp. produce low levels of nivalenol, zearalenone, and moniliformin in the field.

Occurrence of mycotoxins

- DON is found in practically all samples of crude cereal grain, compound feed for animals and cereal food products such as flour, bran and oats flakes. In wheat, the annual mean DON concentration ranged from 60 µg/kg to 400 µg/kg in the period 2001-2011. In oats, the annual mean DON concentration ranged from 40 µg/kg to 2150 µg/kg in the same period.
- T-2 and HT-2 toxins are also widespread, particularly in unprocessed oats, but also in other grains such as barley. The annual mean levels in crude oats grain varied from 20

µg/kg to 50 µg/kg for the T-2 toxin, and from 50 µg/kg to 250 µg/kg for the HT-2 toxin in the period 2001-2011.

- Zearalenone has been found in cereal grains, particularly in oats. It has only been found at very low concentrations in processed grain products intended for human consumption.
- Norway import cereals grown in other countries. These might be contamineded with mycotoxins or levels of mycotoxins other than found in Norwegian-grown cereals. The countries from which Norway imports cereals and cereal products changes continuously.
- *Penicillium* and *Aspergillus* spp. produce mycotoxins during storage, but only low levels of storage mycotoxins have been detected in Norwegian cereals. Ochratoxin A is produced by fungi in these genera. Aflatoxins are produced primarily by the two species *A. parasiticus* and *A. flavus*.
- Ochratoxin A is a storage mycotoxin of concern in Norwegian-produced grain. The occurrence of ochratoxin A in stored grain has been analysed annually in a limited number of samples. The concentrations found in stored Norwegian grain have been low, indicating that the present handling and storage procedures are sufficient to limit the production of ochratoxin A. Aflatoxins does only occure in grains imported from tropical climates, i.e. mainly in maize, but have also been found in basmati rice.
- VKM notes that with regard both to ochratoxin A and aflatoxins non-grain food items are important sources of human exposure in the Norway. But assessment of this exposure is outside the terms of reference of this opinion.
- The results from a pilot study on ergot and ergot alkaloids in rye, barley and wheat in Norway indicates that ergot and ergot alkaloids may be a more significant problem in Norwegian cereal than so far considered.

Cereal species and varieties

- In Norway, oats generally have higher mycotoxin content than other cereals. Winter wheat contains less mycotoxins than spring wheat. There seem to be minor differences in mycotoxin content between cereal varieties. There are regional and annual variations in mycotoxin concentrations in all cereal species.

Variation over time

- *Fusarium avenaceum* and *F. culmorum* both develop under rather cool and humid conditions. *Fusarium graminearum* prefers a warm and humid climate, while *F. langsethiae* is associated with relatively dry and warm conditions. During the last ten years, *Fusarium* infections of cereal seed have more than doubled in oats, barley and spring wheat, compared with the three previous decades. During most of the last ten years, precipitation during the critical period from flowering to harvest has been above average.
- In accordance with an increased infection rate of *Fusarium graminearum* the last decade show a strong increase in the median concentration of DON in crude grains of oats and alsoin wheat but less so.
- Since *Fusarium langsethiae,* until it was described in 2004,was identified as *F. poae*, it is not possible to determine whether any changes in long-term trend in its prevalence

have taken place. In contrast to DON, there are no clear change in trends in the occurrence of the T-2 and HT-2 toxins in crude grains during the last decade.

- Other mycotoxins than the trichothecenes have not been systematically surveyed in cereal grain in Norway, and time trends are therefore not known. It could be relevant to pay more attention to zearalenone which is produced by the same *Fusarium* species as DON.

Regional differences

- In some years there are regional differences in mycotoxin content, but these are not consistent over time. The recent use of fungicides in districts with historically severe *Fusarium* head blight epidemics may mask the differences between districts.

Resistant cereal varieties

- Growing resistant cereal varieties would reduce the risk for mycotoxin contamination of cereals, but complete resistance to *Fusarium* head blight in cereals has never been achieved. Wheat breeders have developed varieties with partial resistance to *Fusarium* head blight, but so far no highly resistant varieties that are adapted to the Norwegian climate have been introduced. There are slight differences in *Fusarium* head blight resistance between barley varieties. Increased *Fusarium* head blight resistance has high priority in Norwegian cereal breeding. Among new, Norwegian oat varieties, some seem to have moderate *Fusarium* head blight resistance.

Pesticides

- Some fungicides in the triazole group reduce the DON content of cereals by 50 % compared with untreated controls. The timing of fungicide application is critical. Best *Fusarium* head blight control is achieved when fungicides are sprayed at flowering. Previously, oats were not sprayed with fungicides, but during recent years spraying to control *Fusarium* head blight in oats has become more common practice. There has been no reduction of T-2 and HT-2 contamination after fungicide application in oats. In districts with frequent *Fusarium* head blight problems, barley has also been sprayed at flowering.

Agricultural practices

- As *Fusarium* spp. survive in crop residues, tillage and crop rotation are important to decrease the risk of *Fusarium* head blight and mycotoxin contamination. Ploughing reduces the amount of plant material at the soil surface more than harrowing and other reduced tillage practices. In order to decrease soil erosion and water pollution, the use of reduced tillage is encouraged in Norway. However, this practice increases the amount of plant residue on the soil surface and may also increase the risk for mycotoxin contamination of cereal grain.

- Fields with cereals as the previous crop are at higher risk of *Fusarium* head blight and mycotoxin contamination than fields with non-cereals in the rotation. The main DON-producer, *F. graminearum*, has airborne spores enabling long distance transport from surrounding fields. Hence, the positive effects of crop rotation and tillage in a single field may thus be reduced by airborne spores.

- Similar to the effect of rainfall, it is reasonable to believe that irrigation in the period around heading and flowering also increases the risk of *Fusarium* head blight and mycotoxin contamination.

- An integrated approach, with a combination of crop rotation, ploughing, growing cereal varieties with the best available resistance, and fungicide treatment at flowering, is a strategy for controlling *Fusarium* head blight and reducing mycotoxin contamination of cereals.

Climate factors

- Temperature and humidity from flowering until harvest determine the risk of *Fusarium* infection and production of mycotoxins in the cereal grains. One of the main challenges for cereal cultivation in Norway is an extensive cereal monoculture, with limited use of crop rotation, combined with reduced tillage. Under humid conditions these factors may contribute to increased occurrence of mycotoxin-producing fungi.
- During the last decade, an increased precipitation in July, which is the flowering time for spring cereals, has occurred in parallel with an increase in mycotoxin contamination of cereals. In the same period, *F. graminearum* has increased in frequency.
- The relationship between weather, disease incidence and severity of mycotoxin-producing fungi, has prompted extensive development of mathematical models for mycotoxin risk management. When linked to a real-time weather monitoring system, model predictions provide information about the need for, and timing of, fungicide application.
- The yearly average temperature in Norway is expected to rise by between 2.3 and 4.6 °C by year 2100. This will lead to an earlier flowering in cereals. One important question concerns the level of precipitation during the flowering period. Precipitation in the flowering period promotes the occurrence of mycotoxins in cereals and is, therefore, a critical factor. In the last five growing seasons there has been more precipitation than normal in the flowering period for cereals in Norway. If such weather conditions are representative of the future climate in Norway, then we can expect significantly increased problems with mycotoxins in cereals in the years to come.

Organic farming

- There are several reports from field trials in which organically grown cereals have been shown to contain less mycotoxin than the equivalent cereals grown with mineral fertilizers and pesticide application. Others report that there are no differences between the two cropping systems. Some of these studies were conducted before the recent approval of fungicides that control DON-producing fungi. In organic farming, ploughing is common and the cropping system frequently includes non-cereal crops. Farmers with monocultures of organically grown cereals practice fallow periods to control weeds. These are all factors that reduce the risk for *Fusarium* head blight and mycotoxin contamination of cereals.

Storage conditions

- In Norway, about one third of the domestic grain is stored on the farms for a period following harvest. Most years, and especially in wet autums, the grain has to be dried to prevent fungal growth during storage. Cooling will also slow down the growth of storage fungi. Commercial grain is commonly traded at moisture content of 15 %. For long term storage grain should be dried to 14.5% or less.

Animal health

The trichothecene mycotoxin levels in prepared compound feed are often related to the content of oats in the formulations. The composition of cereal grain in different feed recipes may vary considerably for different species and there are different recipes for feed for the same species as well.

Addition of chemical adsorbants, though important in the treatment of aflatoxin-contaminated grain, has so far not been documented to be effective against trichothecenes in feed. There may be a risk for binding of chemical adsorbants to other kind of substances including important nutrients. Among potential mycotoxin-degrading microbes tested, the rumen bacteria BBSH 797 has shown good trichotecene-degrading effect *in vitro*, but the product needs further testing of the trichotecene-reducing and clinical effect *in vivo*. If found to be effective, the risk in using these products in feed is expected to be insignificant.

VKM has decided to use feed concentrations as a dose metric (instead of amount/kg body weight) in this risk assessment. Animal exposure in this report is therefore expressed as dietary concentrations.

The risk characterisation of the mycotoxins in feed combines the data of the hazard characterisation for each animal with the corresponding data of exposure. The resulting characterisation of the risk is categorized as negligible, low, moderate or high for the various mycotoxins and the various animal species or production forms.

- **Pig**

  Pig is particularly sensitive to mycotoxins compared to other domestic animals.
  - **Deoxynivalenol (DON)** reduces performance and animal welfare and there is **high risk** for such effects due to its considerable occurrence in cereal grains, even if the current use of oats and maize in Norwegian pig feed is low. The adverse effect levels vary considerably among feeding studies with growing pigs (NOAELs vary from 0.6 to 0.9 mg/kg feed and LOAELs from 0.35 to 2 mg/kg feed). The current analysis results of DON in feed for pigs show mean and median concentrations around 0.4 mg/kg and a 99 percentile at 1.5 mg/kg. This occurrence level may therefore reduce feed intake and performance in the pig barns.
  - **T-2 and HT-2 toxins**: The risk for effects of these more potent trichothecenes in pigs is regarded to be low. Most of the current feed samples seems to be below the adverse effect level. The critical effects are haematological and immunological. The benchmark dose level for T-2 derived by EFSA is 0.17 mg/kg diet. There is lack of toxicity studies of HT-2 in pigs as in other species but its potency is generally regarded to be similar as of T-2.
  - **Nivalenol**: The risk for effects is regarded as negligible due to the low occurrence of this trichothecene.
  - **Zearalenone**: The risk for adverse effects of this estrogenic mycotoxin is considered low. NOAEL is at 0.2 mg/kg feed, and the current use of oats and maize in Norwegian pig feed is limited.
  - **Fumonisins:** The risk for effects is low to negligible with our identified NOAEL at 1 mg/kg diet (No Observed Effect Level (NOEL) at 0.3 mg/kg diet) for pulmonary and performance effects and with the low use of maize in Norwegian pig feed.
  - **Enniatins and beauvericin**: Toxicity data are missing. Due to relatively low and very low occurrence, respectively, and assumed low bioavailability in all species,

the risk for adverse effects is considered to be low and negligible, for enniantins and beauvericin, respectively.

- **Moniliformin**: The risk for adverse effects is negligible due to low occurrence and low toxicity (identified NOAEL 25 mg/kg diet).
- **Ochratoxin A:** EFSA derived a LOAEL for nephrotoxicity at 0.2 mg/kg feed, which is about 1000 times higher than the mean level of ochratoxin A in cereals in Norway. However, more recent findings show reduced sperm quality in boars at 0.003 mg/kg as well as reduced performance of growing pigs at 0.025 mg/kg feed, which indicate a moderate to high risk for adverse effects .
- **Aflatoxins:** The risk for adverse effects is negligible. Only imported ingredients, particularly maize, may contain aflatoxins. However, the levels are shown to be low in maize used in pig feed. The NOEL is 0.2 mg/kg diet for liver effects.

- **Poultry**

  The sensitivity for mycotoxins in poultry may vary considerably for different species and production forms but is in general less than for pigs.

  - **DON**: The risk for effects in broiler chickens and laying hens is considered moderate and low respectively. Recent studies on broiler chicken have shown adverse effects on intestinal villi and immune responses (LOAEL 1.7 at mg/kg diet). Laying hens are more tolerant than chicks, however in breeder hens there is a risk for foetal effects.
  - **T-2 and HT-2**: The risk for effects is regarded as moderate for broiler chicken, turkeys and ducks, and lower for laying hens. T-2 and HT-2 show LOAELs at 0.2-0.5 mg/kg diet in broiler chicken, turkeys and ducks based on oral lesions or reduced weight gain, and 2 mg/kg diet in laying hens based on reduced production and hatchability.
  - **Nivalenol**: The risk for adverse effects is regarded as negligible due to the low occurrence of this trichothecene.
  - **Zearalenone**: The risk for adverse effects is negligible under normal practical feeding conditions as poultry are rather tolerant for this toxin.
  - **Fumonisins**: The risk for effects is considered low. LOAELs for fumonisin B1 are 20 mg/kg diet for broiler chicken and 5 mg/kg for ducks based on increased sphinganine/sphingosine ratio. Fumonisins are found in maize, of which poultry may be fed on relatively large amounts. However, the risk of reaching levels that produce adverse effects is considered as low.
  - **Enniatins and beauvericin**: The risk for adverse effects is considered low and negligible, respectively, due to relatively low and very low occurrence, respectively, as well as suggested low bioavailability of both toxins and low toxicity of beauvericin shown in chicken.
  - **Moniliformin**: The risk adverse for effects is regarded as negligible. Effects in broiler chicken (liver toxicity) and turkeys (enlarged heart) were shown at far higher levels than measured in Norwegian cereals.
  - **Ochratoxin A**: The risk for adverse effects is considered as low. LOAELs are 0.1-0.5 mg/kg diet in broilerchickens, based on immunological responses. The concentrations measured in Norwegian cereals are lower than those expected to produce effects.
  - **Aflatoxins**: The risk for adverse effects may be low in ducks and negligible in other poultry species. Aflatoxins show different toxicity in various species, ducks being about 10 times (effect level 0.030 mg B1/kg diet) more sensitive for liver

toxic effect than broiler chickens and turkeys, and far more sensitive than laying hens.

- **Ruminants**

  The sensitivity for mycotoxins in ruminants is generally low. However, disturbed rumen metabolism due to e.g. excessive amounts of compound feed may increase the sensitivity to mycotoxins.

  - **DON**: The risk for adverse effects in healthy ruminants fed normal amounts of compound feed (below about 50 % at dry weight diet) is considered negligible. Increased risk of adverse effects to DON is observed if fed higher amounts of compound feed.
  - **T-2 and HT-2**: The risk for adverse effects is low. Although there are indications of ruminants being rather sensitive the exposure is low with the current restricted level of oats, and with roughage as a major feed component.
  - **Zearalenone**: The risk for reproductive effects is considered low for ewes and regarded as negligible for heifers due to lower sensivity.
  - **Fumonisins, ochratoxin A and aflatoxins**: The risk for adverse effects is negligible due to low sensitivity
  - **Enniatins, beauvericin and moniliformin**: Toxicity data on ruminants are lacking, but the risk is considered negligible due to relatively low occurrence, and assuming the same low bioactivity as in other species.

- **Horse**

  Horses show moderate sensitivity to a range of mycotoxins. As the major horse diet is roughage and horse owners show more care with using the traditionally important oats for horses, the exposure of mycotoxins via cereals are limited.

  - **DON**: The risk for adverse effects is considered low, when fed a normal diet based on roughage and a normal level of oats in the compound feed at about 30 %. Horses fed DON at approximately 5 mg/kg total diet have shown reduced feed intake and body weight gain or biochemical changes.
  - **T-2 and HT-2**: The risk for effects is normally considered as low. Toxicity data on T-2 and HT-2 in horses are very limited, but feed refusal and skin lesions were observed fed T-2 at approximately 0.7 mg/kg total diet.
  - **Zearalenone**: The risk is regarded negligible. Approximately 1 mg/kg total diet did not produce adverse effects in horses.
  - **Fumonisins**: The risk for adverse effects in Norwegian horses is regarded negligible, as horses normally ingest low amounts of maize. Fumonisin B1 below 6 mg/kg total diet is regarded to be safe regarding encephalomalicia in horses.
  - **Enniatins, beauvericin and moniliformin**: Toxicity data on horses are lacking but the risk is considered negligible due to relatively low occurrence, and assuming the same low bioactivity as in other species.
  - **Ochratoxin A and aflatoxins**: Toxicity in horses is unknown. The low exposure indicates negligible risk of these toxins.

- **Rabbit**

  The toxicity data base for mycotoxins in rabbit is scarce, but available data indicates a moderate sensitivity to various mycotoxins.

- **DON and T-2 and HT-2:** A low risk for adverse effects is indicated from the effect data base and the assumed exposure.
- **Zearalenone and fumonisins**: A moderate risk for adverse effects is indicated from the toxicity data base and the assumed exposure.
- **Enniatins, beauvericin and moniliformin**: Toxicity data on rabbit are lacking, but the risk is considered negligible due to relatively low occurrence, and assuming the same low bioactivity as in other species.
- **Ochratoxin A and aflatoxins**: The risk for effects may be negligible as their effect levels are far higher than the measured concentrations in small grain cereals and maize in Norway.

- **Dog and cat**

  The knowledge on the toxicity of mycotoxins in dog and cat is generally limited.
  - **DON**: The risk for adverse effects in dog seems to be low based on data on reduced feed intake and weight gain at 2.7 mg/kg diet and the sparse knowledge of DON in dog feed sampled in Norway (mean 0.41 mg/kg). For cat the risk is considered to be negligible based on an estimated effect threshold at 7.7 mg/kg diet and the lower level of cereals in cat feed.
  - **T-2 and HT-2**: The risk for effects is considered negligible for dog. Due to lack of effect data for T-2 and HT-2 in dog, an effect threshold is estimated by EFSA from the bench mark dose level in pig. This estimated effect level is more than 10 times higher than the mean concentration of sum T-2 and HT-2 in dog feed sampled in Norway. Cats are particular susceptible to T-2, but the data do not allow the determination of adverse effect levels. Feed for cats generally contain lower amounts of cereals than that of dog feed and the risk for cats is still considered to be low.
  - **Zearalenone**: The risk for adverse effects is considered low for dogs based on our identified LOAEL (metabolic changes) at approximately 1.3 mg/kg diet, which is about 10 times higher than the mean concentration in dog feed sampled in Norway. There are no data for cats, but with lower amounts of cereals in cat feed the risk is considered negligible.
  - **Fumonisins**: The risk for adverse effects may be considered low for dog based on the low fumonisin concentrations shown in samples of dog feed in Norway and despite the fact that the fumonisin commodity maize is an ingredient of concern in dog feed. Adverse effect levels for fumonisin in dogs and cats have not been identified. In lack of toxicity data the effect level found in pig as a sensitive monogastric species is used in the assessment of dogs and cats. With a lower level of maize in cat feed the risk for cats is considered negligible.
  - **Enniatins, beauvericin and moniliformin**: Toxicity data on dog and cat are lacking, but the risk for effects is considered negligible due to relatively low occurrence of these toxins, and in addition assuming similarly low bioactivity as in other species.
  - **Ochratoxin A**: The risk for effects is considered negligible for dog. Ochratoxin A has produced nephrotoxic effects in dog at approximately 5 mg/kg diet (our identified LOAEL) and above. This level is 4-5 orders higher than ochratoxin A levels shown in Norwegian cereals and as measured in dog feed sampled in Norway. There is no data for cats, but with a large margin of exposure for dogs, the risk is also considered negligible for cats.
  - **Aflatoxins**: The risk for adverse effects is considered low to negligible for dogs. Hepatotoxicity occur above 0.2 mg/kg diet (NOAEL), which is far above the trace

concentrations found in the samples of maize and dog feed, analysed in Norway. However, aflatoxins are known to show a very variable occurrence in maize and other tropical feed ingredients. This implies that a risk can not be totally ruled out. For cat there is no toxicity data, but with lower amounts of feed ingredient sources in cat feed that may contain aflatoxin, the risk is considered lower than for dog.

- **Fish (primarily salmon and trout)**
  - **DON, T-2 and HT-2, zearalenone, fumonisins and ochratoxin A:** The risk for adverse effects is considered as negligible based on the maximum theoretical concentrations of the mycotoxins in the feed for salmon and rainbow trout and the effect data available. Storage of fish feed at the farm level may affect levels of mycotoxins, today no data exist showing actual concentrations in feed when fed to the fish. Future surveillance should therefore include samples taken at the farm level.
  - **Enniatins, beauvericin and moniliformin**: Toxicity data on fish are lacking but the risk for adverse effects is considered negligible due to relatively low occurrence, and assuming a similarly low bioactivity as in other species.
  - **Aflatoxins**: The risk for effects in rainbow trout is regarded to be moderate, as the highest theoretical level in salmonid feed is at the same level as the LOAEL (0.0004 mg/kg diet). For Atlantic salmon there is lack of effect data.

Human health

During the years 2008-2011, selected *Fusarium* toxins were analysed in commercially available wheat and oat products for human consumption, such as flour, bran, groat, and oat flakes. Commercial powder-based porridges and breakfast cereals were analysed for trichothecenes and zearalenone, and maize products for fumonisin. For the human exposure assessment, VKM chose to use the lowest and highest mean mycotoxin concentrations of the four years (2008-2011) for each of the four flour products: sieved wheat flour, milled wheat flour, wheat bran and oat flakes. This choice was made in order to take the annual variations in fungal infections and toxin concentrations into account. Since the samples represent the available food in the market, the potential mixture of grains from several harvest years is taken into account.

The current exposure to some mycotoxins in grain-based products represents a concern for some consumer groups in Norway.

- The contribution from animal–derived food items to the dietary intake of mycotoxins is low and of little significance compared to the intake from plant sources. Ochratoxin A is the only mycotoxin assessed in this report with known potential for transfer from feed to humans through meat consumption due to long plasma half-life. Even so, other intake estimates have shown that a maximum of 10% of the total dietary intake of ochratoxin A is from consumption of meat, milk and eggs. Therefore, VKM considers that with the present levels of ochratoxin A in Norwegian grain, the transfer of ochratoxin A from animals to humans is low. Aflatoxin M1 may be excreted in milk from dairy cows receiving feed contaminated with aflatoxins B1. This is, however, not a risk to human health with the current levels of aflatoxins in cattle feed.

The dietary exposure to DON and zearalenone, respectively, were estimated based on occurrence data in Norwegian cereal products and consumption data from national dietary surveys. The exposures were compared with internationally derived TDIs for the respective

toxins. Scenarios for the intake of sum of T-2 and HT-2 toxins and nivalenol have been performed. The estimated intakes from these scenarios were compared with the corresponding TDI.

In the characterization of risk, VKM makes use of the terms "of no concern" or "of concern" depending on whether the calculated exposure is below and/or close to, or exceeding, the respective TDI. Human exposure below or close to TDI is considered to be of no concern, whereas exposure exceeding the TDI is considered to be of concern. Correspondingly, for characterization of risk for acute dietary exposure, the calculated exposure via one single meal is compared to ARfD. Acute exposure similar to or below the ARfD is considered to be of no concern.

Deoxynivalenol (DON)

Deoxynivalenol (DON) is the main mycotoxin of concern in Norwegian grain. It is the most prevalent mycotoxin in Norwegian grain and is present in virtually all samples of flour and oat flakes.

- The estimated mean and high (95-percentile) exposures to DON in years with low mean concentration of DON in the flour, respectively, were in the range of, or exceeded the TDI by almost 2 times in 1-year-old infants and 2-year-old children. In years with high mean DON concentration, the high (95-percentile) exposures exceeded the TDI up to 3.5 times for 1-, 2-, 4- and 9-year-olds. VKM concludes that exceeding the TDI at mean or high exposures to DON in infants and children is of concern, although the TDI is not a threshold for toxicity.
- The estimated dietary intakes of DON in adolescence and in the adult population are equal to or below the TDI and are therefore not a health concern.
- Acute exposure to DON is equal to or below the ARfD and is therefore of no concern.

Zearalenone

- The estimated intakes of zearalenone are below the TDI for all age groups. Exposure to zearalenone is of no concern for all age groups.

Sum of T-2 and HT-2 toxins

The dietary intakes of sum of T-2 and HT-2 toxins could not be estimated due to the high number of samples below the limit of detection. Scenarios were made to illustrate the potential intakes of sum of T-2 and HT-2 toxins. The scenarios are considered to represent an overestimation.

- These scenarios indicate that the dietary intake of the sum of T-2 and HT-2 toxins in 1- and 2-year-olds may exceed the TDI, while the 4-year-olds with high exposure have an intake in the range of the TDI. According to the exposure scenarios, the exposures to the sum of T-2 and HT-2 toxins in 9- and 13-year-olds are below the TDI. Furthermore, both the mean and high exposures in adults are below the TDI. VKM concludes that according to the performed exposure scenarios, the dietary intake of the sum of T-2 and HT-2 is potentially of concern for the youngest age groups.

Nivalenol

The dietary exposure to nivalenol in Norway could not be estimated due to a high number of samples that were below the limit of detection.

- The scenarios for intake of nivalenol were below the TDI for all age groups. The intake of nivalenol is of no concern for all age groups.

Other mycotoxins

- The intake of fumonisins was not estimated due to few occurrence data. Maize and maize-based products are the only commodities known to contain significant levels of fumonisins. Due to the low maize consumption in Norway, fumonisins in grain-products are considered to be of no concern.
- The levels of ochratoxin A in Norwegian grain is low and is considered to be of no concern for human health in Norway.
- Aflatoxins are normally not produced under the conditions of Norwegian climate. Occurrence data for aflatoxins in Norwegian grain-based food products were not available. VKM considers the potential occurrence of aflatoxins in Norwegian grains to be of no concern for human health.
- No assessment of the emerging mycotoxins enniatins, beauvericin and moniliformin in grain could be made due to the lack of occurrence data, as well as toxicity data for these toxins. VKM recognizes the presence of moniliformin, enniatins and beauvericin in Norwegian grains, which potentially might be of risk for human health.

Concluding remarks

In cereals, the most important mycotoxin-producing fungi are *Fusarium* species, which infect cereals during the growing season and cause yield loss and mycotoxin contamination of the grain. With the current occurrence of mycotoxins in grains in Norway VKM identified deoxynivalenol (DON) as the main mycotoxin of concern for human and animal health.

- Pig is in comparison to other domestic animals, particularly sensitive to DON. At current levels of DON in pig feed there is a high risk that DON reduces the pigs´ performance and welfare.

- For humans, the calculated exposure shows that infants and children exceed the TDI for DON from consumption of flour and oat flakes. Exceeding the TDI is of concern, although the TDI is not a threshold for toxicity.

Because of these findings and since cereal grain is an important feed and food ingredient VKM is of the opinion that steps to reduce levels of DON in feed and foods are warranted.

- VKM identified a number of agricultural factors influencing the *Fusarium* infection rate and consequently the mycotoxin production such as crop rotation, ploughing, resistance of cereal varieties, and fungicide treatment at flowering.

- VKM also notes that given the content of mycotoxins in flour for use in Norway attention should also be paid to the content of mycotoxins in imported grains.

- Special attention should be paid to cereals intended for infants and children.

Although there is a large annual variation in the occurrence of DON, VKM notes that during the last decade, parallel to an increased precipitation during the flowering period, there has been a strong increase in the infection rate and occurrence of DON in oats and wheat. Future climate change in Norway, with increased temperature and possibly increased precipitation during the flowering period, would imply a significant increase in problems with *Fusarium* infection and occurrence of mycotoxins in cereals in the years to come.
VKM also identified important gaps in knowledge and data with respect to plant production, especially concerning *Fusarium* infection rates and protective measures against infection. Also, data on occurrence and data on toxicity of mycotoxins, particularly in some domestic animal species and for emerging toxins, are lacking.

References

Aamot HU, Hofgaard IS, Brodal G, Elen O, Klemsdal SS (2008). *Fusarium graminearum* in Norwegian cereals. J Plant Pathol 90 (3, Supplement):79.

Abbas HK, Mirocha CJ, Berdal BP, Sundheim L, Gunther R, Johnsen B (1987). Isolation and toxicity of *Fusarium* species from various areas of Norway. Acta Agric Scand Sect B Soil Plant Sci 37:427-435.

Abbas HK, Mirocha CJ, Gunther R (1989). Mycotoxins produced by toxic *Fusarium* isolates obtained from agricultural and nonagricultural areas (Arctic) of Norway. Mycopathologia 105:143-151.

Abbas HK, Mirocha CJ, Vesonder RF, Gunther R (1990). Acute toxic effects of an isoloate of moniliformin-producing *Fusarium oxysporum* and purified moniliformin on rats. Arch Environ Contam Toxicol 19:433-436.

Abd-Allah GA, El-Fayoumi RI, Smith, MJ, Heckmann RA, O'Neill KL (1999). A comparative evaluation of aflatoxin B-1 genotoxicity in fish models using the Comet assay. Mutat Res Genet Toxicol Environ Mutagen 446:181-188.

Abdelhamid AM, Kelada IP, Ali MM, el-Ayouty SA (1992). Influence of zearalenone on some metabolic, physiological and pathological aspects of female rabbits at two different agfes. Arch Tierernahrung 42:63-70.

Abdel-Wahhab MA, Hasan AM, Aly SE, Mahrous KF (2005). Adsorption of sterigmatocystin by montmorillonite and inhibition of its genotoxicity in the Nile tilapia fish (*Oreachromis nilaticus*). Mutat Res Genet Toxicol Environ Mutagen 582:20-27.

Abrahamsen U, Åssveen M, Uhlen A, Olberg E (2005). Dyrkings- og avlingspotensialet av rybs, raps og erter i Norge. Husdyrforsøksmøtet 2005:367-370

Adams CM, Bischoff K (2011). Mycotoxins – aflatoxins. In: Small animal toxicology. Eds: Osweiler et al. Blackwell publishing Ames, Iowa, USA.

Alabouvette C (1999). *Fusariu* wilt suppressive soils: an example of disease-suppressive soils. Australasian Plant Pathol 28:57-64.

Ali S, Rivera VV, Secor GA (2005). DISEASE NOTES - First Report of *Fusarium graminearum* Causing Dry Rot of Potato in North Dakota. Plant Dis 89:105.

Allen NK, Burmeister HR, Weaver GA, Mirocha CJ (1981a). Toxicity of dietary and intravenously administred moniliformin to broiler chickens. Poult Sci 60:1415-1417.

Allen NK, Mirocha CJ, Aakhus-Allen S, Bitgood JJ, Weaver G, Bates F (1981b). Effects of dietary zearalenone on reproduction of chickens. Poult Sci 60:1165-1174.

Altomare C, Logrieco A, Bottalico A, Mulé G, Moretti A, Evidente A (1995). Production of type A trichothecenes and enniatin B *Fusarium sambucinum* Fuckel sensu lato. Mycopathologia 129:177-181.

Ambrecht BH, Geleta JN, Shalkop WT, Durbin CG (1971). A subacute exposure of beagle dogs to aflatoxins. Toxicol Appl Pharmacol 19: 579-585.

Anadon A, Arzo MA, Bories G (2005). Opinion of the scientific panel on additives and products of substances used in animal feed on a request from the commission on the safety of the product "Biomin BBSH 797" for piglets, pigs for fattening and chicken for fattening. EFSA J 169:1-14.

Angsubhakorn S, Poomvises P, Romruen K, Newberne PM (1981). Aflatoxicosis in horses. J Am Vet Med Assoc 178(3):274-278.

Arafa AS, Bloomer RJ, Wilson HR, Simpson CF, Harms RH (1981). Susceptibility of various poultry species to dietary aflatoxin. Br Poult Sci 22(5):431-6.

Aravind KL, Patil VS, Devegowda G, Umakantha B, Ganpule SP (2003). Efficacy of esterified glucomannan to counteract mycotoxicosis in naturally contaminated feed on performance and serum

biochemical and hematological parameters in broilers. Poult Sci 82:571-576.

Arkoosh MR, Kaattari SL (1987). Effect of early aflatoxin-B1 exposure on in vivo and in vitro antibody-responses in rainbow-trout, *Salmo gairdneri*. J Fish Biol 31:19-22.

Armbrecht BH, Geleta JN, Shalkop WT, Durbin CG (1971). A subacute exposure of beagle dogs to aflatoxins. Toxicol Appl Pharmacol 19:579-585.

Arukwe A, Grotmol T, Haugen TB, Knudsen FR, Goksoyr A (1999). Fish model for assessing the in vivo estrogenic potency of the mycotoxin zearalenone and its metabolites. Sci. Total Environ. 236: 153-161.

Asao T, Buchi G, Abdel-Kader MM, Chang SB, Wick EL, Wogan GN (1965). The structures of aflatoxins B and G1. J Am Chem Soc 87:882-886.

Bai G, Shaner G (2004). Management and resistance in wheat and barley to *Fusarium* head blight. Annu RevPhytopathol 42:135–161.

Bailey GS, Williams DE, Hendricks JD, (1996). Fish models for environmental carcinogenesis: The rainbow trout. Environ Health Perspect 104:5-21.

Bakan B, Pinson L, Cahagnier B, Melcion D, Semon E, Richard-Molard D (2001). Toxigenic potential of *Fusarium culmorum* strains isolated from French wheat. Food Addit Contam 18:998-1003.

Baker DC, Rottinghouse GE (1999). Chronic experimental fumonisin intoxication of calves. J vet Diagn Invest 11(3):289-292.

Beaver RW, Wilson DM, James MA, Haydon KD, Colvin BM, Sangster LT, Pikul AH, Groopman JD (1990). Distribution of aflatoxins in tissues of growing pigs fed an aflatoxin-contaminated diet amended with a high affinity aluminosilicate sorbent. Vet Hum Toxicol 32: 16-18.

Becker BA, Pace L, Rottinghaus GE, Shelby R, Misfeldt M, Ross PF (1995). Effects of feeding fumonisin B1 in lactating sows and their suckling pigs. Am J Vet Res 56:1253–1258.

Behm C, Degen GH, Follmann W (2009). The *Fusarium* toxin enniatin B exerts no genotoxic activity, but pronounced cytotoxicity in vitro. Mol Nutr Food Res 53:423-430.

Benbrook CM (2006). Breaking the mold-impacts of organic and conventional farming systems on mycotoksins in food and livestock feed. The Organic Center 1-40.

Bergamini E, Catellani D, Dall'asta C, Galaverna G, Dossena A, Marchelli R, Suman M (2010). Fate of Fusarium mycotoxins in the cereal product supply chain: the deoxynivalenol (DON) case within industrial bread-making technology. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 27(5):677-687.

Bergsjø B, Herstad O, Nafstad I (1993). Effects of feeding deoxynivalenol-contaminated oats on reproduction performance in white leghorn hens. British Poult Sci 34: 147-159.

Bernhoft A, Behrens G, Ingebrigtsen K, Langseth W, Berndt S, Haugen TB, Grotmol T (2001). Placental transfer of the estrogenic mycotoxin zearalenone in rats. Reprod Toxicol 15: 545-550.

Bernhoft A, Clasen PE, Kristoffersen AB, Torp M (2010). Less *Fusarium* infestation and mycotoxin contamination in organic than in conventional cereals. Food Addit Contam Part A, 27: 842-852.

Bernhoft A, Keblys M, Langseth W, Åkesson CP, Oswald IP, Larsen HJS (2000). A study on immuntoxicity of HT-2 and T-2 toxins in minipigs. Abstract presented at X International IUPAC Symposium on Mycotoxins and Phycotoxins, Brasil, May 2000.

Bernhoft A, Keblys M, Morrison E, Larsen HJ, Flåøyen A (2004). Combined effects of selected *Penicillium* mycotoxins on in vitro proliferation of porcine lymphocytes. Mycopathologia. 158(4):441-450.

Bernhoft A, Torp M, Clasen P-E, Løes A-K, Kristoffersen AB (2012). Influence of agronomic and climatic factors on *Fusarium* infestation and mycotoxin contamination in Norway. Food Addit Contam 29:1129-1140.

Berntssen MHG, Olsvik PA, Torstensen BE, Julshamn K, Midtun T, Goksøyr A, Johansen J, Sigholt T, Joerum N, Jakobsen J-V, Lundebye A-K, Lock E-J (2010). Reducing persistent organic pollutants while maintaining long chain omega-3 fatty acid in farmed Atlantic salmon using of decontaminated fish oils for an entire production cycle. Chemosphere 81:242-252.

Bérubé M-E, Vanasse A, Rioux S, Bourget N, Dion Y, Tremblay G (2012). Effect of glyphosate on *Fusarium* head blight in wheat and barley under different soil tillages. Plant Dis 96:338-344.

Beyer M, Klix MB, Klink H, Verreet JA (2006). Quantifying the effects of previous crop, tillage and trazole fungicides on the deoxynivalenol content of wheat grain- a review. J Plant Dis Protect 113:241-246.

Biehl ML, Prelusky DB, Koritz GD, Hartin KE, Buck WB, Trenholm HL (1993). Biliary excretion and enterohepatic cycling of zearalenone in immature pigs. Toxicol Appl Pharmacol 121(1):152-159.

Binder EM, Binder J, Ellend N, Schaffer E, Krska R, Braun R (1998). Microbiological degradation of deoxynivalenol and 3-acetyl-deoxynivalenol. In: Mycotoxins and phycotoxins: developments in chemistry, toxicology and food safety. (eds. Miraglia M, van Egmond HP, Brera C, Gilbert J) Alaken, Fort Collins, pp. 279-285.

Binder J, Horvath EM, Schatzmayr G, Ellend N, Danner H, Krska R, Braun R (1997). Screening for deoxynivalenol-detoxifying anaerobic rumen microorganisms. Cer Res Commun 25:343-346.

Biomin GmbH (2009). Mycofix Plus 3.0 - Always ahead in mycotoxin risk management. Product brochure. Herzogenburg, Austria: Erber AG.

Biomin GmbH (2011). Mycofix product line - naturally ahead in mycotoxin risk management. www. biomin. net.

Biró K (2003). Adverse effects of deoxynivalenol and ochratoxin a in farm animals. Comparative in vivo and in vitro studies. Thesis University of Utrecht.

Biró K, Barna-Vetró I, Pécsi T, Szabó E, Winkler G, Fink-Gremmels J, Solti L (2003). Evaluation of spermatological parameters in ochratoxin A-challenged boars. Theriogenology 60:199-207.

Birzele B, Meier A, Hindorf H, Dehne HW (2002). Epidemiology of *Fusarium* and deoxynivalenol content in winter wheat in the Rhineland, Germany. Eur J Plant Pathol 108:667-673.

Birzele B, Prange A, Krämer J (2000). Deoxynivalenol and ochratoxin A in German wheat and changes of level in relation to storage parameters. Food Addit Contam 17(12):1027-1035.

Bischoff K, Garland T (2009). Aflatoxicosis in dogs. In: Kirks Current veterinary Therapy XIV (Eds: Bonagura JD and Twedt DC). Saunders Elsevier 11830 Westland Drive. St Louis. Missouri 63146.

Blandino M, Haidukowski M, Pascale M, Plizzari L, Scudellari D, Reyneri A (2012). Integrated strategies for the control of *Fusarium* head blight and deoxynivalenol contamination in winter wheat. Field Crops Res 133:139-149.

Blank R, Rolfs J-P, Südekum K-H, Frolich AA, Marquardt RR, Wolfram S (2003). Effects of chronic ingestion of ochratoxin A on blood levels and excretion of the mycotoxin in sheep. J Agric Food Chem 51:6899-6905.

Blount WP (1961). Turkey “X” disease. J Brit Turkeys Fed 9:52-61.

Bortell RL, Asquith RL, Edds GT, Simpson CF, Aller WW (1983). Acute experimentally induced aflatoxicosis in the weaning pony. Am J Vet Res 44:2110-2114.

Bosch U, Mirocha CJ, Abbas HK, Di Menna M (1989). Toxicity and toxin production by *Fusarium* isolates from New Zealand. Mycopathologia 108:73-79.

Bouaziz C, Bouslimi A, Kadri R, Zaied C, Bacha H, Abid-Essefi S (2012). The in vitro effects of zearalenone and T-2 toxins on Vero cells. Exp Toxicol Pathol. 2012 Mar 17. Available online 17 March 2012, http://dx.doi.org/10.1016/j.etp.2012.02.005.

Boudergue C, Burel C, Dragacci S (2009). Review of mycotoxin-detoxifying agents used as feed

additives: mode of action, efficacy and feed/food safety. Scientific Report submitted to EFSA - European Food Safety Authority 2009; 191 pp. (www.efsa.europa.eu/en/supporting/doc/22e.pdf)

Bouhet S, Oswald IP (2007). The intestine as a possible target for fumonisin toxicity. Mol Nutr Food Res 51:925-31.

Boys E (2011). HGCA grain storage guide for cereals and oilseeds. Third edition. The Home- Grown Cereals Authority, UK. 28pp. www.hgca.com/grainstorage.

Bretz M, Beyer M, Cramer B, Knecht A, Humpf HU (2006). Thermal degradation of the Fusarium mycotoxin deoxynivalenol. J Agric Food Chem 23(17):6445-6451.

Brodal G (1991). Såkornbeising etter behov. Nye analyserutiner ved Statens frøkontroll fra høsten 1990 [Fungicide treatment of cereal seed according to need. New routine analyses at the State Seed Testing Station from autumn 1990]. Statens fagtjeneste for landbruket, Faginfo no 2:205-211.

Brodal G, Henriksen B, Sundheim L (2009). Sjukdommer i korn, oljevekster og kjernebelgvekster. In: Brandsæter LO, Mangerud K, Birkenes SM, Brodal G, Andersen A. (red), Plantevern og plantehelse i økologisk landbruk, Bind 3: Korn, oljevekster og kjernebelgvekster. Bioforsk Fokus 4 (4), 107-150.

Broomhead JN, Ledoux DR, Bermudez AJ, Rottinghaus GE (2002). Chronic effects of moniliformin in broilers and turkeys fed dietary treatments to market age. Avian Dis 46:901-908.

Bruchim Y, Segev G, Sela U, Bdolah-Abram T, Salomon A, Aroch I (2011). Accidental fatal aflatoxicosis due to contaminated commercial diet in 50 dogs. Res Vet Sci 93(1):279-87.

Bryden WL (2012). Mycotoxin contamination of the feed supply chain: Implications for animal productivity and feed security. Animal Feed Science and Technology 173(1-2):134-158.

Bullerman L and Bianchini A (2007). Stability of mycotoxins during food processing. IntJ Food Microbiol 119(1–2):140–146.

Caloni F, Spotti M, Auerbach H, Op den Camp H, Gremmels JF, Pompa G (2000). In vitro metabolism of fumonisin B1 by ruminal microflora. Vet Res Commun 24:379-87.

Carlson DB, Williams DE, Spitsbergen JM, Ross PF, Bacon CW, Meredith FI, Riley RT (2001). Fumonisin B1 promotes aflatoxin B1 and N-methyl-N'-nitro-nitrosoguanidine-initiated liver tumors in rainbow trout. Toxicol Appl Pharmacol 172:29-36.

Castells M, Marín S, Sanchis V, Ramos AJ (2005). Fate of mycotoxins in cereals during extrusion cooking: a review. Food Addit Contam 22(2):150-157.

Castells M, Pardo E, Ramos AJ, Sanchis V, Marín S (2006). Reduction of ochratoxin A in extruded barley meal. J Food Prot 69(5):1139-43.

Cavret S, Laurent N, Videmann B, Mazallon M, Lecoeur S (2010). Assessment of deoxynivalenol (DON) adsorbents and characterisation of their efficacy using complementary in vitro tests. Food Addit Contam Part A 27:43-53.

Cavret S, Lecoeur S (2006). Fusariotoxin transfer in animal. Food Chem Toxicol 44(3):444-53.

Cawood ME, Gelderblom WCA, Alberts JF, Snyman SD (1994). Interaction of 14C-labelled fumonisin B1 mycotoxins with primary rat hepatocyte cultures. Food Chem Toxicol 32:627.

Celik I, Oguz H, Demet O, Donmez HH, Boydak M, Sur E (2000). Efficacy of polyvinylpolypyrrolidone in reducing the immunotoxicity of aflatoxin in growing broilers. Brit Poult Sci41:430-439.

Celius T, Haugen TB, Grotmol T, Walther BT (1999). A sensitive zonagenetic assay for rapid in vitro assessment of estrogenic potency of xenobiotics and mycotoxins. Environ Health Perspect 107:63-68.

Celius T, Matthews JB, Giesy JP, Zacharewski TR (2000). Quantification of rainbow trout (*Oncorhynchus mykiss*) zona radiata and vitellogenin mRNA levels using real-time PCR after in vivo treatment with estradiol-17 beta or alpha-zearalenol. J Steroid Biochem Mol Biol 75: 109-119.

Chakraborty S, Newton AC (2011). Climate change, plant diseases and food security: an overview. Plant Pathol 60:2–14

Chala A, Weinert J, Wolf GA (2003). An integrated approach to the evaluation of the efficacy of fungicides against *Fusarium culmorum*, the cause of head blight of wheat. J Phytopathol 151:673-678

Champeil A, Doré T, Fourbet JF (2004). *Fusarium* head blight: epidemiological origin of the effects of cultural practices on head blight attacks and the production of mycotoxins by *Fusarium* in wheat grains. Plant Sci 166:1389-1415.

Charmley E, Trenholm HL, Thomson BK, Vudathala D, Nicholson JWG, Prelusky DB, Charmley LL (1993). Influence oflevel of deoxynivalenol in the diet of diary cows in feed intake, milk production, and its composition. J Dairy Sci 76:3580-3587.

Chavez -Sanchez MC, Palacios CAM, Moreno IO (1994). Pathological effects of feeding young *Oreochromis-niloticus* diets supplemented with different levels of aflatoxin B-1. Aquaculture 127:49-60.

Chi MS, Mirocha CJ, Kurtz HJ, Weaver GA, Bates F, Robison T, Shimoda W (1980a). Effect of dietary zearalenone on growing broiler chicks. Poult Sci 59:531-536.

Chi MS, Mirocha GJ, Weaver GA, Kurtz HJ (1980b). Effect of zearalenone on female white leghorn chickens. Appl Environ Microbiol 39:1026-1030.

Chi MS, Robison TS, Mirocha CJ, Swanson SP and Shimoda W (1978). Excretion and tissue distribution of radioactivity from tritium-labeled T-2 toxin in chicks. Toxicology and Applied

Pharmacology 45: 391-402.

Choi CY, Nakajima-Adachi H, Kaminogawa S, Sugita-Konishi Y (2000). Nivalenol inhibits total and antigen-specific IgE production in mice. Toxicol Appl Pharmacol 165(1):94-8.

Choo TM, Vigier B, Shen QQ, Martin RA, Ho KM, Savard M (2004). Barley traits associated with resistance to *Fusarium* head blight and deoxynivalenol accumulation. Phytopathology 94:1145-1150.

Christensen CM, Kaufmann HH (1965). Deterioration of stored grain by fungi. Ann Rev Phytopathol 3:69-84.

Clasen P-E, Bremnes NB, Vidnes A, Bernhoft A (2002). *Fusarium*-mykotoksiner i korn – fordeling mellom skall og kjerne. I Kaurstad E. Husdyrforsøksmøtet 2002. NLH, side 169-172.

Coakley SM, Scherm H, Chakraborty S (1999). Climate Change and Plant Disease Management. Ann Rev Phytopathol 37:399-426.

Coenen M, Boyens B (2001). Capacity of zeolithe to depress the oestrogenic effects of zearalenone. Proceedings of the Society of nutrition and physiology 2001, 10: 177.

Coker RD (1979). Aflatoxin: past, present and future. Tropical Science 21: 143-162.

Colhoun J, Taylor GS, Tomlinson R (1968). *Fusarium* diseases of cereals II. Infection of seedlings in relation to environmental factors. Transact Brit Mycol Soc 51:397-407.

Commission of the European Union (1998). Commission Regulation (EC) No. 1525/98 of 16 July 1998 amending Regulation (EC) No. 194/97 of 31 January 1997 setting maximum levels for certain contaminants in foodstuffs. Off J Eur Commun L201:43–45.

Constable PD, Foreman JH, Waggoner AL, Smith GW, Eppley RM, Tumbleson ME, Haschek WM (2000). The mechanism of fumonisin mycotoxicosis in horses. Draft report on USDA-CSREES Grant#928-39453, pp.1-28.

Côté L-M, Dahlem AM, Yoshizawa T, Swanson SP, Buck WB (1986). Excretion of deoxynivalenol and its metabolite in milk, urine, and feces of lactating dairy cows. J Dairy Sci 69:2416-2423.

Coulombe RA, Bailey GS, Nixon JE (1984). Comparative activation of aflatoxin-B1 to mutagens by isolated hepatocytes from rainbow trout (*Salmo gairdneri*) and Coho salmon (*Oncorhynchus kisutch*).

Carcinogenesis 5:29-33.

Covarelli L, Beccari G, Steed A, Nicholson P (2012). Colonization of soft wheat following infection of the stem base by *Fusarium culmorum* and translocation of deoxynivalenol to the head. Plant Pathol 61(6):1121–1129. Article first published online: 15 FEB 2012. DOI: 10.1111/j.1365-3059.2012.02600.x

Culler MD, Miller-Garvin JE, Dill-Macky R (2007). Effect of extended irrigation and host resistance on deoxynivalenol accumulation in *Fusarium*-infected wheat. Plant Dis 91:1464-1472.

Cysewski SJ, Pier AC, Baetz AL, Cheville NF (1982). Experimental equine aflatoxicosis. Toxicol Appl Pharmacol 65(3):354-365.

Dänicke S, Brũssow KP, Goyarts T, Valenta H, Ueberschär KH, Tiemann U (2007). On the transfer of the *Fusarium* toxins deoxynivalenol (DON) and zearalenone (ZON) from the sow to the full-term piglet during the last third of gestation. Food Chem Toxicol 45: 1565-1574.

Dänicke S, Valenta H, Döll S, Ganter M, Flachowsky G (2004). On the effectiveness of a detoxifying agent in preventing fusario-toxicosis in fattening pigs. Anim Feed Sci Technol 114:141-157.

Dantzer WR, Hopper J, Mullin K, Hendrich S, Murphy PA (1999). Excretion of (14)C-fumonisin B(1), (14)C-hydrolyzed fumonisin B(1), and (14)C-fumonisin B(1)-fructose in rats. J Agric Food Chem 47:4291-4296.

Davis RA, Huggins DR, Cook JR, Paulitz TC (2009). Nitrogen and crop rotation effects on *Fusarium* crown rot in no-till spring wheat. Can J Plant Pathol 31: 456-467.

de Luna L, Bujold I, Carisse O, Paulitz TC (2002). Ascospore gradients of *Gibberella zeae* from overwintered inoculum in wheat fields. Can J Plant Pathol 24: 457-464

De Nijs M (1997). Public Health Aspects of *Fusarium* Mycotoxins in Food in The Netherlands: A Risk Assessment, Dissertation, Wageningen Agricultural University, Wageningen.

De Nijs M, van Egmond HP, Nauta M, Rombouts FM, Notermans SH (1998). Assessment of human exposure to fumonisin B1. J Food Protect 61:879-84.

De Wolf E, Lipps P, Miller D, Knight P, Molineros J, Francl L, Madden L (2004). Evaluation of prediction models for wheat *Fusarium* head blight in the US, 2004. In: S.M. Canty, T. Boring, J. Wardwell, R.W. Ward (Eds.), Proceedings of the Second International Symposium on *Fusarium* Head Blight; incorporating the Eighth European *Fusarium* Seminar, 11–15 December 2004, Orlando, FL, USA. East Lansing, MI, Michigan State University, vol. 2, pp. 439.

De Wolf ED, Madden LV, Lipps PE (2003). Risk assessment models for wheat *Fusarium* head blight epidemics based on within-season weather data. Phytopathology93:428–435.

Del Ponte EM, Fernandes JMC, Pavan W (2005). A risk infection simulation model for *Fusarium* head blight of wheat. Fitopatologia Brasileira 30:634-642.

Deng SX, Tian LX, Liu FJ, Jin SJ, Liang GY, Yang HJ, Du ZY, Liu YJ (2010). Toxic effects and residue of aflatoxin B1 in tilapia (*Oreochromis niloticus x O. aureus*) during long-term dietary exposure. Aquaculture 307: 233-240.

Denning DW, Quiepo SC, Altman DG, Makarananda K, Neal GE, Camallere EL, Morgan MR, Tupasi TE (1995). Aflatoxin and outcome from acute lower respiratory infection in children in The Philippines. Ann Trop Paediatr 15:209-216.

Desjardins AE, Proctor RH, Bai G, McCormick SP, Shaner G, Buechley G, Hohn TM (1996). Reduced virulence of trichothecene-nonproducing mutants of *Gibberella zeae* in wheat field tests. Mol PlantMicrobe Inter 9:775-781.

Detrixhe P, Chandelier A, Cavelier M, Buffet D, Oger R (2003). Development of an agro-meteorological model integrating leaf wetness duration estimation to assess the risk of head blight infection in wheat. Asp Appl Biol 68:199–204.

Diaz GJ (2002). Evaluation of the efficacy of a feed additive to ameliorate the toxic effects of 4,15-diacetoxiscirpenol in growing chicks. Poult Sci 81:1492-1495.

Diaz GJ, Cortes A, Roldan L (2005). Evaluation of the efficacy of four feed additives against the adverse effects of T-2 toxin in growing broiler chickens. J Appl Poult Res 14:226-231.

Diaz GJ, Squires EJ, Julian RJ, Boermans HJ (1994). Individual and combined effects of T-2 toxin and DAS in laying hens. Br Poult Sci 35(3):393-405.

Diaz-Llano G, Smith TK (2006). Effects of feeding grains naturally contaminated with *Fusarium* mycotoxins with and without a polymeric glucomannan mycotoxin adsorbent on reproductive performance and serum chemistry of pregnant gilts. J Anim Sci 84: 2361-2366.

Dilkin P, Direito G, Simas MM, Mallmann CA, Corrêa B (2010). Toxicokinetics and toxicological effects of single oral dose of fumonisin B1 containing *Fusarium* verticillioides culture material in weaned piglets. Chem Biol Interact 185:157-62.

Dill-Macky R (2008). Cultural control practices for *Fusarium* head blight: Problems and solutions. Cer Res Commun 36: 653-657.

Dill-Macky R (2010). *Fusarium* head blight (scab). In WW Bockus, RL Bowden, RM Hunger, WL Morrill, TD Murray, RW Smiley (Eds.). Compendium of Wheat Diseases and Pests (pp. 34–36). St. Paul, MN: APS Press.

Dill-Macky R, Jones RK (2000). The effect of previous crop residues and tillage on *Fusarium* head blight of wheat. Plant Dis 84: 71-76.

Directorate of fisheries (2009). Nøkkeltall fra norsk havbruksnæring. ISBN 82-91065-15-2.

Divon H, Razzaghian J, Udnes-Aamot H, Klemsdal S (2011). *Fusarium langsethiae*, investigation of alternative infection routes in oats. Eur J Plant Pathol 132(1): 147-161.

Dohnal V, Jezkova A, Jun D and Kuca K (2008). Metabolic pathways of T-2 toxin. Current Drug

Metabolism 9: 77-82.

Döll S, Baardsen G, Moller P, Koppe W, Stubhaug I, Danicke S (2010). Effects of increasing concentrations of the mycotoxin deoxylnivalenil, zearalenone or ochratoxin A in diets for Atlantic salmon (*Salmo salar*) on growth performance and health. International Symposium on Nutrition and Feeding in Fish ISFNF, May 31-June 4, Qingdao China.

Döll S, Dänicke S, Schnurrbusch U (2003). The effect of increasing concentrations of *Fusarium* toxins in the diets for piglets on histological parameters of the uterus. Mycotox Res 19:73-76.

Döll S, Dänicke S, Valenta H, Flachowsky G (2004). In vitro studies on the evaluation of mycotoxin detoxifying agents for their efficacy on deoxynivalenol and zearalenone. Arch Anim Nutr 58: 311-324.

Döll S, Gericke S, Dänicke S, Raila J, Ueberschar KH, Valenta H, Schnurrbusch U, Schweigert FJ, Flachowsky G (2005). The efficacy of a modified aluminosilicate as a detoxifying agent in *Fusarium* toxin contaminated maize containing diets for piglets. J Anim Physiol Anim Nutr 89:342-358.

Döll S, Valenta H, Danicke S, Flachowsky G (2002). *Fusarium* mycotoxins in conventionally and organically grown grain from Thuringia/Germany. Landbauforschung Volkenrode 52: 91-96

Dombrink-Kurtzman MA (2003). Fumonisin and beauvericin induce apoptosis in turkey peripheral blood lymphocytes. Mycopathologia 156:357-364.

Duthie JA, Hall R (1987). Transmission of *Fusarium graminearum* from seed to stems of winter wheat. Plant Pathol 36:33-37.

Dwiwedi P, Wangikar PB, Sinha N (2004). Teratogenic effects of ochratoxin A in rabbits. World Rabbit Sci 12:159-171.

Eaton DL, Groopman JD (1994). The toxicology of aflatoxins. Human health, veterinary and agricultural significance, Academic Press, San Diego, California.

Edwards SG (2004). Influence of agricultural practices on *Fusarium* infection of cereals and subsequent contamination of grain by trichotecene mycotoxins. Toxicol Lett 153:29-35.

Edwards SG (2009a). *Fusarium* mycotoxin content in UK organic and conventional barley. Food Addit Contam 26(8): 1185-1190.

Edwards SG (2009b). *Fusarium* mycotoxin content of UK organic and conventional oats. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 26(7):1063-1069.

Edwards SG (2009c). *Fusarium* mycotoxin content of UK organic and conventional wheat. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 26(4): 496-506.

Edwards SG, Barrier-Guillot B, Clasen PE, Hietaniemi V, Pettersson H (2009). Emerging issues of HT-2 and T-2 toxins in European cereal production. World Mycotox J 2:173-179.

Edwards SG, Dickin ET, MacDonald S, Buttler D, Hazel CM, Patel S, Scudamore KA (2011). Distribution of Fusarium mycotoxins in UK wheat mill fractions. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 28(12):1694-704.

EFSA (2004a). Opinion of the Scientific Panel on Contaminants in the Food Chain on a request from the Commission related to aflatoxin B1 as undesirable substance in animal feed (Request N° EFSA-Q-2003-035 (adopted on 3 February 2004). EFSA J 39: 1-27.

EFSA (2004b). Opinion of the Scientific Panel on Contaminants in the Food Chain on a request from the Commission related to zearalenone as undesirable substance in animal feed (Question N° EFSA-Q-2003-037 Adopted on 28 July 2004. EFSA J 89: 1-35.

EFSA (2004c). Scientific Opinion of CONTAM Panel: Opinion of the Scientific Panel on contaminants in the food chain [CONTAM] related to deoxynivalenol (DON) as undesirable substance in animal feed. Question number: EFSA-Q-2003-036. EFSA J 73: 1-42.

EFSA (2004d). Opinion of the scientific panel on contaminants in the food chain on a request from the commission related to ochratoxin as undesirable substance in animal feed. EFSA J 101: 1-36.

EFSA (2004e). Opinion of the Scientific Panel on Contaminants in the Food Chain on a request from the Commission related to aflatoxin B_1 as undesirable substance in animal feed. EFSA J 39: 1-27.

EFSA (2005). Opinion of the scientific panel on contaminants in the food chanin on a request from the Commission related to fumonisins as undesirable substances in animal feed. EFSA J 235:1-32.

EFSA (2006). Opinion of the scientific panel on contaminants in the food chain on a request from the commission related to ochratoxin A in food. EFSA J 365:1-56.

EFSA (2007). Opinion of the scientific panel on contaminants in the food chain [CONTAM] related to the potential increase of consumer health risk by a possible increase of the existing maximum levels for aflatoxins in almonds, hazelnuts and pistachios and derived products. EFSA J 446: 1 – 127. http://www.efsa.europa.eu/en/efsajournal/doc/446.pdf

EFSA (2009). Effects on public health of an increase of the levels for aflatoxin total from 4 µg/kg to 10 µg/kg for tree nuts other than almonds, hazelnuts and pistachios - Statement of the Panel on Contaminants in the Food Chain. http://www.efsa.europa.eu/en/efsajournal/doc/s1168.pdf

EFSA (2011a). "Scientific Opinion on the risks for animal and public health related to the presence of T-2 and HT-2 toxin in food and feed1." EFSA Journal 9(12):2481.

EFSA (2011b). Scientific Opinion on the risks for public health related to the presence of zearalenone in food. EFSA Journal 9(6):2197.

EFSA (2011c). Scientific Opinion on the risks for animal and public health related to the presence of Alternaria toxins in feed and food. EFSA Journal 9(10):2407

Eiblmeier P, Lepschy von Gleissenthall J (2007). Risk evaluation of deoxynivalenol levels in Bavarian wheat from survey data. J Plant Dis Protect 114:69-75.

Elen O, Hofgaard IS, Brodal G, Aamot HU, Jestoi M, Klemsdal SS (2008). Effect of fungicide treatment in field trials on different *Fusarium* spp and mycotoxins. Proceedings from 10th International *Fusarium* workshop and *Fusarium* genomics workshop 90(3): pp. 3. Date of conference: 30-8-2008.

Elen O, Langseth W, Liu W, Haug G, Skinnes H, Gullord M, Sundheim L (1997). The content of deoxynivalenol and occurrence of *Fusarium* spp. in cereals from field trials in Norway. Cer Res Commun 25: 585-586.

Elen ON, Liu W, Langseth W, Skinnes H, Gullord M, Sundheim L (2003). Deoxynivalenol content of cereal grain from naturally infected and artificially inoculated plants in field trials in Norway. Acta Agric Scand - Sect B - Soil Plant Sci 53: 183-189.

Ellis RW, Clements M, Tibbetts A, Winfree R (2000). Reduction of the bioavailability of 20 mu g/kg aflatoxin in trout feed containing clay. Aquaculture. 183:179-188.

El-Sayed YS, Khalil RH (2009). Toxicity, biochemical effects and residue of aflatoxin B-1 in marine water-reared sea bass (*Dicentrarchus labrax* L.). Food Chem Toxicol 47:1606-1609.

El-Sayed YS, Khalil RH, Saad TT (2009). Acute toxicity of ochratoxin-A in marine water-reared sea bass (*Dicentrarchus labrax* L.). Chemosphere 75:878-882.

Eriksen GS, Alexander J (Eds) (1998). *Fusarium* toxins in cereals - a risk assessment. TemaNord 1998: 502. Nordic Council of Ministers. Copenhagen.

Eriksen GS, Petterson H (2004). Toxicological evaluation of trichothecenes in animal feed. Anim Feed Sci Technol 114: 205-239.

Essigmann JM, Croy RG, Bennett RA, Wogan GN (1982). Metabolic activation of aflatoxin B1: Patterns of DNA adduct formation, removal, and excretion in relation to carcinogenesis. Drug Metabol Rev 13:581-602.

EU (2001). Commission Regulation (EC) No 466/2001 of 8 March 2001 setting maximum levels for certain contaminants in foodstuffs. http://www.caobisco.com/doc_uploads/legislation/466-2001EN.pdf

EU (2003). Directive 2002/32/EC of the European Parliament and of the Council of 7 May 2002 on undesirable substances in animal feed. http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CONSLEG:2002L0032:20061020:EN:PDF

Fæste CK, Ivanova L, Uhlig S (2011). In vitro metabolism and inhibition studies of the mycotoxin enniatin B in different species. Drug Metabol Disp (submitted).

Fernandes JM, Cunha GR, Del Ponte E, Pavan W, Pires JL, Baethgen W, Gimenez A, Magrin G, Travasso MI (2004). Modeling *Fusarium* head blight in wheat under climate change using linked process-based models. In: S.M. Canty, T. Boring, J. Wardwell, R.W. Ward (Eds.), Proceedings of the Second International Symposium on *Fusarium* Head Blight; incorporating the Eighth European Fusarium Seminar, 11–15 December 2004, Orlando, FL, USA. East Lansing, MI, Michigan State University 2:441–444.

Fernandez MR, Huber D, Basnyat P, Zentner RP (2008). Impact of agronomic practices on populations of *Fusarium* and other fungi in cereal and noncereal crop residues on the Canadian Prairies. Soil Tillage Res 100:60-71.

Fernandez MR, Selles F, Gehl D, DePauw RM, Zentner RP (2005). Crop production factors associated with *Fusarium* head blight in spring wheat in Eastern Saskatchewan. Crop Sci 45: 1908.

Fernandez MR, Zentner RP, Basnyat P, Gehl D, Selles F, Huber D (2009). Glyphosate associations with cereal diseases caused by *Fusarium* spp. in the Canadian Prairies. Eur J Agr 31:133-143.

Ficheaux AS, Sibiril Y, Parent-Massin D (2012). Co-exposure of *Fusarium* mycotoxins: In vitro myelotoxicity assessment on human hematopoietic progenitors. Toxicon 60(6):1171–1179.

Fincham JE, Marasas WF, Taljaard JJ, Kriek NP, Badenhorst CJ, Gelderblom WC, Seier JV, Smuts CM, Faber M, Weight MJ (1992). Atherogenic effects in a non-human primate of *Fusarium*

moniliforme cultures added to a carbohydrate diet, Atherosclerosis 94:13–25.

Fink-Gremmels J, Malekinejad H (2007). Clinical effects and biochemical mechanisms associated with exposure to the mycoestrogen zearalenone. Anim Feed Sci Technol 137:326-341.

Fináková S, Proska B, Šturdíková M (2007). Biosynthesis and biological activities of enniatins. Pharmazie 62:563-568.

Follstad MN, Christensen CM (1965). Effect of storage for three months at 14.1 % moisture content and 21 ^{o}C upon the microflora of barley kernels. Phytopathology 55:399-400.

Forsell JH, Witt MF, Tai JH, Jensen R, Pestka JJ (1986). Effects of 8-week exposure of the B6C3F1 mouse to dietary deoxynivalenol (vomitoxin) and zearalenone. Food Chem Toxicol. 1986:213-219.

Freimund S, Sauter M, Rys P (2003). Efficient adsorption of the mycotoxins zearalenone and T-2 toxin on a modified yeast glucan. J Environ SciHealth B38:243-255.

Friend DW, Thompson BK, Trenholm HL, Boermans HJ, Hartin KE, Panich PL (1992). Toxicity of T-2 toxin and its interaction with deoxynivalenol when fed to yound pigs. Can J Anim Sci 72:703-711.

Frøseth RB (2011). Økologisk håndbok, matvekster-KORN, Agropub. http://agropub.no/id/2380?hidemenu=true&kap=kap25 , 1-22. 2011. Ref Type: Electronic Citation.

Fuchs E, Binder EM, Heidler D, Krska R (2002). Structural characterization of metabolites after the microbial degradation of type A trichothecenes by the bacterial strain BBSH 797. Food Addit Contam 19:379-386.

Gajecka M, Przybylska-Gornowicz B, Bremski K, Polak M, Jakimiuk E, Skorska-Wyszynska E, Zielonka L, Gajecki M (2008). Ultrastructural changes of ovarian follicle and corpus luteum after experimental zearaleonone mycotoxicosis in bitch. Polish J Vet Sci 11(4):327-337.

Gallagher EP, Stapleton PL, Slone DH, Schlenk D, Eaton DL (1996). Channel catfish glutathione S-transferase isoenzyme activity toward (+/-)-anti-benzo[a]pyrene-trans-7,8-dihydrodiol-9,10-epoxide. Aquat Toxicol 34:135-150.

Garaleviciene D, Pettersson H, Elwinger K (2002). Effects on health and blood plasma parameters of laying hens by pure nivalenol in the diet. J Anim Physiol Anim Nutr 86(11-12):389-398.

Gäuman E, Naef-Roth ST, Ettlinger L (1950). Zur gewinnung von enniatinen aus dem myzel verschiedener Fusarien. Phytopath Zeitschrift 16(3):289-299.

Gautam P, Dill-Macky R (2012). Free water can leach mycotoxins from *Fusarium*-infected wheat heads. J Phytopathol 160:484–490.

Gelderblom WC, Jaskiewicz K, Marasas WF, Thiel PG, Horak RM, Vleggaar R, Kriek NP (1988). Fumonisins-novel mycotoxins with cancerpromoting activity produced by *Fusarium moniliforme*. Appl Environ Microbiol 54:1806-1811.

Gelineau-van Waes JB, Starr L, Maddox JR, Aleman F, Voss KA, Wilberding J, Riley RT (2005). Maternal fumonisin exposure and risk for neural tube defects: mechanisms in an in vivo mouse model. Birth Defects Res. Part A: Clin Mol Teratol 73:487–497.

Gilsinger J, Kong L, Shen X, Ohm HH (2005). DNA markers associated with low *Fusarium* head blight incidence and narrow flower opening in wheat. Theoret Appl Genet 110:1218-1225.

Glavits R, Sandor GS, Vanyi A, Gajdacs G (1983). Reproductive disorders caused by trichothecene mycotoxins in a large-scale pig herd. Acta Vet Hung 31(4):173-180.

Glynn NC, Hare MC, Parry DW, Edwards SG (2005). Phylogenetic analysis of EF-1 alpha gene sequences from isolates of *Microdochium nivale* leads to elevation of varieties *majus* and *nivale* to species status. Mycol Res 109: 872-880.

Golkari S, Gilbert J, Slusarenko K, Fernando WGD, Brûlé-Babel A (2008). Effect of rotation on colonization of field stubble by *Fusarium* species. Cer Res Commun 36: 555-561.

Gong YY, Cardwell K, Hounsa A, Egal S, Turner PC, Hall AJ, Wild CP (2002). Dietary aflatoxin exposure and impaired growth in young children from Benin and Togo: cross sectional study. Brit Med J325:20-21.

Gosman N, Chandler E, Thomsett M, Draeger R, Nicholson P (2005). Analysis of the relationship between parameters of resistance to *Fusarium* head blight and in vitro tolerance to deoxynivalenol of the winter wheat cultivar WEK0609. Eur J Plant Pathol 111:57-66.

Gottschalk C, Barthel J, Engelhardt G, Bauer J, Meyer K (2007). Occurrence of type A trichothecenes in conventionally and organically produced oats and oat products. Mol Nutr Food Res 51:1547-1553.

Gouze ME, Laffitte J, Dedieu G, Galinier A, Thouvenot JP, Oswald IP, Galtier P (2005). Individual and combined effects of low oral doses of deoxynivalenol and nivalenol in mice. Cellular and molecular biology 51: 809-817.

Government of Alberta (2002). Alberta *Fusarium graminearum* management plan. Developed by the provincial *Fusarium* action committee August 2002. Contact: Alberta Agriculture and Rural Development. http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/agdex5210#management.

Grenier B, Oswald IP (2011). Mycotoxin co-contamination of food and feed: meta-analysis of publications describing toxicological interactions. World Mycotox J 4:285-313.

Groopman JD, Donahue PR, Zhu JQ, Chen JS, Wogan GN (1985). Aflatoxin metabolism in humans: detection of metabolites and nucleic acid adducts in urine by affinity chromatography. Proc Natl Acad Sci USA 82:6492-6496.

Groopman JD, Wang JS, Scholl P (1996). Molecular biomarkers for aflatoxins: from adducts to gene mutations to human liver cancer. Can J Physiol Pharmacol74:203-209.

Guan S, He JW, Young JC, Zhu HH, Li XZ, Ji C, Zhou T (2009). Transformation of trichothecene mycotoxins by microorganisms from fish digesta. Aquaculture. 290:290-295.

Guengerich FP (2005). Principles of covalent binding of reactive metabolites and examples of activation of bis-electrophiles by conjugation. Arch Biochem Biophys 433:369-78.

Guerre P, Eeckhoutte C, Larrieu G, Burgat V, Galtier P (1996). Dose-related effect of aflatoxin B1 on liver drug metabolizing enzymes in rabbit. Toxicology 108(1-2):39-48.

Gundersen GI, Bye AS, Berge B, Hoem B, Knutsen SS (2009). Jordbruk og miljø - Tilstand og utvikling 37:1-102 Statistisk Sentralbyrå.

Guo XW, Fernando WGD, Bullock P, Sapirstein H (2010). Quantifying cropping practises in relation to inoculum levels of *Fusarium graminearum* on crop stubble. Plant Pathol 59:1107-1113.

Gupta S, Montllor C, Hwang Y-S (1995). Isolation of Novel Beauvericin Analogues from the Fungus *Beauveria bassiana*. J Nat Prod 58(5).733–738.

Gurung NK, Rankins DL, Shelby RA, Goel S (1998). Effects of fumonsin B1-contaminated feeds on weaning Angora goats. J Anim Sci 76:2863–2870.

Haave R (1985). Forekomst og patogenitet av *Fusarium*-arter på korn i Norge. Dr. scient. avhandling, Norges landbrukshøgskole 82 p.

Hagelberg S, Hult K, Fuchs R (1989). Toxicokinetics of ochratoxin-A in several species and its plasma-binding properties. J Appl Toxicol 9:91-96.

Halstensen AS, Nordby KC, Klemsdal SS, Elen O, Clasen C-E, Eduard W (2006). Toxigenic *Fusarium* spp. as determinants of trichothecene mycotoxins in settled grain dust. J Occup Environ Hygiene 3:651-659.

Hamill RL, Higgens CE, Boaz HE, Gorman M (1969). The structure of beauvericin, a new depsipeptide antibiotic toxic to *Artemia salina*. Tetrahedron Lett 49:4255-4258.

Hammer P, Bluthgen A, Walte HG (1996). Carry-over of fumonisin B1 into the milk of lactating

cows. Milchwissenschaft 51:691–695.

Han D, Xie S, Zhu X, Yang Y, Guo Z (2010). Growth and hepatopancreas performances of gibel carp fed diets containing low levels of aflatoxin B-1. Aquac Nutr 16:335-342.

Hanssen-Bauer I, Drange H, Førland EJ, Roald LA, Børsheim KY, Hisdal H, Lawrence D, Nesje A, Sandven S, Sorteberg A, Sundby S, Vasskog K, Ådlandsvik B (2009). Klima i Norge 2100. Bakgrunnsmateriale til NOU Klimatilplassing, Norsk klimasenter Oslo

Harvey RB, Edrington TS, Kubena LF, Rottinghaus GE, Turk JR, Genovese KJ, Nisbet DJ (2001). Toxicity of moniliformin from *Fusarium fujikuroi* culture materal to growing barrows. J Food Protect 64(11):1780-1784.

Harvey RB, Ellissalde MH, Kubena LF, Weaver EA, Corrier DE, Clement BA (1992). Immunotoxicity of ochratoxin-A to growing gilts. Am J Vet Res 53(10):1966-1970.

Harvey RB, Kubena LF, Corrier DE, Witzel DA, Phillips TD, Heidelbaugh ND (1986). Effects of deoxynivalenol in a wheat ration fed to growing lambs. Am J Vet Res 47:1630-1632.

Harvey RB, Kubena LF, Elissalde MH, Rottinghaus GE, Corrier DE (1994). Administration of ochratoxin A and T-2 toxin to growing swine. Am J Vet Res 55(12):1757-61.

Harvey RB, Kubena LF, Phillips TD, Huff WE, Corrier DE (1989). Prevention of aflatoxicosis by addition of hydrated sodium calcium aluminosilicate to the diets of growing barrows. Am J Vet Res 50:416-420.

Haschek WM, Gumprecht LA, Smith G, Tumbleson ME, Constable PD (2001). Fumonisin toxicosis in swine: an overview of procine pulmonary edema and current perspectives. Environ Health Perspect 109 (suppl 2):251-257.

Hassan AM, Kenawy AM, Abbas WT, Abdel-Wahhab MA (2010). Prevention of cytogenetic, histochemical and biochemical alterations in Oreochromis niloticus by dietary supplement of sorbent materials. Ecotox Environ Safe 731890-1895.

Hasso SA (2003). Non-fatal aflatoxicosis in Arabian horses in Iraq. Vet Rec 152(21):657-658.

Hazel CM, Patel S (2004). Influence of processing on trichothecene levels. Toxicol Lett 153(1):51–59.

He P, Young LG, Forsberg C (1992). Microbial transformation of deoxynivalenol (vomitoxin). App Environ Microbiol 58:3857-3863.

Hedman R, Pettersson H (1997). Transformation of nivalenol by gastrointestinal microbes. Arch Anim Nutr 50:321-329.

Hedman R, Pettersson H, Engström B, Elwinger K, Fossum O (1995). Effects of feeding nivalenol-contaminated diets to male broiler chickens. Poultry Science 74:620-625.

Hedman R, Pettersson H, Lindberg JE (1997). Absorption and metabolism ov nivalenol in pigs. Arch Animal Nutr 50:13-24.

Heier T, Jain SK, Kogel KH, Pons-Kuhnemann J (2005). Influence of N-fertilization and fungicide strategies on *Fusarium* head blight severity and mycotoxin content in winter wheat. J Phytopathol 153(9):551-557.

Henriksen B (1999). Factors affecting *Fusarium* infection and mycotoxin content in cereal grains. Norges landbrukshøgskole. Dr. Scient. Theses 1999:4.

Henriksen B, Elen ON (2005). Natural *Fusarium* grain infection level in wheat, barley and oat after early application of fungicides and herbicides. J Phytopathol 153:214-220.

Henry MH, Wyatt RD, Fletchert OJ (2000). The toxicity of purified fumonisin B1 in broiler chicks. Poult Sci 79(10):1378-1384.

Hestbjerg H, Felding G, Elmholt S (2002). *Fusarium culmorum* infection of barley seedlings: Correlation between aggressiveness and deoxynivalenol content. J Phytopathol150:308-316.

Hietaniemi V, Rämö S, Manninen P, Parikka P, Hankomäki J (2009). The effect of cleaning and de-hulling on the trichothecene content in oats and barley. Presentation at Nordforsk mould and mycotoxin seminar 14-14 April, 2009, Uppsala, Sweden.

Hilton AJ, Jenkinson P, Hollins TW, Parry DW (1999). Relationship between cultivar height and severity of *Fusarium* ear blight in wheat. Plant Pathol 48:202-208.

Hoerr F (2008). Mycotoxicosis. In: Diseases of poultry. (Ed: Saif YM). Blackwell publishing. Ames, Iowa, USA.

Hoerr FJ, Carlton WW, Yagen B (1981). The toxicity of T-2 toxin and diacetoxyscirpenol in combination for broiler chickens. Food Cosmet Toxicol 19:185-188.

Hofgaard IS (2012). Effect of cultivation practice on prevalence of *Fusarium* and mycotoxins (HT2/T2 and DON) in oats and spring wheat in Norway. Presentation at the European Commission *FUSARIUM* – toxin forum, Albert Borschette Conference Centre, Brussels, Belgium, Friday 3rd of February 2012

Hofgaard IS, Aamot HU, Klemsdal SS, Elen O, Jestoi M, Brodal G (2010a). Occurrence of *Fusarium* spp. and mycotoxins in Norwegian wheat and oats. In: Hofgaard IS and Fløystad E (Ed.) Proceedings from Bioforsk FOKUS. 5(7) 9. Bioforsk. Date of conference: 23.11.2010.

Hofgaard IS, Brodal G, Elen O, Aamot HU, Jestoi M, Klemsdal SS (2010b). Occurrence of *Fusarium* spp. and mycotoxins in Norwegian oats and spring wheat sampled in a six-year period from 2004-2009. In: Arseniuk E, Czembor E and Goral T (Ed.) Proceedings from 11th European *Fusarium* Seminar. (pp. 171-172). Date of conference: 20-9-2010.

Hofgaard IS, Seehusen T, Abrahamsen U, Razzaghian J, Le VH, Elen O, Riley H, Strand E, Brodal G (2012). Impact of agricultural practices on mycotoxin contamination of oats and spring wheat in Norway. Abstract and poster at the 7th Conference of The World Mycotoxin Forum® and the XIIIth IUPAC International Symposium on Mycotoxins and Phycotoxins, "WMFmeetsIUPAC", Rotterdam, the Netherlands, 5-9 November 2012.

Homdork S, Fehrmann H, Beck R (2000). Influence of different storage conditions on the mycotoxin production and quality of *Fusarium*-infected wheat grain. J Phytopathol 148:7-15.

Hooft JM, Elmor HI, Encarnacao P, Bureau DP (2011). Rainbow trout (*Oncorhynchus mykiss*) is extremely sensitive to the feed-borne *Fusarium* mycotoxin deoxynivalenol (DON). Aquaculture. 311:224-232.

Hoogenboom LAP, Bokhorst JG, Nordholt MD, Broex NJG, Mevius D, Meijs JAC and van der Roest J (2007). Contaminants and micro-organisms in organic food products. 1-4, RIKILT Institute of Food Safety; Wageningen, The Nethelands.

Hooker DC, Schaafsma AW (2003). The DONcast model: using weather variables pre- and post-heading to predict deoxynivalenol content in winter wheat. Asp Applied Biol 68:117–122.

Hooker DC, Schaafsma AW (2004). The DONcast model: predicting deoxynivalenol (DON) in wheat. In: S.M. Canty, T. Boring, J. Wardwell, R.W. Ward (Eds.), Proceedings of the Second International Symposium on *Fusarium* Head Blight; incorporating the Eighth European *Fusarium* Seminar, 11–15 December 2004, Orlando, FL, USA. East Lansing, MI, Michigan State University, 2, pp. 458

Hooker DC, Schaafsma AW, Tamburic-Ilincic L (2002). Using weather variables pre- and post-heading to predict deoxynivalenol content in winter wheat. Plant Dis 86: 611-619.

Horevaj PGL, Gale LR, Milus EA (2011). Resistance in Winter Wheat Lines to Initial Infection and Spread Within Spikes by Deoxynivalenol and Nivalenol Chemotypes of *Fusarium graminearum*. Plant Dis 95:31-37.

Howard PC, Eppley RM, Stack ME, Warbritton A, Voss KA, Lorentzen RJ, Kovach RM, Bucci TJ (2001). Fumonisin b1 carcinogenicity in a two-year feeding study using F344 rats and B6C3F1 mice. Environ Health Perspect. 109(Suppl 2):277–282.

Hsu IC, Smalley EB, Strong FM, Ribelin WE (1972). Identification of T-2 toxin in mouldy corn associated with a lethal toxicosis a dairy cattle. Appl Microbiol 24:684-687.

Huang Y, Wong PTW (1998). Effect of *Burkholderia (Pseudomonas) cepacia* and soil type on the control of crown rot in wheat. Plant Soil 203:103-108.

Hughes DM, Gahl MJ, Graham CH, Grieb SL (1999). Overt signs of toxicity to dogs and cats of dietary deoxynivalenol. J Anim Sci 77:693-700.

Humpf HU, Schmelz EM, Meredith FI, Vesper H, Vales TR, Wang E, Menaldino DS, Liotta DC, Merrill AH Jr. (1998). Acylation of naturally occurring and synthetic 1-deoxysphinganines by ceramide synthase. Formation of N-palmitoyl-aminopentol produces a toxic metabolite of hydrolyzed fumonisin, AP1, and a new category of ceramide synthase inhibitor. J Biol Chem 273:19060-19064.

Hussain Z, Khan MZ, Khan A, Javed I, Saleemi MK, Mahmood S, Asi MR (2010). Residues of aflatoxin B1 in broiler meat: effect of age and dietary aflatoxin B1 levels. Food Chem Toxicol 48:3304-7.

Huszenicza G, Fekete S, Szigeti G, Kulcsar M, Febel H, Kellems RO, Nagy P, Cseh S, Veresegyhazy T, Hullar I (2000). Ovarian consequences of low dose peroral *fusarium* (T-2) toxin in a ewe and heifer model. Theriogenology 53:1631-1639.

Huwig A, Freimund S, Käppeli O, Dutler H (2001). Mycotoxin detoxication og animal feed by different adsorbents. Toxicol lett 122: 179-188.

IARC (1993). Some naturally occurring substances: Food items and constituents, heterocyclic aromatic amines and mycotoxins. IARC Monogr Eval Carcinog Risks Hum 56:245–395.

Imathiu SM, Edwards SG, Ray RV and Back MA (2012). *Fusarium langsethiae* – a HT-2 and T-2 Toxins Producer that Needs More Attention. J Phytopathol. n/a. doi: 10.1111/jph.12036

Imathiu SM, Ray RV, Back M, Hare MC, Edwards SG (2009). *Fusarium langsethiae* pathogenicity and aggressiveness towards oats and wheat in wounded and unwounded in vitro detached leaf assays. Eur J Plant Pathol 124:117-126.

Inch SA, Gilbert J (2003). Survival of *Gibbrella zeae* in *Fusarium*-damaged wheat kernels. Plant Disease, 87, 282-287

Ingalls JR (1996). Influence if deoxynivalenol on feed consumption by dairy cows. Anim Feed Sci Technol 60:297-300.

Ismail Y, McCormick S, Hijri M (2011). A Fungal Symbiont of Plant-Roots Modulates Mycotoxin Gene Expression in the Pathogen *Fusarium sambucinum*. PLoS ONE 6(3): e17990. doi:10.1371/journal.pone.0017990

Ivanova L, Fæste CK, Uhlig S (2011). Phase I metabolism of enniatin B in liver microsomes. Anal Biochem Chem (accepted).

Ivanova L, Skjerve E, Eriksen GS, Uhlig S (2006). Cytotoxicity of enniatins A, A1, B, B1, B2 and B3 from *Fusarium avenaceum*. Toxicon 47:868-876.

Ivanova L, Uhlig S, Eriksen GS, Johannessen LE (2010). Enniatin B1 is a substrate of intestinal P-glycoprotein, multidrug resistance-associated protein 2 and breast cancer resistance protein. World Mycotox J 3:271-281.

Iverson F, Armstrong C, Nera E, Truelove J, Fernie S, Scott P, Stapley R, Hayward S, Gunner S (1995). Chronic feeding study of deoxynivalenol in B6C3F1 male and female mice. Teratog Carcinog Mutagen15:283–306.

Jantrarotai W, Lovell RT (1990). Subchronic toxicity of dietay aflatoxin B1 to channel catfish. J Aquat Anim Health 2:248-254.

Jard G, Liboz T, Mathieu F, Guyonvarc'h A, Lebrihi A (2011). Review of mycotoxin reduction in food and feed: from prevention in the field to detoxification by adsorption or transformation. Food

Addit Contam Part A 28:1590-1609.

Javed T, Bunte RM, Dombrink-Kurtzman MA, Richard JL, Bennett GA, Côté LM, Buck WB (2005). Comparative pathologic changes in broiler chicks on feed amended with *Fusarium proliferatum* culture material or purified fumonisin B1 and moniliformin. Mycopathol 159:553-564.

JECFA (1987). Evaluation of certain food additives and contaminants. Thirty-first report of the Joint FAO/WHO Expert Committee on Food Additives. WHO Technical Report Series, No. 759, 1987, and corrigendum. http://whqlibdoc.who.int/trs/WHO_TRS_759.pdf

JECFA (1997). Evaluation of certain food additives and contaminants (Forty-sixth report of the Joint FAO/WHO Expert Committee on Food Additives). WHO Technical Report Series, No. 868, 1997. http://whqlibdoc.who.int/trs/WHO_TRS_868.pdf

JECFA (1998). Forty-ninth report of the Joint FAO/WHO Expert Committee on Food Additives, WHO Technical Report Series. World Health Organisation, Geneva, Switzerland. http://www.inchem.org/documents/jecfa/jecmono/v040je16.htm

JECFA (2000). Zearalenone. In Safety evaluation of certain food additives and contaminants WHO Food additives series 44. Geneva.

JECFA (2001). Evaluation of certain mycotoxins in food. Fifty-sixth report of the Joint FAO/WHO Expert Committee on Food Additives, WHO Technical Report Series 906. World Health Organisation, Geneva, Switzerland. http://whqlibdoc.who.int/trs/WHO_TRS_906.pdf

JECFA (2007). Sixty-eighth report of the Joint FAO/WHO Expert Committee on Food Additives. http://whqlibdoc.who.int/publications/2007/9789241209472_eng.pdf

JECFA (2011). Evaluation of certain contaminants in food. Seventy-second report og the Joint FAO/WHO Expert Committee on Food Additives. Deoxynivalenol. WHO Technical Report Series page 37-48.

Jennings P, Coates ME, Walsh K, Turner JA, Nicholson P (2004). Determination of deoxynivalenol- and nivalenol-producing chemotypes of *Fusarium graminearum* isolated from wheat crops in England and Wales. Plant Pathol 53:643–652

Jestoi M (2008). Emerging *Fusarium* mycotoxins fusaprofilin, beauvericin, enniatins, and moniliformin – a review. Crit Rev Food Sci Nutr 48:21-49.

Jestoi M, Carmela SM, Merja K, PirjoV, Aldo R, Alberto R, Kimmo P (2004). Levels of mycotoxins and sample cytotoxicity of selected organic and conventional grain-based products purchased from Finnish and Italian markets. Molecul Nutr Food Res 48:299-307.

Jestoi M, Jonsson M, Koivisto P, Heinonen A, Isoniemi A, Kokkonen M, Peltonen K (2010). Acute oral toxicity of *Fusarium*-mycotoxin moniliformin. Toxicol Lett Abstract 196 S:333.

Jestoi M, Rokka M, Järvenpää E, Peltonen K (2009). Determination of *Fusarium* mycotoxins beauvericin and enniatins (A, A1, B, B1) in eggs of laying hens using liquid chromatography-tandem mass spectrometry (LC-MS/MS). Food Chem 115:1120-1127.

Jestoi M, Rokka M, Peltonen K (2007). Residues of emerging *Fusarium*–mycotoxins and coccidiostats in Finnish poultry tissues. Molec Nutr Food Res 51:625-637.

Johnson PJ, Casteel SW, Messer NT (1997). Effect of feeding deoxynivalenol (vomitoxin)-contaminated barley to horses. J Vet Diagn Invest 9:219-221.

Jones RK (1999). Seedling blight development and control in spring wheat damaged by *Fusarium graminearum* group 2. Plant Dis 83:1013-1018.

Jørstad I (1945). Parasittsoppene på kultur- og nyttevekster i Norge. I. Sekksporesopper (Ascomycetes) og konidiesopper (Fungi imperfecti). Meld. Statens Plantepatologiske Institutt. Nr.1. 142 p.

Jubert C, Mata J, Bench G, Dashwood R, Pereira C, Tracewell W, Turteltaub K, Williams D, Bailey G

(2009). Effects of chlorophyll and chlorophyllin on low-dose aflatoxin B(1) pharmacokinetics in human volunteers. Cancer Prev Res (Phila) 2:1015-22.

Juhasz J, Nagy P, Huszenicza G, Szigeti G, Reiczigel J, Kulcsar M (1997). Long term exposure to T-2 *Fusarium* mycotoxin fails to alter luteal function, follicular activity and embryo recovery in mares. Equine Vet J Suppl 25:17-21.

Juhasz J, Nagy P, Kulcsar M, Szigeti G, Reiczigel J, Huszenicza G (2001). Effect of low-dose zearalenone exposure on luteal function, follicular activity and uterine oedema in cycling mares. Acta Vet Hung 49:211-222.

Kabak B (2009). The fate of mycotoxins during thermal food processing. J Sci Food Agric 89:549-554.

Karakilcik AZ, Zerin M, Arslan O, Nazligul Y, Vural H (2004). Effects of vitamin C and E on liver enzymes and biochemical parameters of rabbits exposed to aflatoxin B1. Vet Hum Toxicol 46(4):190-192.

Karaman MK, Basmacioglu HB, Ortatatli MO, Oguz HO (2005). Evaluation of the detoxifying effect of yeast glucomannan on aflatoxicosis in broilers as assessed by gross examination and histopathology. Brit Poult Sci 46:394-400.

Kégl T, Ványi A (1991). T-2 fusariotoxicosis in a cattle stock. Magyar Allatorvosok Lapja. 46:467-471.

Kensler TW, Chen JG, Egner PA, Fahey JW, Jacobson LP, Stephenson KK, Ye L, Coady JL, Wang JB, Wu Y, Sun Y, Zhang QN, Zhang BC, Zhu YR, Qian GS, Carmella SG, Hecht SS, Benning L, Gange SJ, Groopman JD, Talalay P (2005). Effects of glucosinolate-rich broccoli sprouts on urinary levels of aflatoxin-DNA adducts and phenanthrene tetraols in a randomized clinical trial in He Zuo township, Qidong, People's Republic of China. Cancer Epidemiol Biomarkers Prev 14:2605-2613.

Khera KS, Whalen C, Angers G (1986). A teratology study on vomitoxin (4-deoxynivalenol) in rabbits. Food Chem Toxicol 24(5):421-424.

Khol-Parisini A, Hellweg P, Razzazi-Fazeli E, Saalmueller A, Strasser A, Tichy A, Zentek J (2012). Highly deoxynivalenol contaminated oats and immune function in horses. Arch Anim Nutr 66(2):149-161.

Khonga EB, Sutton JC (1988). Inoculum production and survival of *Gibberella zeae* in maize and wheat residues. Can J Plant Pathol 10:232-239.

Kiecana I, Mielniczuk E, Kaczmarek Z, Kostecki M, Golinski P (2002). Scab response and moniliformin accumulation in kernels of oat genotypes inoculated with *Fusarium avenaceum* in Poland. Eur J Plant Pathol 108:245-251.

Kiessling KH, Pettersson H, Sandholm K, Olsen M (1984). Metabolism of aflatoxin, ochratoxin, zearalenone, and three trichothecenes by intact rumen fluid, rumen protozoa, and rumen bacteria. Appl Environ Microbiol 47:1070.

Kirby GM, Wolf CR, Neal GE, Judah DJ, Henderson CJ, Srivatanakul P, Wild CP (1993). In vitro metabolism of aflatoxin B1 by normal and tumorous liver tissue from Thailand. Carcinogenesis14:2613-2620.

Kitchen BN, Carlton WW, Hinsman EJ (1977c). Ochratoxin A and citrinin induced nephrosis in beagle dogs. III. Terminal renalultrastructural alterations. Vet Pathol 14:392-406.

Kitchen BN, Carlton WW, Tuite J (1977a). Ochratoxin A and citrinin induced nephrosis in beagle dogs. I. Clinical and clinicopathological features. Vet Pathol 14:154-172.

Kitchen BN, Carlton WW, Tuite J (1977b). Ochratoxin A and citrinin induced nephrosis in beagle dogs. II. Pathology. Vet Pathol 14:261-272.

Knudsen IMB, Debosz K, Hockenhull J, Jensen DF, Elmholt S (1995). Biocontrol of seedling diseased of barley and wheat caused by *Fusarium culmorum* and *Bipolaris sorokiniana* – Effects of selected

fungal antagonists on growth and yield components. Plant Pathol 44(3):467-477.

Knudsen IMB, Elmholt S, Hockenhull J, Jensen DF (1995). Distribution of Saprophytic Fungi Antagonistic to *Fusarium-Culmorum* in 2 Differently Cultivated Field Soils, with Special Emphasis on the Genus *Fusarium*. Biol Agric Horticult 12:61-79.

Knutsen AK, Torp M, Holst-Jensen A (2004). Phylogenetic analyses of the *Fusarium poae*, *Fusarium poae* and *Fusarium langsethiae* species complex based on partial sequences of translation elongation factor-1 alpha gene. Int J Food Microbiol 95:287-295.

Koch HJ, Pringas C, Maerlaender B (2006). Evaluation of environmental and management effects on *Fusarium* head blight infection and deoxynivalenol concentration in the grain of winter wheat. Eur J Agr 24:357-366.

Köpke U, Thiel B, Elmholt S (2007). Strategies to reduce mycotoxin and fungal alkaloid contamination in organic and conventional cereal production systems. In: Cooper J, Niggli U, Leifert C (editors). Handbook of organic food safety and quality. Boca Raton (FL): CRC Press, p. 353-391.

Kosiak B, Torp M, Skjerve E, Andersen B (2004). *Alternaria* and *Fusarium* in Norwegian grain of reduced quality-a matched pair sample study. Int J Food Microbiol 93:51-62.

Kosiak B, Torp M, Skjerve E, Thrane U (2003). The prevalence and distribution of *Fusarium* species in Norwegian cereals: a survey. Acta Agric Scand Sect B Soil Plant Sci 53:168-176.

Kovacic S, Pepeljnjak S, Petrinec Z, Klaric MS (2009). Fumonisin B-1 neurotoxicity in young carp (*Cyprinus carpio* L.). Arh Hig Rada Toksikol 60:419-426.

Kremer RJ, Means NE (2009). Glyphosate and glyphosate-resistant crop interactions with rhizosphere microorganisms. Eur J Agr 31:153-161.

Krishnamachari KA, Bhat RV, Nagarajan V, Tilak TB (1975). Hepatitis due to aflatoxicosis. An outbreak in Western India. Lancet 1:1061-1063.

Kriss AB, Paul PA, Madden LV (2010). Relationship Between Yearly Fluctuations in *Fusarium* head blight. Intensity and environmental variables: A Window-Pane analysis. Phytopathology 100:784-797.

Kristiansen AL, Andersen LF, Lande B (2009). Småbarnskost 2 år. Landsomfattende kostholdsundersøkelse blant 2 år gamle barn. Helsedirektoratet, Oslo. Available in Norwegian at http://www.helsedirektoratet.no/publikasjoner/rapport-smabarnskost-2-aringer-2009/Publikasjoner/rapport-smabarnskost-2-aringer-2009.pdf

Krogh P (1991). Porcine nephropathy associated with ochratoxin A. In: Smith JE, Henderson RS. (eds): Mycotoxins and animal foods. CRC Press Inc., Boca Raton. Chapter 26, pp 627-645.

Krogh P, Axelsen NH, Elling F, Gyrd-Hansen N, Hald B, Hyldgaard-Jensen J, et al (1974). Experimental porcine nephropathy. Changes of renal function and structure induced by ochratoxin A-contaminated feed. Acta Pathol Microbiol Scand Sect A, Suppl 246:1-21.

Krogh P, Gyrd-Hansen N, Hald B, Larsen S, Nielsen JP, Smith M, Ivanoff C, Meisner H (1988). Renal enzyme activities in experimental ochratoxin A-induced porcine nephropathy: diagnostic potential of phosphoenolpyruvate carboxykinase and gamma-glutamyl transpeptidase activity. J Toxicol Environ Health 23(1):1-14.

Kroslák M (2002). Efficacy, and acceptability of fusafungine, a local treatment for both nose and throat infections, in adult patients with upper respiratory tract infections. Curr Med Res Opin 18:194-200.

Krska R, Welzig E, Boudra H (2007). Analysis of *Fusarium* toxins in feed. Anim Feed Sci Technol 137:241-264.

Kubena LF, Harvey RB, Buckley SA, Bailey RH, Rottinghous GE (1999). Effects of long-term feeding studies of diets containing moniliformin supplied by *Fusarium fujikuroi* culture material and fumonisin supplied by *Fusarium moniliforme* culture material to laying hens. Poult Sci 78:1499-1505.

Kubena LF, Harvey RB, Phillips TD, Corrier DE, Huff WE (1990). Diminution of aflatoxicosis in growing chickens by the dietary addition of a hydrated, sodium calcium aluminosilicate. Poult Sci 69:727-735.

Kubena LF, Huff WE, Harvey RB, Phillips TD, Rottinghaus GE (1989). Individual and combined toxicity of deoxynivalenol and T-2 toxin in broiler chicks. Poult Sci 68(5):622-6.

Kubena LF, Swanson SP, Harvey RB, Fletcher OJ, Rowe LD, Phillips TD (1985). Effects of feeding deoxynivalenol (vomitoxin)-contaminated wheat to growing chicks. Poult Sci 64: 1649-1655.

Kubosaki A, Aihara M, Park BJ, Sugiura Y, Shibutani M, Hirose M, Suzuki Y, Takatori K, Sugita-Konishi Y (2008). Immunotoxicity of nivalenol after subchronic dietary exposure to rats. Food Chem Toxicol 46(1):253-258.

Kuiper-Goodman T, Scott PM, McEwen NP, Lombaert GA, Ng W (1996). Approaches to the risk assessment of fumonisins in corn-based foods in Canada. Adv Exp Med Biol 392:369–393.

Kurek E, Jaroszuk-Scisel J. 2003. Rye (Secale cereale) growth promotion by Pseudomonas fluorescens strains and their interactions with *Fusarium culmorum* under various soil conditions. Biol Control 26:48-56.

LaBorde JB, Terry KT, Howard PC, Chen JJ, Collins TFX, Shackelford ME, Hansen DK (1997). Lack of embryotoxicity of fumonisin B1 in New Zealand white rabbits. Fundam Appl Toxicol 40:120-128.

Laca A, Mousia Z, Diaz M, Webb C, Pandiella SS (2006). Distribution of microbial contamination within cereal grains. J Food Engin 72:332-338.

Lacey J, Bateman GL, Mirocha CJ (1999). Effects of infection time and moisture on development of ear blight and deoxynivalenol production by *Fusarium* spp. in wheat. Ann Appl Biol 134:277-283.

Lairon D (2010). Nutritional quality and safety of organic food. A review. Agr Sustain Dev 30:33-41.

Lancova K, Hajslova J, Kostelanska M, Kohoutkova J, Nedelnik J, Moravcova H, Vanova M (2008). Fate of trichothecene mycotoxins during the processing: milling and baking. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 25(5):650-659.

Langseth W (2000). Kartlegging av mykotoksiner i norsk matkorn. 1990-1998.

1-1Veterinærinstituttet, Oslo, Norway.

Langseth W, Bernhoft A, Rundberget T, Kosiak B, Gareis M (1999). Mycotoxin production and cytotoxicity of *Fusarium* strains isolated from Norwegian cereals. Mycopathologia 144:103-113.

Langseth W, Elen ON (1996). Differences between barley, oats and wheat in the occurrence of deoxynivalenol and other trichothecenes in Norwegian grain. J Phytopathol 144:113-118.

Langseth W, Elen ON (1997). The occurence of deoxynivalenol in Norwegian cereals- differences between years and districts, 1988-1996. Acta Agric Scand Sect B Soil Plant Sci 47:176-184.

Langseth W, Rundberget T (1999). The occurrence of HT-2 toxin and other trichothecenes in Norwegian cereals. Mycopathologia 147:157-165.

Langseth W, Stenwig H, Sogn L, Mo E (1993). Growth of moulds and production of mycotoxins in wheat during drying and storage. Acta Agric Scand Sect B Soil Plant Sci 43:32-37.

Larsson P, Ngethe S, Ingebrigtsen K, Tjalve H (1992). Extrahepatic disposition of H-3 aflatoxin-B1 in the rainbow trout (*Oncorhynchus-mykiss*). Pharmacol Toxicol 71:262-271.

Lee DJ, Wales JH, Ayres JL, Sinnhuber RO (1968). Synergism between Cyclopropenoid Fatty Acids and Chemical Carcinogens in Rainbow Trout (*Salmo Gairdneri*). Cancer Res 28(11): 2312.

Lee MD, Calazzo JL, Staley AL, Lee JC, Warren MS, Fuernkranz H, Chamberland S, Lomocskaya O, Miller GH (2001). Microbial fermentation-derived inhibitors of efflux-pump-mediated drug resistance. Il Farmaco 56:81–85.

Leitgeb R, Lew H, Khidr R, Böhm J, Zollitsch W, Wagner E (2000). Influence of *Fusarium* toxins on

growth and carcass characteristics of turkeys. Bodenkultur 51(3):171-178.

Lemmens M, Buerstmayr H, Krska R, Schuhmacher R (2004a). The effect of inoculation treatment and long-term application of moisture on *Fusarium* head blight symptoms and deoxynivalenol contamination in wheat grains. Eur J Plant Pathol110:299-308.

Lemmens M, Haim K, Lew H, Ruckenbauer P (2004b). The effect of nitrogen fertilization on *Fusarium* head blight development and deoxynivalenol contamination in wheat. J Phytopathol 152:1-8.

Lemmens-Gruber R, Rachoy B, Steininger E, Kouri K, Saleh P, Krska R, Josephs R, Lemmens M (2000). The effect of the *Fusarium* metabolite beauvericin on electromechanical and physiological properties in isolated smooth and heart muscle preparations of guinea pigs. Mycopathologia 149:5-12.

Leslie JF, Summerell BA (2006). The *Fusarium* Laboratory Manual. Blackwall Publishing. 388 pp.

Leung MCK, Smith TK, Karrow NA, Boermans HJ (2007). Effects of foodborne *Fusarium* mycotoxins with and without a polymeric glucomannan mycotoxin adsorbent on food intake and nutrient digestibility, body weight and physical and clinicopathologic variables of mature dogs. Am J Vet Res 68(10):1122-1129.

Lewis L, Onsongo M, Njapau H, Schurz-Rogers H, Luber G, Kieszak S, Nyamongo J, Backer L, Dahive A, Misore A, DeCock K, Rubin C (2005). Aflatoxin contamination of commercial maize products during an outbreak of acute aflatoxicosis in eastern and central Kenya. Environ Health Perspect 113:1763–1767.

Li YC, Ledoux DR, Bermudez AJ, Fritsche KL, Rottinghous GE (2000). Effects of moniliformin on performance and immune function of broiler chicks. Poult Sci 79:26-32.

Lillemo M, Skinnes H, Bjørnstad Å, Buraas T, Reitan L, Bergersen S, Dieseth JA (2013). Valg av resistente sorter for å redusere omfanget av mykotoksiner i hvete, bygg og havre. Jord- og Plantekultur 2013, Bioforsk Fokus 8(1): 91-97.

Liu W, Sundheim L (1996). Nitrate nonutilizing mutants and vegetative compatibility groups in *Fusarium poae*. Fungal Genet Biol 20:12-17.

Liu W, Sundheim L, Langseth W (1998). Trichothecene production and the relationship to vegetative compatibility groups in *Fusarium poae*. Mycopathologia 140:105-114.

Liu WZ, Langseth W, Skinnes H, Elen ON, Sundheim L (1997). Comparison of visual head blight ratings, seed infection levels, and deoxynivalenol production for assessment of resistance in cereals inoculated with *Fusarium culmorum*. Eur J Plant Pathol 103:589-595.

Logrieco A, Moretti A, Ritieni A, Caiaffa MF, Macchia L (2002). Beauvericin: chemistry, biology and significance. In: Upadhyay RK (Ed.), Advances in microbiol toxin research and its biotechnological exploitation. Kluwer Academic, New York, pp 23-30.

Logrieco A, Moretti, Castella G, Kostecki M, Golinski P, Ritieni A, Chelkowski J (1998). Beauvericin production by *Fusarium* species. Appl Environ Microbiol 64:3084-3088.

Lohmann A (1988). Analytische, pharmakologische und mikrobiologische Untersuchungen des ionophoren Antibiotikums Fusafungin. PhD thesis, Westfälische Wilhelms Universität, Münster, Germany.

Lopes PRS, Pouey J, Enke DBS, Mallmann CA, Kich HA, Soquetta MB (2009). Use of adsorbent in diets containing aflatoxin for silver catfish fingerlings. Rev Bras Zootecn 38:589-595.

Lori GA, Sisterna MN, Sarandón SJ, Rizzo I, Chidichimo H (2009). *Fusarium* head blight in wheat: Impact of tillage and other agronomic practices under natural infection. Crop Protection 28(6): 495–502.

Lumlertdacha S, Lovell RT (1995). Fumonisin-contaminated dieatry corn reduced survival and antibody production by channel catfish challenged with Edwardsielle ictaluri. J Aquat Anim Health 7:1-8.

Lumlertdacha S, Lovell RT, Shelby RA, Lenz SD, Kemppainen BW (1995). Growth, hematology, and histopathology of channel catfish, *Ictalurus-punctatus,* fed toxins from *Fusarium-moniliforme*. Aquaculture 130:201-218.

Lutsky I, Mor N (1981). Experimental alimental toxic aleukia in cats. Lab Anim Sci 31:43-47.

Lutsky I, Mor N, Yagen B, Joffe AZ (1978). The role of T-2 toxin in experimental alimentary toxic aleukia: A toxicity study in cats. Toxicol Appl Pharmacol 43:111-124.

Madden LV, Lipps PE, De Wolf E (2004). Developing forecasting systems for *Fusarium* Head Blight. In: Canty SM, Boring T, Wardwell J, Ward RW (Eds.), Proceedings of the Second International Symposium on *Fusarium* Head Blight; incorporating the Eighth European *Fusarium* Seminar, 11–15 December 2004, Orlando, FL, USA. East Lansing, MI, Michigan State University, vol. 2, pp. 458.

Madden UA, Stahr HM (1995). Retention and distribution of aflatoxin in tissues of chicks fed aflatoxin-contaminated poultry rations amended with soil. Vet Hum Toxicol 37:24-29.

Madgwick JW, West J, White RP, Semenov MA, Townsend JA, Turner JA, Fitt B (2011). Impacts of climate change on wheat anthesis and *fusarium* ear blight in the UK. Eur J Plant Pathol 130(1):117-131.

Magan N, Hope R, Cairns V, Aldred D (2003). Post-harvest fungal ecology: Impact of fungal growth and mycotoxin accumulation in stored grain. Eur J Plant Pathol 109:723-730.

Magan N, Hope R, Colleate A, Baxter ES (2002). Relationship between growth and mycotoxin productiopn by *Fusarium* species, biocides and environment. EurJ Plant Pathol 108:685-690.

Magan N, Medina A, Aldred D (2011). Possible climate-change effects on mycotoxin contamination of food crops pre- and postharvest. Plant Pathol 60:150–163.

Mahnine N, Meca G, Elabidi A, Fekhaoui M, Saoiabi A, Font G, Mañes J, Zinedine A (2011). Further data on the levels of emerging *Fusarium* mycotoxins enniatins (A, A1, B, B1), beauvericin and fusaproliferin in breakfast and infant cereals from Morocco. Food Chem 124:481-485.

Maiorano A, Blandino M, Reyneri A, Vanara F (2008). Effects of maize residues on the *Fusarium* spp. infection and deoxynivalenol (DON) contamination of wheat grain. Crop Protect 27:182-188.

Malagutti L, Zannotti M, Scampini A, Sciaraffia F (2005). Effects of ochratoxin A on heavy pig production. Anim Res 54(3):179-184.

Maldonado-Ramirez SL, Schmale DG, Bergstrom GC (2005). The relative abundance of viable spores of *Gibberella zeae* in the planetary boundary layer suggests the role of long-distance transport in regional epidemics of *Fusarium* head blight. AgricForest Meteorol 132:20-27.

Manning BB, Li MH, Robinson EH (2005). Aflatoxins from moldy corn cause no reductions in channel catfish *Ictalurus punctatus* performance. J World Aquacult Soc 36:59-67.

Manning BB, Li MH, Robinson EH, Gaunt PS, Camus AC, Rottinghaus GE (2003a). Response of channel catfish to diets containing T-2 toxin. J Aquat Anim Health15:229-238.

Manning BB, Terhune JS, Li MH, Robinson EH, Wise DJ, Rottinghaus GE (2005c). Exposure to feedborne mycotoxins T-2 toxin or ochratoxin a causes increased mortality of channel catfish challenged with Edwardsiella ictaluri. J Aquat Anim Health 17:147-152.

Manning BB, Ulloa RM, Li MHH, Robinson EH, Rottinghaus GE (2003b). Ochratoxin A fed to channel catfish (*Ictalurus punctatus*) causes reduced growth and lesions of hepatopancreatic tissue. Aquaculture. 219:739-750.

Mao H (1985). Isolation and identification of beauvericin from Bai Jiang Yong. Zhongcaoyao 16:293-294.

Maragos CM, Richard JL (1994). Quantitation and stability of fumonisin B_1 and B_2 in milk. J AOAC Int 77:1162–1177.

Marasas WF (1996). Fumonisins: history, world-wide occurrence and impact. Adv Exp Med Biol

3921-17.

Marasas WF (2001). Discovery and occurrence of the fumonisins: a historical perspective. Environ Health Perspect 109(2):239-243.

Martin RA, MacLeod JA, Caldwell C (1991). Influences of production inputs on incidence of infection by *Fusarium* species on cereal seed. Plant Dis 75(8):784-788.

Martinez-de Anda A, Valdivia AG, Jaramillo-Juarez F, Reyes JL, Ortiz JL, Quezada T, de Luna MC, Rodriguez ML (2010). Effects of aflatoxins chronic intoxication in renal function of laying hens. Poult Sci 89:1622-1628.

Martinez-Larranaga M, Anadon A, Diaz MJ, Fernandez-Cruz ML, Martinez MA, Frejo MT, Martinez M, Fernandez R, Anton RM, Morales ME, Tafur M (1999). Toxicokinetic and oral bioavailability of fumonisin B1. Vet Hum Toxicol 41:357–362.

Matvaretabellen (1995). Den store matvaretabellen. Universitetsforlaget. ISBN 82-00-41607-0.

http://www.matportalen.no/verktoy/Matvaretabellen/article27210.ece/BINARY/Matvaretabellen+1995

Matvaretabellen (2006). http://www.matportalen.no/verktoy/Matvaretabellen/gamle_tabeller

Matvaretabellen (2012). http://www.matvaretabellen.no/

McKee TC, Bokesch HR, McCormick JL, Rashid MA, Spielvogel D, Gustafson KR, Alavanja MM, Cardelline JH, Boyd MR (1997). Isolation and characterization of new anti-HIV and cytotoxic leads from plants, marine, and microbial organisms. J Nat Prod 60:431-438.

McKenzie KS, Sarr AB, Mayura K, Bailey RH, Miller DR, Rogers TD, Norred WP, Voss KA, PlattnerRD, Kubena LF, Phillips TD (1997). Oxiative degradation and detoxification of mycotoxins using a novel source of ozone. Food Chem Toxicol 35:807-820.

McKenzie RA, Blaney BJ, Connole MD, Fitzpatrick LA (1981). Acute aflatoxicosis in calves fed peanut hay. Austr Vet J 57:284-286.

McMullen M, Bergstrom G, De Wolf E, Dill-Macky R, Hershman D, Shaner G, Van Sanford D (2012). A unified effort to fight an enemy of wheat and barley: *Fusarium* head blight. Plant Dis 96(12):1712-1728.

McMullen M, Halley S, Schatz B, Meyer S, Jordahl J, Ransom J (2008). Integrated strategies for *Fusarium* head blight management in the United States. Cer Res Commun 36:563-568.

Mei L, Zhang L, Dai R (2009). An inhibition study of beauvericin on human and rat cytochrome P450 enzymes and its pharmacokinetics in rats. J Enzyme Inhib Med Chem 24:753-62.

Meissonnier GM, Laffitte J, Raymond I, Benoit E, Cossalter A.M, Pinton P, Bertin G, Oswald IP, Galtier P (2008). Subclinical doses of T-2 toxin impair acquired immune response and liver cytochrome P450 in pigs. Toxicology 247:46-54.

Meredith FI, Riley RT, Bacon CW, Williams DE, Carlson DB (1998). Extraction, quantification, and biological availability of fumonisin B-1 incorporated into the Oregon test diet and fed to rainbow trout. J Food Protect 61:1034-1038.

Mesterházy A (1995). Types and components of resistance to *Fusarium* head blight of wheat. Plant Breeding, 114, 377-386

Mesterházy A (2003). Control of *Fusarium* head blight of wheat by fungicides. In: Leonard KJ and Bushnell WR (eds.) *Fusarium* head blight of wheat and barley. (pp 363-380) APS press, Minnesota, USA

Mesterházy A, Bartók T, Kászonyi G, Varga M, Tóth B, Varga J (2005). Common resistance to different *Fusarium* spp. causing *Fusarium* head blight in wheat. Eur J Plant Pathol112:267-281.

Mesterházy A, Bartók T, Mirocha CM, Komoróczy R (1999). Nature and resistance of wheat to

Fusarium head blight and deoxynivalenol contamination and their consequences for breeding. Plant Breeding 118. 97-110.

Mesterházy A, Kászonyi G, Tóth B, Bartok T, Varga M (2004). Prothioconazole fungicides against FHB in wheat, 2003/2004 results. pp. 355-358. Proceedings of the 2nd International Symposium on *Fusarium* Head Blight; incorporating the 8th European *Fusarium* Seminar, Orlando, FL, USA

Mézes, M and Balogh, K (2009). Mycotoxins in rabbit feed: a review. World rabbit science 17(2):53-62.

Miedaner T (1997). Breeding wheat and rye for resistance to *Fusarium* diseases. Plant Breeding, 116:201-220.

Miedaner T, Cumagun CJR, Chakraborty S (2008). Population genetics of three important head blight pathogens *Fusarium graminearum*, *F. pseudograminearum* and *F. culmorum*. J. Phytopathol 156:129–139.

Miller DM, Wilson DM (1994). Veterinary diseases related to aflatoxins. In: Eaton DL and Groopman JD (Eds). The Toxicology of Aflatoxins: Human Health, Veterinary and Agricultural Significance. Academic Press. NY, pp 347-364.

Miller JD, ApSimon JW, Blackwell BA, Greenhalgh R, Taylor A (2001). Deoxynivalenol: a 25 year perspective on a trichothecene of agricultural importance. In Fusarium: Paul E. Nelson memorial symposium, eds. B.a. Summerell, J.f: Leslie, D. Backhouse, W.L. Bryden and L.W. Burgess. St Paul, MN: APS Press pp. 310-320.

Miller JD, Culley J, Fraser K, Hubbard S, Meloche F, Quellet T, Seaman WL, Seifert KA, Turkington K, Voldeng H (1998). Effect of tillage practice on *Fusarium* head blight of wheat. Can J Plant Pathol 20:95-103.

Mirocha CJ, Pathre SV, Robison TS (1981). Comparative metabolism of zearalenone and transmission into bovine-milk. Food and Cosmetics Toxicology 19: 25-30.

Missmer SA, Suarez L, Felkner M, Wang E, Merrill Jr AE, Rothman KJ, Hendricks KA (2006). Exposure to fumonisins and the occurrence of neural tube defects along the Texas–Mexico border, Environ Health Perspect114:237–241.

Moeser S (2001). Einfluss von Zearalnon und Zeranol bei Kalbinnen. Dissertation, Veterinaermedizinsche Universität Wien.

Molnar O, Schatzmayr G, Fuchs E, Prillinger H (2004). Trichosporon mycotoxinivorans sp. nov., a new yeast species useful in biological detoxification of various mycotoxins. Syst Appl Microbiol 27:661-671.

Moretti A, Mulé G, Ritieni A, Logrieco A (2007). Further data on the production of beauvericin, enniatins and fusaprofilin and toxicity to Artemia salina by *Fusarium* species of *Gibberella fujikuroi* species complex. Int J Food Microbiol 118:158-163.

Morgan MK, Bursian SJ, Rottinghaus GH, Bennet GA, Render JA, Aulerich RJ (1998). Subacute and reproductive effects in mink from exposure to *Fusarium fujikuroi* culture material (M-1214) containing known concentrations on moniliformin. Arch Environ Contam Toxicol 35:513-517.

Morrison E, Rundberget T, Kosiak B, Aastveit AH, Bernhoft A (2001). Cytotoxicity of trichothecenes and fusarochromanone produced by *Fusarium equiseti* strains isolated from Norwegian grain. Mycopathologia 153: 49-56.

Moschini RC, Fortugno C (1996). Predicting wheat head blight incidence using models based on meteorological factors in Pergamino, Argentina. Eur J Plant Pathol 102:211–218.

Moschini RC, Pioli R, Carmona M, Sacchi O (2001). Empirical Predictions of Wheat Head Blight in the Northern Argentinean Pampas Region. Crop Sci 41:1541–1545.

Munger H, Vanasse A, Rioux S, Bourget N, Légère A (2010). Conservation tillage and low-input farming system: relation between yield, weed population and *Fusarium* head blight in spring wheat

(*Triticum aestivum* L.). Proceedings of Canadian Weed Science Society 64th Annual Meeting. November 2010. http://www.weedscience.ca/media/annual-meeting/2010_abstracts_regina.pdf

Naik DM, Busch LV, Barron GL (1978). Influence of temperature on the strain of *Fusarium graminearum* Schwabe in zearalenone production. Can J Plant Sci 58:1095-1097.

Nakajima T, Naito S (1995). Reassessment of mycotoxin productivity of *Microdochium nivale* in Japan. Ann Phytopathol Soc Jpn 61:357-361.

Nasri T, Bosch RR, Voorde S, Fink-Gremmels J (2006). Differential induction of apoptosis by type A and B trichothecenes in Jurkat T-lymphocytes. Toxicology In Vitro 20(6): 832-840.

Nelson M, Schneider NR, Doster AR, Carson MP, Klopfenstein T (1984). Vomitoxin-contaminated wheat – pathology, toxicity in cattle. Nebraska Beef Cattle report MP-47, pp. 3-12.

Neslon PE, Desjardins AE, Plattner RD (1993). Fumonisins, mycotoxins produced by *Fusarium* species: biology, chemistry, and significance. Ann Rev Phytopathol 31:233-252.

Newberne PM, Butler WH (1969). Acute and chronic effects of aflatoxin on the liver of domestic and laboratory animals: a review. Cancer Res 29:236-250.

Newman SJ, Smith JR, Stenske KA, Newman LB, Dunlap JR, Imerman PM, Kirk CA (2007). Aflatoxicosis in nine dogs after exposure to contaminated commercial dog food. J Vet Diagn Invest 19(2):168-75.

Ngethe S, Horsberg TE, Ingebrigtsen K (1992). The disposition of H-3 aflatoxin-B1 in the rainbow trout (*Oncorhynchus-mykiss*) after oral and intravenous administration. Aquaculture 108:323-332.

Ngethe S, Horsberg TE, Mitema E, Ingebrigtsen K (1993). Species-differences in hepatic concentration of orally-administered H-3 Afb(1) between rainbow trout (*Oncorhynchus-mykiss*) and tilapia (*Oreochromis niloticus*). Aquaculture. 114:355-358.

Noser J, Schmutz HR, Schmid S, Schneider P (2007). Bestimmung von Enniatinen in Getreideprodukten aus dem Schweizer Markt. Lebensmittelchemie 61:49-72.

Nowicki TW, Gaba WD, Dexter JE, Matsuo RR, Clear RM (1988). Retention of the Fusarium mycotoxin deoxynivalenol in wheat during processing and cooking of spaghetti and noodles. J Cereal Sci 8(2):189–202.

Obst A, Günther B, Beck R, Lepschy J, Tischner H (2002). Weather conditions conducive to *Gibberella zeae* and *Fusarium graminearum* head blight of wheat. J Appl Genet 43A:185-192.

Oerke E-C, Meier A, Dehne H-W, Sulyok M, Krska R, Steiner U (2010). Spatial variability of *Fusarium* head blight pathogens and associated mycotoxins in wheat crops. Plant Pathol 59:671-682.

Ojcious DMA, Zychlinsky A, Zheng LM, Young DE (1991). Ionophore-induced apoptosis: role of DNA fragmentation and calcium fluxes. Exp Cell Res 197:43-49.

Omura S, Koda H, Nishida H (1991). Hypolipemics containing beauvericin as acylcoenzyme A cholesterol acyltransferase inhibitor. Patent JP 89-16115019890623.

Onji Y, Dohi Y, Aoki, Y, Moriyama T, Nagami H, Uno M, Tanaka T, Yamazoe Y (1989). Deepoxynivalenol - a new metabolite of nivalenol found in the excreta of orally-administered rats. Journal of agricultural and food chemistry 37: 478-481.

Orlando B, Barrier-Guillot B, Gourdain E, Maumen C (2010). Identification of agronomic factors that influence the levels of T-2 and HT-2 toxins in barley grown in France. World Mycotox J 3: 169-174

Osweiler GD, Ensley SM (2012). Mycotoxins in grains and feed. In: Zimmerman JJ, Karriker LA, Ramirez A, Schwartz KJ, Stevenson GW (eds). Diseases of swine (10th edition). Wiley-Blackwell. Chichester, West-Sussex, UK.

Osweiler GD, Kehrli ME, Stabel JR, Thurston JR, Ross PF, Wilson TM (1993). Effects of fumonisin-contaminated corn screenings on growth and health of feeder calves. J Anim Sci 71:459-66.

Ottinger CA, Kaattari SL (2000). Long-term immune dysfunction in rainbow trout (*Oncorhynchus mykiss*) exposed as embryos to aflatoxin B-1. Fish Shellfish Immunol 10:101-106.

Ouyang YL, Azcona-Olivera JI, Pestka JJ (1995). Effects of trichothecene structure on cytokine secretion and gene expression in murine CD4+ T-cells. Toxicology 1041(3):187-202.

Øverby NC, Andersen LF (2002). Ungkost 2000. Landsomfattende kostholdsundersøkelse blant elever i 4.- og 8. klasse i Norge. Sosial- og helsedirektoratet, Oslo.

Available in Norwegian at: *http://www.helsedirektoratet.no/publikasjoner/ungkost-2000-landsomfattende-kostholdsundersokelse-blant-elever-i-4-og-8klasse-i-norge/Publikasjoner/ungkost-2000-landsomfattende-kostholdsundersokelse-blant-elever-i-4-og-8klasse-i-norge.pdf*

Øverby NC, Kristiansen AL, Andersen LF, Lande B (2009). Spedkost 12 måneder. Landsomfattende kostholdsundersøkelse blant 12 måneder gamle barn (Spedkost 2006 - 2007). Helsedirektoratet, Oslo. Available in Norwegian at *http://www.helsedirektoratet.no/publikasjoner/rapport-spedkost-12-maneder-2009/Publikasjoner/rapport-spedkost-12-maneder-2009.pdf*

Papendick II, Cook RJ (1974). Plant Water Stress and Development of *Fusarium* Foot Rot in Wheat Subjected to Different Cultural Practices. Phytopathology 64:358-363.

Pardo E, Marín S, Ramos AJ (2006). Ecophysiology of ochratoxigenic *Aspergillus ochraceus* and *Penicillium verrucosum* isolates. Predictive models for fungal spoilage prevention a review. Food Addit Contam 23:398–410.

Parlat SS, Özcan M, Oguz H (2001). Biological suppression of aflatoxicosis in Japanese quail (*Coturnix coturnix japonica*) by dietary addition of yeast (Saccharomyces cerevisiae). Res Vet Sci 71:207-211

Parry DW, Jenkinson P, McLeod L (1995) *Fusarium* ear blight (scab) in small grain cereals - a review. Plant Pathol 44:207-238.

Paterson RRM, Lima N (2010). How will climate change affect mycotoxins in food? Food Res Int 43:1902–1914.

Paul PA, El-Allaf SM, Lipps PE, Madden LV (2003). Development of *Fusarium* head blight of winter wheat in Ohio influenced by planting date, cultivar maturity, and inoculum level. In: Canty SM, Lewis J and Ward RW (Ed.) Proceedings from 2003 National *Fusarium* Head Blight Forum Proceedings. Kinko's, Okemos. Date of conference: 13-12-2003, Michigan State University, USA

Peeters H, Zocher R, Kleinkauf H (1988). Synthesis of beauvericin by a multifunctional enzyme. The Journal of Antibiotics XLI(3):352-359.

Peltonen K, Jestoi M, Eriksen GS (2010). Health effects of moniliformin a poorly understood *Fusarium* mycotoxin. World Mycotox J 3:403-414.

Pepeljnjak S, Petrinec Z, Kovacic S, Segvic M (2003). Screening toxicity study in young carp (*Cyprinus carpio* L.) on feed amended with fumonisin B-1. Mycopathologia 156:139-145.

Pereyra SA, Dill-Macky R (2008). Colonization of the residues of diverse plant species by *Gibberella zeae* and their contribution to *Fusarium* head blight inoculum. Plant Dis 92: 800-807.

Pereyra SA, Dill-Macky R, Sims AL (2004). Survival and inoculum production of *Gibberella zeae* in wheat residue. Plant Dis 88:724-730.

Pestka JJ (2010). Deoxynivalenol: mechanisms of action, human exposure, and toxicological relevance. Arch Toxicol 84(9):663-679.

Pestka JJ, Uzarski RL, Islam Z (2005). Induction of apoptosis and cytokine production in the mJurkat human T cells by deoxynivalenol: role of mitogen-activated protein kinases and comparison to other 8-ketotrichothecenes. Toxicology 206(2): 207-219.

Petersen A, Thorup I (2001). Preliminary evaluation of fumonisins by the Nordic countries and occurrence of fumonisins (FB_1 and FB_2) in corn-based food on the Danish market. Food Addit

Contam 18:221-226.

Pettersson H, Hedman R, Engstrom B, Elwinger K, Fossum O (1995). Nivalenol in Swedish cereals – occurrence, production and toxicity towards chickens. Food Addit Contam 12(3):373-376.

Pfohl-Leszkowicz A, Manderville RA (2007). Ochratoxin A: An overview on toxicity and carcinogenicity in animals and humans. Mol Nutr Food Res 51:61-99.

Phillips TD, Kubena LF, Harvey RB, Taylor DR, Heidelbaugh ND (1988). Hydrated sodium calcium aluminosilicate: a high affinity sorbent for aflatoxin. Poult Sci 67:243-247.

Pier AC, Cysewski SJ, Richard, JL, Baetz AL, Mithchell L (1976). Experimental mycotyoxicosis in calves with aflatoxin, ochratoxin, rubratoxin and T-2 toxin. In: Proceedings of the 80th Annual meeting of the US animal health association. Miami Beach, Florida. Richmond VA. US Animal Health Association pp 130-148.

Pinton P, Accensi F, Beauchamp E, Cossalter A-M, Callu P, Grosjean F, Oswald IP (2008). Ingestion of deoxynivalenol (DON) contaminated feed alters the pig vaccinal immune responses. Toxicol Lett 177:215-222.

Pirrung MC, Nauhaus SK, Singh B (1996). Cofactor-directed, time-dependent inhibition of thiamine enzymes by the fungal toxin moniliformin. J Org Chem 61(8):2592-2593.

Pitt JI, Hocking AD (2009). Fungi and Food Spoilage. Gaithersburg, Maryland, Aspen.

Plakas SM, Loveland PM, Bailey GS, Blazer VS, Wilson GL (1991). Tissue disposition and excretion of C-14-labeled aflatoxin B-1 after oral administration in Channel catfish. Food Chem Toxicol 29:805-808.

Plank B, Schuh M, Binder EM (2009). Investigations on the effect of two feed additives, Biomin® BBSH 797 and Mycofix Plus® 3.E as detoxificants of DON contaminated feed of piglets. (Untersuchungen über die detoxifizierende Wirkung der Futtermittelzusätze Biomin® BBSH 797 und Mycofix Plus® 3.E in Bezug auf DON im Ferkelaufzuchtfutter). Wiener Tierarzt Monatsschr 96:55-71.

Plattner RD, Nelson PE (1994). Production of beauvericin by a strain of *Fusarium proliferatium* isolated from corn fodder for swine. Appl Environ Microbiol 60(10):3894-3896.

Pleadin J, Persi N, Mitak M, Terzic S, Milic D, Vulic A, Brstilo M (2012). Biochemical changes in pig serum after ochratoxin A exposure. Bull Environ Contam Toxicol 88:1043-1047.

Plumlee K (2004). Clinical veterinary toxicology. Mosby. 118030 Westland industrial drive. St Luis, Missouri. USA.

Poapolathep A, Ohtsuka R, Kiatipattanasakul W, Ishigami N, Nakayama H, Doi K (2002). Nivalenol – induced apoptosis in thymus, spleen and Peyer's patches of mice. Exp Toxic Pathol 53: 441-446.

Poapolathep A, Sugita-Konishi Y, Doi K, Kumagai S (2003). The fates of trichothecene mycotoxins, nivalenol and fusarenon-X, in mice. Toxicon 41: 1047-1054.

Pocsfalvi G, Di Langa G, Ferranti P, Ritieni A, Randazzo G, Malorni A (1997). Observation of non-covalent interactions between beauvericin and oligonucleotides using electrospray ionization mass spectrometry. Rapid Commun Mass Spectrom11:265-272.

Pollestad ML, Øverby NC, Andersen LF (2002). Kosthold blant 4-åringer. Landsomfattende kostholdsundersøkelse. UNGKOST-2000. Sosial- og helsedirektoratet, Oslo. Available in Norwegian at *http://www.helsedirektoratet.no/publikasjoner/ungkost-2000-kosthold-blant-4-aringer/Publikasjoner/ungkost-2000-kosthold-blant-4-aringer.pdf*

Pollmann DS, Koch BA, Seitz LM, Mohr HE, Kennedy GA (1985). Deoxynivalenol-contaminated wheat in swine diets. J Anim Sci 60(1):239-247.

Poston HA, Coffin JL, Combs GF (1982). Biological effects of dietary T-2 toxin on rainbow-trout, *Salmo-gairdneri*. Aquat Toxicol 2:79-88.

Powell J, Swanton C (2008). A critique of studies evaluating glyphosate effects on diseases associated

with *Fusarium spp.* Weed Res 48:307-318.

Prabu PC, Dwivedi P, Sharma AK (2013). Toxicopathological studies on the effects of aflatoxin B(1), ochratoxin A and their interaction in New Zealand White rabbits. Exp Toxicol Pathol 65(3):277-86.

Prandini A, Sigolo S, Filippi L, Battilani P, Piva G (2009). Review of predictive models for *Fusarium* head blight and related mycotoxin contamination in wheat. Food Chem Toxicol 47:927–931.

Prelusky DB, Hamilton RM, Trenholm HL (1989). Transmission of residues to eggs following long-term administration of 14C-labelled deoxynivalenol to laying hens. Poult Sci 68(6):744-748.

Prelusky DB, Scott PM, Trenholm HL, Lawrence GA (1990). Minimal transmission of zearalenone to milk of dairy cows. Journal of Environmental Science and Health, Part B 25: 87-103.

Prelusky DB, Trenholm HL, Rotter BA, Miller JD, Savard ME, Yeung JM, Scott PM (1996). Biological fate of fumonisin B1 in food producing animals. Adv Exp Med Biol 392:265–278.

Prelusky DB, Trenholm HL, Savard ME (1994).Pharmacokinetic fate of ^{14}C-labelled fumonisin B_1 in swine. Nat Toxins 2:73-80.

Proctor RH, Hohn TM, McCormick SP (1997). Restoration of wild-type virulence to Tri5 disruption mutants of *Gibberella zeae* via gene reversion and mutant complementation. Microbiology 143: 2583-2591.

Qu B, Li HP, Zhang JB, Xu YB, Huang T, Wu AB, Zhao CS, Carter J, Nicholson P, Liao YC (2008). Geographic distribution and genetic diversity of *Fusarium graminearum* and *F. asiaticum* on wheat spikes throughout China. Plant Pathol 57:15-24.

Queslati S, Meca G, Mllliki A, Ghorbel A, Mañes J (2011). Determination of *Fusarium* mycotoxins enniatins, beauvericin and fusaproliferin in cereals and derived products from Tunisia. Food Control, doi: 10.1016/j.foodcont.2011.02.015.

Rafai P, Bata A, Vanyi A, Papp Z, Brydl E, Jakab L, Tuboly S, Tury E (1995a). Effect of various levels of T-2 toxin on the clinical status, performance and metabolism of growing pigs. Vet Rec 136:485-489.

Rafai P, Tuboly S, Bata A, Tilly P, Vanyi A, Papp Z, Jakab L, Tury E (1995b). Effect of various levels of T-2 toxin in the immune system of growing pigs. Vet Rec 136:511-514.

Raju MVLN, Devegowda G (2000) Influence of esterified-glucomannan on performance and organ morphology, serum biochemistry and haematology in broilers exposed to individual and combined mycotoxicosis (aflatoxin, ochratoxin and T-2 toxin). Brit Poult Sci 41:641-650.

Ramirez ML, Chulze S, Magan N (2004). Impact of environmental factors and fungicides on growth and deoxynivalenol production by *Fusarium graminearum* isolates from Argentinian wheat. Crop Protect 23;117-125.

Ramos AJ, Hernández E (1996). In situ absorption of aflatoxins in rat small intestine. Mycopathologia 134:27-30.

Rasmussen HB, Larsen K, Hald B, Moeller B, Elling F (1986). Outbreak of liver cell carcinoma among saltwater reared rainbow trout in Denmark. Dis Aquat Org 1:191-196.

Rasmussen PH (2010). Masked mycotoxions. Oral presentation at workshop, Nordic mycotoxin workshop SVA. Uppsala, Sweden April 2010.

Rauber RH, Dilkin P, Giacomini L, Zde Almeida CAA, Mallmann CA (2007). Performance of turkey poults fed different doses of aflatoxins in the diet. Poultry Science 86(8):1620-1624

Rawal S, Kim JE, Coulombe R Jr (2010). Aflatoxin B1 in poultry: toxicology, metabolism and prevention. Res Vet Sci 89(3):325-31

Raymond SL, Smith TK, Swamy HVLN (2003). Effects of feeding a blend of grains naturally contaminated with *Fusarium* mycotoxins on feed intake, serum chemistry, and haematology of horses,

and the efficacy of a polymeric glucomannan mycotoxin adsorbent. J Anim Sci 81: 2123-2130.

Raymond SL, Smith TK, Swamy HVLN (2005). Effects of feeding a blend of grains naturally contaminated with *Fusarium* mycotoxins on feed intake, metabolism, and indices of athletic performance of exercised horses. J Anim Sci 83:1267-1273.

Rheeder JP, Marasas WF, Vismer HF (2002). Production of fumonisin analogs by *Fusarium* species. Appl Environ Microbiol 68:2101-2105.

Ribelin WE, Fukushima K, Still PE (1978). The toxicity of ochratoxin to ruminants. Can J Comp Med 42:172.

Richard JL, Meerdink G, Maragos CM, Tumbleson M, Bordson G, Rice LG, Ross PF (1996). Absence of detectable fumonisins in the milk of cows fed *Fusarium proliferatum* (Masushima) Nirenberg culture material. Mycopathologia. 133:123-126.

Riley RT, An N-H, Showker JL, Yoo H-S, Norred WP, Chamberlain WJ, Wang E, Merrill AH Jr, Motelin G, Beasley VR, Haschek WM (1993). Alteration of tissue and serum sphinganine to sphingosine ratio: an early biomarker in pigs of exposure to fumonisin-containing feeds. Toxicol Appl Pharmacol 118:105–112.

Riley RT, Wang E, Schroeder JJ, Smith ER, Plattner RD, Abbas H, Yoo HS, Merrill AH Jr (1996). Evidence for disruption of sphingolipid metabolism as a contributing factor in the toxicity and carcinogenicity of fumonisins. Nat Toxins 4:3-15.

Rimestad AH, Løken EB, Nordbotten A (2000). The Norwegian food composition table and calculation system used at the Institute for Nutrition Research. Norw J Epidemiol 10:107-110.

Ritieni A, Moretti A, Logrieco A, Bottalico A, Randazzo G, Monti SM, Ferracane R, Fogliano V (1997). Occurrence of fusaproliferin, fumonisin B1 and beauvericin in maize from Italy. J Agric Food Chem45:4011–4016.

Roll-Hansen F (1940). Undersøkelser av *Gibberella saubinetii* (Mont.) Sacc. som fotsyke på havre. Meld. Statens frøkontroll 1939-40, 32-38.

Ross PF, Nelson PE, Owens DL, Rice LG, Nelson HA, Wilson TM (1994). Fumonisin B2 in cultured *Fusarium proliferatum*, M-6104, causes equine leukoencephalomalaciea. J Vet Diag Invest 6(2):263-265.

Rossi V, Giosuè S, Delogu G (2003a). A model estimating risk for *Fusarium* mycotoxins in wheat kernels. Asp Appl Biol 68:229–234.

Rossi V, Giosuè S, Pattori E, Spanna F, Del Vecchio A (2003). A model estimating the risk of *Fusarium* head blight on wheat. EPPO Bulletin 33: 421–425.

Rossi V, Scandolara A, Battilani P (2006). Effect of temperature, time and strains on *Fusarium verticillioides* sporulation. Book of abstracts “IX European Fusarium Seminar”, 4 19-22 September 2006, Wageningen, The Netherlands, pp. 149.

Rotter BA, Prelusky DB, Fortin A, Miller JD, Savard ME (1997). Impact of pure fumonisin B1 on various metabolic parameters and carcass qualityof growing-finishing swine – preliminary findings. Can J Anim Sci 77:465-470.

Rotter BA, Thompson BK, Prelusky DB, Trenholm HL, Stewart B, Miller JD, Savard ME (1996). Response of growing swine to dietary exposure to pure fumonisin B1 during and eight-week period: growth and clinical parameters. Nat Toxins 4(1):42-50.

Rudd JC, Horsley RD, McKendry AL, Elias EM (2001). Host plant resistance genes for *Fusarium* head blight: Sources, mechanisms, and utility in conventional breeding systems. Crop Sci 41(3):620-627.

Rustemeyer SM, Lamberson WR, Ledoux DR, Rottinghaus GE, Shaw DP, Cockrum RR, Kessler KL, Austin KJ, Cammack KM (2010). Effects of dietary aflatoxin on the health and performance of

growing barrows. J Anim Sci 88(11):3624-3630.

Rustom IYS (1997). Aflatoxin in food and feed: occurrence, legislation and inactivation by physical methods. Food Chem 59:57-67.

Salem MH, Kamel KI, Yousef MI, Hassan GA, EL-Nouty FD (2001). Protective role of ascorbic acid to enhance semen quality of rabbits treated with sublethal doses of aflatoxin B(1). Toxicology 162(3):209-18.

Salgado JD, Wallhead M, Madden LV, Paul PA (2011). Grain harvesting strategies to minimize grain quality losses due to *Fusarium* head blight in wheat. Plant Dis 95:1448-1457.

Samson RA, Frisvad JC (2004). *Penicillium* subgenus *Penicillium*: new taxonomic schemes and mycotoxins and other extrolites. Utrecht, The Netherlands, Centaalbureau voor Schimmelcultures.

Samson RA, Houbraken J, Thrane U, Frisvad JC, Andersen B (2010). Food and Indoor Fungi. CBS Laboratory Manual Series 2. Utrecht, The Netherlands: CBS-Knaw Fungal Biodiversity Centre. 390 pp.

Sandhu BS, Singh B, Brar RS (1998). Haematological and biochemical studies in broiler chicks feed ochratoxin and inoculated with inclusion body hepatitis virus, singly and in concurrence. Vet Res Commun 22:335-346.

Sandhu BS, Singh H, Singh B (1995). Pathological studies in broiler chicks fed aflatoxin or ochratoxin and inoculated with inclusion body hepatitis virus singly and in concurrence. Vet Res Commun 19:27-37.

Santacroce MP, Conversano MC, Casalino E, Lai O, Zizzadoro C, Centoducati G, Crescenzo G, (2008). Aflatoxins in aquatic species: metabolism, toxicity and perspectives. Rev Fish Biol Fish 18:99-130.

SCF (1999). Opinion of the Scientific Committee on Food on *Fusarium* toxins. Part 1: Deoxynivalenol (DON), expressed on 2 December.

SCF (2000a). Opinion of the Scientific Committee on Food on *Fusarium* toxins. Part 2: Zearalenone.

SCF (2000b). Opinion of the Scientific Committee on Food on *Fusarium* toxins. Part 3: Fumonisin B1 (FB1), expressed on 17 October

SCF (2000c). Opinion of the Scientific Committee on Food on *Fusarium* toxins. Part 4: Nivalenol.

SCF (2001). Opinion of the Scientific Committee on Food on *Fusarium* toxins. Part 5: T-2 Toxin and HT-2 Toxin.

Schaafsma AW, Hooker DC (2007). Climatic models to predict occurrence of *Fusarium* toxins in wheat and maize. Int J Food Microbiol 119:116–125.

Schaafsma AW, Tamburic-Ilincic L, Miller JD, Hooker DC (2001). Agronomic considerations for reducing deoxynivalenol in wheat grain. Can J Plant Pathol 23:279-285.

Schatzmayr G, Zehner F, Taubel M, Schatzmayr D, Klimitsch A, Loibner AP, Binder EM (2006). Microbiologicals for deactivating mycotoxins. Mol Nutr Food Res, 50 543-551.

Schjøth JE, Visconti A, Sundheim L (2009). Fumonisins in maize in relation to climate, planting time and hybrids in two agroecological zones in Zambia. Mycopathologia 167:209-219.

Schmidt A, Holst-Jensen A, Klemsdal S, Kullnig-Gradinger CM, Kubicek CP, Mach R, Niessen L, Nirenberg H, Thrane U, Torp M, Yli-Mattila T, Vogel RF (2004). An integrated taxonomic study of *Fusarium langsethiae* (ined.), *F. poae* and *F. sporotrichoides* based on the use of composite dataset. J Food Microbiol 95:341-349.

Schollenberger M, Drochner W, Müller H-M (2007). *Fusarium* toxins of the scirpentriol subgroup: a review. Mycopathologia 164:101-118.

Schollenberger M, Drochner W, Rufle M, Suchy S, Terry-Jara H, Muller HM (2005). Trichothecene

toxins in different groups of conventional and organic bread of the German market. J Food Comp Anal 18:69-78.

Scholtyssek S, Niemiec J, Bauer J (1987). Ochratoxin A in the layer´s feed. 1. Report: Influence on laying performance and egg quality (article in German). Arch Geflügelk 51:234-240.

Schroeder HW, Christensen JJ (1963). Factors affecting resistance of wheat to scab caused by *Gibberella zeae.* Phytopathology 53:831-838.

Schuh M (1996). Schimmelpilzgifte – Probleme beim Rind. Fortschritt Landwirt 21:SB7-SB8.

Schuhmacher-Wolz U, Heine K, Schneider K (2010). Report on toxicity data on trichothecene mycotoxins HT-2 and T-2 toxins. Scientific report submitted to EFSA. Pp 1-57.

Schwake-Anduschus C, Langenkämper G, Unbehend G, Dietrich R, Märtlbauer E, Münzing K (2010). Occurrence of Fusarium T-2 and HT-2 toxins in oats from cultivar studies in Germany and degradation of the toxins during grain cleaning treatment and food processing. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 27(9):1253-60.

Scudamore KA, Guy RC, Kelleher B, MacDonald SJ (2008). Fate of the fusarium mycotoxins, deoxynivalenol, nivalenol and zearalenone, during extrusion of wholemeal wheat grain. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 25(3):331-337.

Seehusen T, Hofgaard IS, Riley H, Elen O, Razzaghian J, Brodal G (2011). Soil tillage and straw treatment in relation to development of *Fusarium* spp. on crop debris and mycotoxins in grain. NJF seminar. 2011. Ref Type: Abstract.

Seeling K, Dänicke S (2005). Relevance of the *Fusarium* toxins deoxynivalenol and zearalenone in ruminant nutrition. J Anim Feed Sci 14:3-40.

Sharom FJ, Lu P, Liu R, Yu X (1998). Linear and cyclic peptides as substrates and modulators of P-glycoprotein: peptide binding and effects on drug transport and accumulation. Biochem J 333:621–630.

Shephard GS, Snijman PW (1999). Elimination and excretion of a single dose of the mycotoxin fumonisin B2 in a non-human primate. Food Chem Toxicol 37:111-116.

Shephard GS, Thiel PG, Sydenham EW, Alberts JF, Cawood ME (1994). Distribution and excretion of a single dose of the mycotoxin fumonisin B1 in a non-human primate. Toxicon 32:735-741.

Shin BS, Hong SH, Bulitta JB, Hwang SW, Kim HJ, Lee JB, Yang SD, Kim JE, Yoon HS, Kim do J, Yoo SD (2009). Disposition, oral bioavailability, and tissue distribution of zearalenone in rats at various dose levels. Journal of Toxicology and Environmental Health, Part A 72: 1406-1411.

Simpson DR, Weston GE, Turner JA, Jennings P, Nicholson P (2001). Differential control of head blight pathogens of wheat by fungicides and consequences for mycotoxin contamination of grain. Eur J Plant Pathol 107:421-431.

Sinovec ZJ, Nesic KV, Sefer DS (2006). Alleviating zearalenone effects on piglet performances by different adsorbents. Proceedings of the 19th IPVS Congress, Copenhagen, Denmark, 1, 297.

Sissener NH, Hemre G-I, Lall SP, Sagstad A, Petersen K, Williams J, Roholff J, Krogdahl A, Sanden M (2011). Are apparent negative effects of feeding GM MON810 maize to Atlantic salmon, *Salmo salar*, caused by confounding factors? Brit J Nutr 106: 42–56.

Skaug MA et al (1998). Ochratoxin A: a naturally occurring mycotoxin found in human milk samples from Norway. Acta Paediatr 87:1275.

Skaug MA, Helland I, Solvoll K, Saugstad OD (2001). Presence of ochratoxin A in human milk in relation to dietary intake. Food Addit Contam 18(4):321-327.

Smith EE, Phillips TD, Ellis JA, Harvey RB, Kubena LF, Thompson J, Newton G (1994). Dietary hydrated sodium calcium aluminosilicate reduction of aflatoxin M1 residue in dairy goat milk and effects on milk production and components. J Anim Sci 72:677-682.

Smith JF, di Menna ME, McGowan LT (1990). Reproductive performance of Coopworth ewes following oral doses of zearalenone before and after mating. J Reprod Fertil 89(1):99-106.

Smith KM (1960). Disease of turkey poults. Vet Rec 72:652.

Smith TK, McMillan EG, Castillo JB (1997). Effect of feeding blends of *Fusarium* mycotoxin-contaminated grains containing deoxynivalenol and fusaric acid on growth and feed consumption of immature swine. J Anim Sci 75(8):2184-91.

Smith-Spangler C, Brandeau ML, Hunter GE, Bavinger JC, Pearson M, Eschbach PJ, Sundaram V, Liu H, Schirmer P, Stave C, Olkin I, Bravata DM (2012). Are organic foods safer or healthier than conventional alternaitives? A systematic review. Ann Int Med 157:348-366.

Snijders CHA (1987). Interactions between winter wheat genotypes and isolates of *Fusarium culmorum*. Meded Faculteit Landbouwwet Rijksuniv. Gent. 52:807-814.

Soliman KM, El-Faramawy AA, Zakaria SM, Mekkawy SH (2001). Monitoring the preventive effect of hydrogen peroxide and γ-radiation of aflatoxicosis in growing rabbits and the effect of Ccooking on aflatoxin residues. J Agric Food Chem 49(7):3291–3295.

Spotti M, Pompa G, Caloni F (2001). Fumonisin B1 metabolism by bovine liver microsomes. Vet Res Commun 25:511-516.

Stenske KA, Smith JR, Newman SJ, Newman LB, Kirk CA (2006). Aflatoxicosis in dogs and dealing with suspected contaminated commercial foods. J Am Vet Med Assoc 228(11):1686-91.

Stenwig H, Liven E (1988a). Mycological examination of improperly stored grains. Acta Agric Scand 38:199-205.

Stenwig H, Liven E (1988b). Mycological survey of animal feeds used in Norway. Acta Agric Scand 38:189-197.

Stevens VL, Nimkar S, Jamison WC, Liotta DC, Merill AH Jr (1990). Characteristics of the growth inhibition and cytotoxicity of long-chain (sphingoid) bases for Chinese hamster ovary cells: Evidence for an involvement of proteinase C. Biochim Biophys Acta 1051(1):37-45.

Stoev SD, Goundasheva D, Mirtcheva T, Mantle PG (2000). Susceptibility to secondary bacterial infections in growing pigs as an early response in ochratoxicosis. Exp Toxicol Pathol 52:287-296.

Stoev SD, Gundasheva D, Zarkov I, Mircheva T, Zapryanova D, Denev S, Mitev Y, Daskalov H, Dutton M, Mwanza M, Schneider Y-J (2011). Experimental mycotoxin nephropathy in pigs provoked by a mouldy diet containing ochratoxin A and fumonisin B1. Exp Toxicol Pathol 64(7-8):733-41.

Stoev SD, Paskalev M, MacDonald S, Mantle PG (2002). Experimental 1 year ochratoxin A toxicosis in pigs Exp Toxicol Pathol 53:481-487.

Stoev SD, Vitanov S, Anguelov G, Petkova Bocharova T, Creppy EE (2001). Experimental porcine nephropathy in pigs provoked by a diet containing ochratoxin A and penicillic acid. Vet Res Commun 25:205-223.

Suga H, karugia GW, Ward T, Gale LR, Tomimura K, Nakajima T (2008). Molecular characterization of the *Fusarium graminearum* species complex in Japan. Phytopathology 98:159-166.

Sugita-Konishi Y, Kubosaki A, Takahashi M, Park BJ, Tanaka T, Takatori K, Hirose M, Shibutani M (2008). Nivalenol and the targeting of the female reproductive system as well as haematopoietic and immune systems in rats after 90-day exposure through the diet. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 25(9):1118-1127.

Sur E, Dönmez HH, Boydak M, Ataman MB (2012). Effects of glucomannan on the sacculus rotundus and peripheral blood lymphocytes in New Zealand rabbits during aflatoxicosis. ScientificWorld Journal 2012:632945. doi: 10.1100/2012/632945. Epub 2012 May 3.

Sypecka Z, Kelly M, Brereton P. 2004. Deoxynivalenol and zearalenone residues in eggs of laying hens fed with a naturally contaminated diet: effects on egg production and estimation of transmission

rates from feed to eggs. J Agric Food Chem 52(17):5463-5471.

Szabo-Fodor J, Kametler L, Posa R, Mamet R, Rajli V, Bauer J, Horn P, Kovacs F, Kovacs M (2008). Kinetics of fumonisin B1 in pigs and persistence in tissues after ingestion of a diet containing high fumonisin concentration. Ceram Res Commun 36:331–336.

Tacon AGJ, Metian M (2008). Global overview on the use of fish meal and fish oil in industrially compounded aquafeeds: Trends and future prospects. Aquaculture 285:146-158.

Takahashi M, Shibutani M, Sugita-Konishi Y, Aihara M, Inoue K, Woo GH, Fujimoto H, Hirose M (2008). A 90-day subchronic toxicity study of nivalenol, a trichothecene mycotoxin, in F344 rats. Food Chem Toxicol 46(1):125-135.

Tangni EK, Waegeneers N, Van Overmeire I, Goeyens L, Pussemier L (2009). Mycotoxin analyses in some home produced eggs in Belgium reveal small contribution to the total daily intake. Sci Total Environ 407(15):4411-8.

Tardieu D, Bailly J-D, Benlashehr I, Auby A, Jouglar J-Y, Guerre P (2009). Tissue persistence of fumonisin B1 in ducks and after exposure to a diet containing the maximum European tolerance for fumonisin in avian feeds. Chem-Biol Interact 182:239-244.

Tedjiotsop Feudijo F, Dornetshuber R, Lemmens M, Hoffmann O, Lemmens-Gruber R, Berger W (2010). Beauvericin and enniatin: emerging toxins and/or remedies? World Mycotox J 3:415-430.

Teich AH, Hamilton JR (1985). Effect of cultural practices, soil phosphorus, Potassium and pH on the incidence of *Fusarium* head blight and deoxynivalenol levels in wheat. Appl Environ Microbiol 49:1429-1431.

Teich AH, Nelson K (1984). Survey of *Fusarium* head blight and possible effects of cultural practices in wheat fields in Lambton County in 1983. Can Plant Dis Surv 64:11-13.

Tejada-Castaneda ZI, Ávila-Gonzalez E, Casaubon-Huguenin MT, Cervantes-Olivares RA, Vásquez-Peláez C, Hernández-Baumgarten EM, Moreno-Martínez E (2008). Biodetoxification of aflatoxin-contaminated chick feed. Poult Sci 87:1569-1576.

Tekle S, Dill-Macky R, Skinnes H, Tronsmo AM, Bjørnstad Å (2012). Infection process of *Fusarium graminearum* in oats (*Avenae sativa* L.) Eur J Plant Pathol 132:431-442.

Thrane U, Adler A, Clasen P-E, Galvano F, Langseth W, Lew H, Logrieco A, Nielsen KF, Ritieni A (2004). Diversity in metabolite production by *Fusarium langsethiae, Fusarium poae*, and *Fusarium sporotrichoides*. Int J Food Microbiol 95:257-266.

Thuvander A, Paulsen JE, Axberg K, Johansson N, Vidnes A, Enghardt-Barbieri H, Trygg K, Lund-Larsen K, Jahrl S, Widenfalk A, Bosnes V, Alexander J, Hult K, Olsen M (2001). Food Chem Toxicol 39(12):1145-1151.

Thuvander A, Wikman C, Gadhasson I (1999). In vitro exposure of human lymphocytes to trichothecenes: individual variation in sensitivity and effects of combined exposure on lymphocyte function. Food Chem Toxicol 37(6):639-48.

Tiemann U, Dänicke S (2007). In vivo and in vitro effects of the mycotoxins zearalenone and deoxynivalenol on different non-reproductive and reproductive organs in female pigs: A review. Food Addit Contam 24(3):306-314.

Tomoda H, Huang XH, Cao J, Nishida H, Nagao R, Okuda S, Tanaka H, Omura S, Arai H, Inoue K (1992). Inhibition of acyl-CoA: cholesterol acyltransferase activity by cyclodepsipeptide antibiotics. J Antibiot (Tokyo) 45:1626-1632.

Torp M, Nirenberg HI (2004). *Fusarium langsethiae* sp. nov. on cereals in Europe. Int J Food Microbiol 95(3):247-256.

Torstensen, BE, Espe M, Sanden M, Stubhaug I, Waagbo R, Hemre GI, Fontanillas R, Nordgarden U, Hevroy EM, Olsvik P, Berntssen MHG (2008). Novel production of Atlantic salmon (*Salmo salar*) protein based on combined replacement of fish meal and fish oil with plant meal and vegetable oil

blends. Aquaculture, 285:193-200.

Totland TH, Melnæs BK, Lundberg-Hallèn N, Helland-Kigen KM, Lund-Blix NA, Myhre JB, Johansen AMW, Løken EB & Andersen LF (2012). Norkost 3 - En landsomfattende kostholdsundersøkelse blant menn og kvinner i Norge i alderen 18-70 år, 2010-11. Helsedirektoratet. Oslo. Available in Norwegian at *http://helsedirektoratet.no/publikasjoner/norkost-3-en-landsomfattende-kostholdsundersokelse-blant-menn-og-kvinner-i-norge-i-alderen-18-70-ar/Publikasjoner/norkost-3-is-2000.pdf*

Trail F (2009). For blighted waves of grain: *Fusarium graminearum* in the postgenomic era. Plant Physiol 14:103-110.

Trenholm HL, Friend DW, Thompson BK, Prelusky DB (1988). Effects of zearalenone and deoxynivalenol combinations fed to pigs. Proc Jpn Assoc Mycotox (Suppl1): 101-102.

Truckness MW, Stoloff L, Young K, Wyatt RD, Miller BL (1983). Aflatoxicol and aflatoxins B1 and M1 in eggs and tissues of laying hens consuming aflatoxins-contaminated feed. Poult Sci 62:2176-2182.

Tuan NA, Grizzle JM, Lovell RT, Manning BB, Rottinghaus GE (2002). Growth and hepatic lesions of Nile tilapia (*Oreochromis niloticus*) fed diets containing aflatoxin B-1. Aquaculture 212:311-319.

Tuan NA, Manning BB, Lovell RT, Rottinghaus GE (2003). Responses of Nile tilapia (*Oreochromis niloticus*) fed diets containing different concentrations of moniliformin or fumonisin B-1. Aquaculture 217:515-528.

Turner PC, Hopton RP, White KL, Fisher J, Cade JE, Wild CP (2011). Assessment of deoxynivalenol metabolite profiles in UK adults. Food Chem Toxicol 49(1):132-135.

Turner PC, Rothwell JA, White KL, Gong Y, Cade JE, Wild CP (2008). Urinary deoxynivalenol is correlated with cereal intake in individuals from the United kingdom. Environ Health Perspect 116(1):21-25.

Turner PC, White KL, Burley VJ, Hopton RP, Rajendram A, Fisher J, Cade JE, Wild CP (2010). A comparison of deoxynivalenol intake and urinary deoxynivalenol in UK adults. Biomarkers 15(6):553-562.

Ueno Y, Iijima K, Wang SD, Sugiura Y, Sekijima M, Tanaka T, Chen C, Yu SZ (1997). Fumonisins as a possible contributory risk factor for primary liver cancer: a 3-year study of corn harvested in Haiman, China by HPLC and ELISA. Food Chem Toxicol 35:1143–1150.

Ueno Y, Yabe T, Hashimoto H, Sekijima M, Masuda T, Kim DJ, Hasegawa R, Ito N (1992). Enhancement of GST-P-positive liver cell foci development by nivalenol, a trichothecene mycotoxin. Carcinogenesis 13(5):787-91.

Uhlig S, Jestoi M, Parikka P (2007). *Fusarium avenaceum* – The North European situation. Int J Food Microbiol 119:17-24.

Ul-Hassan Z, Khan MZ, Khan A, Javed I (2012b). Immonological status of the progeny of breeder hens kept on ochratoxin A (OTA)- and aflatoxin B1 (AFB1)-contaminated feeds. J Immunotoxicol Early Online 1-11.

Ul-Hassan Z, Khan MZ, Saleemi MK, Khan A, Javed I, Noreen M (2012a). Immonological responses of male White Leghorn chicks kept on ochratoxin A (OTA)-contaminated feed. J Immunotoxicol 9(1):56-63.

Valenta H, Dänicke S (2005). Study on the transmission of deoxynivalenol and de-epoxy-deoxynivalenol into eggs of laying hens using a high-performance liquid chromatography-ultraviolet method with clean-up by immunoaffinity columns. Mol Nutr Food Res 49(8):779-785.

Vallone L, Dragoni I (1997). Investigation of mycotoxins (aflatoxin B1) occurrence in corn silage trench. Atti della Societa Italiana delle Scienze Veterinaire 51:237-238.

van Arendonk JJCM, Niemann GJ, Boon JJ, Lambers H. 1997. Effects of nitrogen supply on the

anatomy and chemical composition of leaves of four grass species belonging to the genus Poa, as determined by image-processing analysis and pyrolysis-mass spectrometry. Plant Cell Environ 20:881-897.

Van der Fels-Klerx HJ, Stratkou I (2010). T-2 toxin and HT-2 toxin in grain and grain-based commodities in Europe occurrence, factors affecting occurrence, co-occurrence and toxicological effects. World Mycotox J 3:349-367.

Van der Westhuizen L, Shephard GS, van Schalkwyk DJ (2001). The effect of a single gavage dose of fumonisin B1 on the sphinganine and sphingosine levels in vervet monkeys. Toxicon 39:273–281.

Van Egmond HP (1989). Aflatoxin M1: Occurrence, toxicity, regulation. In: van Egmond HP(Eds.). Mycotoxins in Dairy Products. Elsevier Applied Science, London and New York.

van Maanen A, Xu XM (2003). Modelling plant disease epidemics. Eur J Plant Pathol 109:669–682.

Vančo B, Šliková S, Šudyová V (2007). Influence of localities and winter wheat cultivars on deoxynivalenol accumulation and disease damage by *Fusarium culmorum*. Biologia 62:62-66.

Vanyi A, Glavits R, Molnar T (1995). Reproductive disorders due to F2 and T-2 toxins in large-scale pig farms. Magyar Allatorvosok Lapja 50(7):424-430.

Veldman A, Meijst JAC, Borggreve GJ, Heeres-van der Tol JJ (1992). Carry-over of aflatoxin from cow's food to milk. Anim Prod 55:163-168.

Vesonder R, Haliburton J, Stubblefield R, Gilmore W, Peterson S (1991). *Aspergillus flavus* and aflatoxins B1, B2, and M1 in corn associated with equine death. Arch Environ Contam Toxicol 20(1):151-3.

Vesonder R, Wu W, McAlpin C (1999). Beauvericin is not an acute toxin to ducklings bioassay. Abstract in Am Phytopatholl Soc Aug 12, 1999.

VKM (2008). Combined toxic effects of multiple chemical exposures. Norwegian Scientific Committee for Food Safety, Report 1:2008.

Voss KA, Riley RT, Bacon CW, Meredith FI, Norred WP (1998). Toxicity and sphinganine levels are correlated in rats fed fumonisin B1 (FB1) or hydrolyzed FB1. Environ Toxicol Pharmacol 5:101-104.

Voss KA, Smith GW, Haschek WM (2007). Fumonisins: Toxicokinetics, mechanism of action and toxicity. Anim Feed Sci Technol 137:299–325.

Voss KA, Snook ME (2010). Stability of the mycotoxin deoxynivalenol (DON) during the production of flour-based foods and wheat flake cereal. Food Addit Contam Part A Chem Anal Control Expo Risk Assess 27(12):1694-1700.

Vudathala DK, Prelusky DB, Ayroud M, Trenholm HL, Miller JD (1994). Pharmacokinetic fate and pathological effects of 14C-fumonisin B1 in laying hens. Nat Toxins 2:81–88.

Waalwijk C, Kastelein P, de Vries I, Kerényi Z, van der Lee T, Hesselink T, Köhl J; Kema G (2003). Major changes in *Fusarium* spp. In wheat in the Netherlands. Eur J Plant Pathol 109:743-754.

Walter S, Nicholson P, FM Doohan (2010). Action and reaction of host and pathogen during *Fusarium* head blight disease. New Phytologist 185:54–66.

Wang E, Ross PF, Wilson TM, Riley RT, Merrill Jr AH (1992). Increases in serum sphingosine and sphinganine and decreases in complex sphingolipids in ponies given feed containing fumonisins, mycotoxins produced by *Fusarium moniliforme*. J Nutr 122:1706–1716.

Ward TJ, Bielawski JP, Kistler HC, Sullivan E, O'Donnell K (2002). Ancestral polymorphism and adaptive evolution in the trichothecene mycotoxin gene cluster of phytopathogenic *Fusarium*. Proc Nat Acad Sci 99:9278-9283.

Ward TJ, Clear RM, Rooney AP, O Donnell K, Gaba D, Patrick S, Starkey DE, Gilbert J, Geiser DM, Nowicki TW (2008). An adaptive evolutionary shift in *Fusarium* head blight pathogen populations is driving the rapid spread of more toxigenic *Fusarium graminerarum* in North America. Fungal Genet

Biol 45(4):473-484.

Weaver GA, Kurtz HJ, Mirocha CJ, bates FY, Behrens JC, Robison TS (1978). Effect of T-2 toxin on porcine reproduction. Can Vet J 19(11):310-314.

Weaver GA, Kurtz HT, Behrens JC, Robison TS, Seguin BE, Bates FY, Mirocha JC (1986). Effect of zearalenone on the fertility of virgin dairy heifers. Am J Vet Res 47:1395-1397.

West JS, Holdgate S, Townsend JA, Edwards SG, Jennings P, Fitt BDL (2012). Impacts of changing climate and agronomic factors on *fusarium* ear blight of wheat in the UK. Fungal Ecol 5:53-61.

Wetscherek W, Huber H, Lew J (1998). Use of mycotoxin contaminated corn and of feed detoxifation additives in pig fattening (Einsatz von mit Mycotxinen kontaminiertem Mais und von Detoxificationsmitteln in der Schweinemast). Proceedings of the Society of Nutrition and Physiology 7:93.

Whitlow LW, Hagler JR (2009). Mycotoxins in Feeds. Feedstuffs, September 16:70-78.

Wogan GN (1966). Chemical nature and biological effects of the aflatoxins. Bacteriol Rev 30:460-470.

Wolzak A, Pearson AM, Coleman TH, Pestka JJ, Gray JI (1985). Aflatoxin deposition and clearance in the eggs of laying hens. Food Chem Toxicol 23:1057-1061.

Wong ZA, Hsieh DP (1980). The comparative metabolism and toxicokinetics of aflatoxin B1 in the monkey, rat, and mouse. Toxicol Appl Pharmacol 55:115-125.

Wozny M, Brzuzan P, Luczynski MK, Gora M, Bidzinska J, Jurkiewicz P (2008). Effects of cyclopenta[c]phenanthrene and its derivatives on zona radiata protein, ER alpha, and CYP1A mRNA expression in liver of rainbow trout (*Oncorhynchus mykiss* Walbaurn). Chem-Biol Interact 174:60-68.

Wu Q, Dohnal V, Huang L, Kuca K, Yuan Z (2010). Metabolic pathways of trichothecenes. Drug Metab Rev 42(2):250-267.

Wu Q, Lohrey L, Cramer B, Yuan Z, Humpf HU (2011). Impact of physicochemical parameters on the decomposition of deoxynivalenol during extrusion cooking of wheat grits. J Agric Food Chem 59(23):12480-12485.

Wu W, Liu T, Vesonder RF (1995). Comparative cytotoxicity of fumonisin B1 and moniliformin in chicken primary cell cultures. Mycopathologia 132(2):111-116.

Wunder W, Korn H (1982). Aflatoxin cancer (heptoma) in the liver of the rainbow trout. Zool Beitr 28:99-109.

Xiao H, Marquardt RR, Frohlich AA, Philipps GD, Vitti TG (1991). Effect of a hay and a grain diet on the bioavailability of ochratoxin A in the rumen of sheep. J Anim Sci 69:3715-3723.

Xu XM (2003). Effects of environmental conditions on the development of *Fusarium* ear blight. Eur J Plant Pathol 109:683–689.

Xu XM, Nicholson P (2009). Community ecology of fungal pathogens causing wheat head blight. Ann Rev Phytopathol 47:83–103.

Xu XM, Nicholson P, Thomsett MA, Simpson D, Cooke BM, Doohan FM, Brennan J, Monaghan S, Moretti A, Mule G, Hornok L, Beki E, Tatnell J, Ritieni A, Edwards SG (2008). Relationship between the fungal complex causing *Fusarium* head blight of wheat and environmental conditions. Phytopathology 98:69-78.

Yan W, Li HB, Cai SB, Ma HX, Rebetzke GJ, Liu CJ (2011). Effects of plant height on type I and type II resistance to *Fusarium* head blight in wheat. Plant Pathol 60:506-512.

Yi C, Kaul HP, Kubler E, Schwadorf K, Aufhammer W (2001). Head blight (*Fusarium graminearum*) and deoxynivalenol concentration in winter wheat as affected by pre-crop, soil tillage and nitrogen fertilization. J Plant Dis Prot 108(3):217-230.

Yildirim M, Manning BB, Lovell RT, Grizzle JM, Rottinghaus GE (2000). Toxicity of moniliformin and fumonisin B-1 fed singly and in combination in diets for young channel catfish *Ictalurus punctatus*. J World Aquacult Soc 31:599-608.

Yli-Mattila T (2010). Ecology and evolution of toxigenic *Fusarium* species in cereals in Northern Europe and Asia. J Plant Pathol 92:7-18.

Yli-Mattila T, Gavrilova O, O'Donnell K, Ward T, Procto R, Gagkava T (2009). Asian *Fusarium langsethiae* and two subgroups of European *F. langsethiae*. Nordforsk Mould and Mycotoxin seminar 2009.

Yli-Mattila T, Mach RL, Alekhina IA, Bulat SA, Koskinen S, Kullnig-Gradinger CM, Kubicek CP, Klemsdal SS (2004). Phylogenetic relationship of *Fusarium langsethiae* to *Fusarium poae* and *Fusarium sporotrichoides* as inferred by IGS, ITS, β-tubulin sequences and UP-PCR hybridization analysis. Int J Food Microbiol 95:267-285.

Young JC (1986). Reduction in levels of deoxynivalenol on contaminated corn by chemical and physical treatment. J Agric Food Chem 34:465-467.

Young JC, Zhou T, Yu H, Zhu H, Gong J (2007). Degradation of trichothecene mycotoxins by chicken intestinal microbes. Food Chem Toxicol 45:136-143.

Young LG, McGirr L, Valli VE, Lumsden JH, Lun A (1983). Vomitoxin in corn fed to young pigs. J Anim Sci 57(3):655-664.

Yunus AW, Blajet-Kosicka A, Kosicki R, Khan MZ, Rehman H, Böhm J (2012a). Deoxynivalenol as a contaminant of broiler feed: Intestinal development, absorptive functionality, and metabolism of the mycotoxin. Poult Sci 91:852-861

Yunus AW, Ghareeb K, Twaruzek M, Grajewski J, Böhm J (2012b). Deoxynivalenol as a contaminant of broiler feed: Effects on bird performance and response to common vaccines. Poult Sci 91:844-851.

Zinedine A, Meca G, Mañes J, Font G (2011). Further data on the occurrence of *Fusarium* emerging mycotoxins (A, A1, B, B1), fusaproliferin and beauvericin in raw cereals commercialized in Morocco. Food Control 22:1-5.

Zollitsch W, Raffaseder C, Bohm J, Wagner E, Leitgeb R (2003). Impact of the mycotoxins moniliformin and beauvericin on growth and carcass traits of broilers. Wiener Tierarzt Monatsschr 90 (9):238-243.

Zomborszky-Kovacs M, Kovacs F, Vetesi F, Repa I, Tornyos G, Toth A (2002). Investtigations into the time- and dose-dependent effect of fumonisin B1 in order to determine tolerable limit values in pigs. Livestock Prod Sci 76(3):251-256.

Appendix A: Occurrence of *Fusarium* species on Norwegian grown cereals

Table A1: *Fusarium* spp. and *Microdochium nivale* isolated from symptomatic cereal plants 1980-83 (Haave 1985).

Isolated species	Wheat	Barley	Oats	Mean
F. avenaceum	21.6	20.5	25.2	25.0
F. culmorum	25.3	25.6	38.2	29.6
F. graminearum	7.4	4.3	11.5	7.8
F. poae	2.5	0.9		1.2
F. equiseti	1.9	0.9		1.0
F. tricinctum	0.6	0.8		0.5
M. nivale	12.3	37.8	15.9	22.2

Table A2: Occurrence (%) of *Fusarium* spp. from seed samples at five experimental fields 1994-97 (Henriksen 1999).

Fusarium spp.	Kvithamar Nord-Trøndelag	Apelsvoll, Oppland	Brandval, Hedmark	Norderås Akershus	Hauer Akershus	Mean
F. avenaceum	32.3	4.0	45.5	5.9	12.4	20.0
F. tricinctum	3.3	10.2	1.2	2.5	32.3	9.9
F. poae	5.4	6.3	4.0	13.0	10.7	7.9
F. culmorum	1.4	0.2	7.5	1.5	2.2	2.5
F. crookwellense	3.0	0.05	5.5	0.6	0.2	1.9
F. graminearum	2.1	0.5	2.4	0	0	1.0

Table A3: Occurrence (%) of *Fusarium* spp. in wheat samples from four regions 1994-96 (Kosiak et al. 2003).

	South-East	Upper East	Mid-Norway	South-West
F. avenaceum	97.6	100.0	100.0	100.0
F. poae	87.8	95.1	42.9	85.4
F. tricinctum	70.7	65.9	50.0	77.1
F. culmorum	65.9	78.0	28.6	68.8
F. graminearum	9.8	36.6	42.9	12.5
F. equiseti	4.9	29.3	7.1	31.3
F. sporotrichioides	4.9	4.9	0.0	8.3
F. langsethiae	76.5	70.6	56.6	70.0

Table A4: Occurrence (%) of *Fusarium* spp. in barley samples from four regions 1994-96 (Kosiak et al. 2003).

	South-East	Upper East	Mid-Norway	South-West
F. avenaceum	100.0	100.0	100.0	97.4
F. poae	94.4	83.3	43.7	91.0
F. tricinctum	90.7	93.6	62.0	100.0
F. culmorum	77.8	69.2	54.9	82.1
F. graminearum	20.4	26.9	31.0	14.1
F. equiseti	14.8	34.6	4.2	34.6
F. sporotrichioides	3.7	7.7	1.4	9.0
F. langsethiae	71.4	84.0	62.5	73.3

Table A5: Occurrence (%) of *Fusarium* spp. in oats samples from four regions 1994-96 (Kosiak et al. 2003).

	South-East	Upper East	Mid-Norway	South-West
F. avenaceum	98.1	98.8	100.0	98.8
F. poae	96.3	90.0	49.1	96.3
F. tricinctum	83.3	90.0	65.5	88.9
F. culmorum	72.2	77.5	45.5	81.5
F. graminearum	13.0	31.3	72.7	28.4
F. equiseti	20.4	30.0	1.8	24.7
F. sporotrichioides	7.4	1.3	0.0	4.9
F. langsethiae	95.5	96.3	30.0	88.5

Table A6: *Fusarium* spp. in settled grain dust measured by PCR (Halstensen et al. 2006)

***Fusarium* spp.**	**% positive samples**	**Median %**	**Maximum %**
F. avenaceum	94	28	99
F. poae	82	25	96
F. langsethiae	80	52	100
F. sporotrichioides	5	0	20
F. culmorum	40	0	92
F. graminearum	9	0	9

Table A7a: Overview of models predicting *Fusarium* head blight (FHB) disease and DON (deoxynivalenol) in wheat. Models produced in Argentina, the USA and Italy predict the severity/risk of *Fusarium* ear blight (FEB) on wheat from observed weather variables at critical crop growth stages; relative humidity (RH), daily rainfall, temperature and solar radiation. These models can be used to determine the optimum timing of fungicide sprays for control of *Fusarium* head blight.

Reference country	Factor predicted	Weather inputs	Critical weather period (crop growth dependent)	Equations	N	Performance
Argentina[d] (2 models)	Percentage of ears with FEB[b]	RH[c], daily rainfall, daily temperature	From 8 days before ear emergence (growth stage 50) until 550° days have elapsed	$PI\%=20.37+8.63\times NP_2-0.49\times DD_{926}$ $DD_{926}=\sum[(MaxT-26)+(9-MinT)]$ $PI\%=18.34+4.12\times NP12-0.45\times DD_{1026}$ $DD_{1026}=\sum[(MaxT-26)+(10-MinT)]$	?	
USA[e] (4 models)	Risk of a FEB epidemic (field disease severity≥10%)	RH[c], daily rainfall, daily temperature	From 7 days before the start of anthesis (growth stage 60) to 10 days after the start of anthesis	$RFE = -3.38 + 6.81 \times TRH9010$ $RFE = -3.73 + 10.5 \times T15307 \times TRH9010$ $RFE = -1.06 - 14.2 \times T15307 \times DPPT7 + 39.5 \times T15307 \times DPPT7 \times TRH9010$ $RFE = -1.54 + 31.8 \times T15307 \times DPPT7 \times TRH9010 - 5.81 \times DPPT7$	?	
Italy[f]	Risk of a FEB epidemic (field disease severity)	RH[c], daily rainfall, daily max./min. temperature	From ear emergence (heading) until harvest	$FHB\ risk = \sum SPO\times DIS\times INF\times GS^H$ $lnDON = 3.0894\times ln(FHB\ risk) - 3.5231$ $lnZON = 0.2113\times exp(0.054\times FHB\ risk)$	?	
Norway[i]	DON	RH[c]	From two weeks before anthesis to the second week after start of anthesis	$DON = -93 + 1357\ Site + 2213\times Site\times Ploughing - 487.7\times Site\times UM1 - 397.8\times Site\times UM3 - 465.7\times Site\times UM4 + 250.2\times Ploughing\times UM2 - 206.7\times Ploughing\times UM3 + 22.05\times UM2\times UM4$	?	
Canada[j] (3 models)	DON	daily rainfall, min temperature	From 4 to 7 days before heading From 7 days before heading to 10 days after heading When RAINB>0 When RAINB=0	$DON = exp[-0.30 + 1.84\times RAINA - 0.43\times (RAINA)^2 - 0.56TMIN] - 0.1$ $DON = exp[-2.15+2.21\times RAINA-0.61\times (RAINA)^2 +0.85\times RAINB+0.52\times RAINC-0.30TMIN - 1.10\times TMAX] - 0.1$ $DON = exp[-0.84+0.78\times RAINA + 0.40\times (RAINC) -0.42TMIN] - 0.1$	399 (5 years)	$R^2 = 0.55$ $R^2 = 0.79$ $R^2 = 0.56$

[a]There is an additional model from Brazil (Del Ponte et al. 2005) that was investigated but it is very complex and requires the calculation or knowledge of many additional factors such as anther extrusion rate (to calculate the area of available tissue for infection), spore cloud density and infection frequency to produce an overall risk of *Fusarium* ear blight (FEB) infection

[b]As these models were produced outside the UK, the disease is referred to as *Fusarium* head blight (FHB) in the original papers (not fusarium ear blight (FEB))

[c]From a statistical viewpoint, the relative humidity is difficult to work with since it cannot be greater than 100%, so that any formula developed using relative humidity as an input would need to take this into account. [d]Moschini et al. 2001. [e]De Wolf et al. 2003. Field disease severity is the mean percentage of ear area with FEB symptoms in the crop (including all plants with/without symptoms)

[f]Rossi et al. 2003b. Field disease severity is the mean percentage of ear area with FEB symptoms (www.ext.nodak.edu). [g]Notation used in the equations:DD_{926} where MaxT is daily maximum temperature >26°C, MinT is daily minimum temperature <9°C (10°C), and summation occurs over the days of the CPL. NP_2 Number of 2 day periods with precipitation≥0.2 mm and RH >81%, NP_{12} Number of NP_2 periods plus the total number of days with precipitation≥0.2 mm and RH >83%, PI% Predictive index, RFE Risk of a FEB epidemic (epidemic if FEB field disease severity≥10%), T15307 Duration (hours) of 15≤T≤30°C in the 7 days before anthesis, TRH9010 Duration (hours) 15≤T≤30°C and RH≥90% during anthesis [h]FHB risk is calculated daily and accumulated until harvest; equations for each of the components (SPO, sporulation rate; DIS, dispersal rate; INF, infection rate; GS, empirical weight) are given in Rossi et al. (2003b). The model assumes that inoculum is always present. [i]Model in operation for the growing season 2009 and 2010 (at http://www.vips-landbruk.no) . Notation used in the equation: UM1 number of days/week with relative humidity>75% in the week that starts two weeks before the start of anthesis; UM2 number of days/week with relative humidity>75% in the week that starts one week before the start of anthesis; UM3 number of days/week with relative humidity>75% in the week that starts at the first day of anthesis; UM4 number of days/week with relative humidity>75% in the week that starts one week after the start of anthesis (i.e. requires availability of weather forecast)

[j]Hooker et al. 2002

Table A7b: Models predicting DON (deoxynivalenol) in mature oat, to be used for quality sorting (Oleif Elen, unpublished).

Reference country	Factor predicted	Weather inputs	Critical weather period (crop growth dependent)	Equations	N	Performance
Norway	DON	RH	From two weeks before anthesis to the second week after start of anthesis	lnDON+3AC = 5.51722 + 0.344003×lnDONyrbef + 0.0365976×Diff_hø-blr + 0.425707×forFORoats – 0.0807057× RR(24h value) Sum B5 – 1.04742×Resistance – 0.0393101× lnDONyrbef *TM(24h value) Average B1 - 0,040377× lnDONåf*UM(24h value) Threshold>80.0 B-5 + 0.0285777× lnDONåf*TM(24h value) Average B-2 + 0.0273926RR(24h value)Sum B5*Resistance + 0,141498×Site=1*TM(24h value) AverageB-3 – 0.283412× Site=1*UM(24h value) Threshold>80.0 B-8	177	$R^2 = 0.65$ R^2(pred) = 0.6

DON = deoxynivalenol; RH = Relavive humidity; 3AC = 3-acetyldeoxynivalenol.

Table A8: Samples of certified seed grain with sclerotia of ergot (Claviceps) 2007-2010. One to six sclerotia were registered per sample. Most samples had only one sclerotium. (Source: Kimen Såvarelaboratorie AS. 03.05.2011).

Species	2007		2008		2009		2010	
	Number of samples with sclerotia	Total number of samples	Number of samples with sclerotia	Total number of samples	Number of samples with sclerotia	Total number of samples	Number of samples with sclerotia	Total number of samples
Barley	10	1173	12	1161	9	1214	9	1274
Oats	3	596	6	658	4	345	5	494
Spring wheat	14	508	5	523	2	432	4	569
Winterwheat	0	252	0	221	1	120	1	140
Total	27	2529	23	2563	16	2111	19	2477

Table A9: Price reduction according to DON (deoxynivalenol) content in oat grain in year 2012. The "basic" price per kilo oat grain in 2012 was 2.10 NOK /kg.

DON content (µg/kg)	Price reduction (øre/kg)
0 - 2000	0
2000 - 5000	9
5001 - 8000	14
> 8000	25

Appendix B: Occurence of mycotocins in cereal grain

DEOXYNIVALENOL (DON): Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	138	103	125	80	0.0	890
	food grain (imported)	27	110	212	20	0.0	660
	bran (Norwegian)	10	19	13	15	5.5	33
1991	food grain (Norwegian)	107	84	61	68	0.0	310
1992	food grain (Norwegian)	112	191	173	173	0.0	895
	food grain (imported)	16	407	421	245	0.0	1300
1993	food grain (Norwegian)	102	94	101	67	0.0	560
	food grain (imported)	29	395	580	154	0.0	2500
1994	food grain (Norwegian)	112	42	67	15	0.0	340
	food grain (imported)	30	79	145	17	0.0	610
1995	food grain (Norwegian)	26	29	31	15	0.0	165
	food grain (imported)	13	154	228	48	0.0	650
1996	food grain (Norwegian)	28	10	0.0	10	0.0	20
	food grain (imported)	14	333	714	30	0.0	2700
1997	food grain (Norwegian)	28	19	17	10	0.0	73
	food grain (imported)	10	356	591	45	0.0	1900
1998	food grain (Norwegian)	35	13	13	10	0.0	85
	food grain (imported)	24	77	75	50	0.0	233
	crude grain	24	18	19	10	-	-
1999	crude grain	83	16	15	10	-	-
2000	crude grain	59	109	241	10	-	-
2001	crude grain	18	155	199	62	-	-
2002	crude grain	74	62	143	23	-	-
2003	crude grain	88	83	172	25	-	-
2004	crude grain	60	256	386	138	-	-
2005	crude grain	93	209	342	76	-	-
2006	crude grain	22	153	411	10	-	-
2007	crude grain	6	130	160	52	-	-
2008	milled flour (Norwegian)	33	141	120	160	0.0	430
	milled flour (imported)	18	150	96	143	8.0	480
	sieved flour (Norwegian)	24	223	117	202	14	492
	sieved flour (imported)	38	201	121	197	0.0	639
	bran (Norwegian)	6	450	315	573	28	820
	bran (imported)	17	359	231	381	35	1093
	groat (Norwegian)	1	8.0	0.0	8.0	8.0	8.0
	infant porridge	4	13	9.0	14	0.0	23
2009	milled flour (Norwegian)	17	229	187	138	0.0	623
	milled flour (imported)	25	113	114	67	0.0	480
	sieved flour (Norwegian)	18	156	74	141	0.0	299
	sieved flour (imported)	31	186	248	121	0.0	1123
	bran (Norwegian)	1	167	0.0	167	167	167
	bran (imported)	19	140	44	129	104	295
	groat (Norwegian)	1	112	0.0	112	112	112
	groat (imported)	1	10	0.0	10	0.0	20
2010	milled flour (Norwegian)	15	210	254	112	24	997
	milled flour (imported)	27	72	35	66	28	161
	sieved flour (Norwegian)	11	237	267	101	47	935
	sieved flour (imported)	39	113	97	85	0.0	446
	bran (Norwegian)	9	263	216	197	60	798
	bran (imported)	17	179	120	161	14	438
	groat (Norwegian)	2	370	358	370	117	623
2011	milled flour	29	240	115	206	0.0	504
	sieved flour	37	200	107	174	44	501
	bran	24	241	275	156	61	1364

DEOXYNIVALENOL (DON): Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	10	33	15	30	0.0	55
	bran (Norwegian)	10	19	13	15	1.5	33
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	crude grain	31	63	256	10	-	-
1999	crude grain	56	11	3.0	10	-	-
2000	crude grain	46	12	5.0	10	-	-
2001	crude grain	38	24	34	5.0	-	-
2002	crude grain	80	27	43	10	-	-
2003	crude grain	110	36	60	10	-	-
2004	crude grain	122	101	253	10	-	-
2005	crude grain	114	113	238	10	-	-
2006	crude grain	88	36	121	10	-	-
2007	crude grain	56	85	143	16	-	-
2008	crude grain	28	173	206	79	-	-
	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	crude grain	13	315	298	260	-	-
	food grain (Norwegian)	3	318	55	298	275	380
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

DEOXYNIVALENOL (DON): Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	20	111	73	98	80	690
	oat flakes (Norwegian)	20	262	182	225	30	270
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	food grain (Norwegian)	3	470	296	610	470	470
1994	food grain (Norwegian)	3	538	665	240	75	1300
1995	food grain (Norwegian)	25	61	57	45	0.0	230
1996	food grain (Norwegian)	14	104	98	82	0.0	266
1997	food grain (Norwegian)	11	61	34	60	20	130
1998	food grain (Norwegian)	22	134	199	52	0.0	849
	crude grain	42	27	40	13	-	-
1999	crude grain	82	55	123	10	-	-
	infant porridge	3	10	0.0	10	0.0	20
2000	crude grain	61	26	50	10	-	-
2001	crude grain	45	39	58	20	-	-
2002	crude grain	77	54	75	25	-	-
2003	crude grain	112	245	575	39	-	-
2004	crude grain	136	842	2882	70	-	-
2005	crude grain	355	1623	3889	391	-	-
2006	crude grain	245	878	2923	71	-	-
2007	crude grain	138	858	1373	261	-	-
2008	crude grain	77	1668	2142	1199	-	-
	oat flakes (Norwegian)	19	261	136	217	94	485
	oat flakes (imported)	12	322	156	372	20	520
	infant porridge	5	34	26	15	0.0	70
2009	crude grain	30	2149	1760	1565	-	-
	oat flakes (Norwegian)	19	388	204	370	119	776
	oat flakes (imported)	15	249	250	132	25	723
2010	oat flakes (Norwegian)	14	178	139	162	0.0	523
	oat flakes (imported)	17	170	94	176	26	337
2011	oat flakes	31	160	77	166	0.0	291

DEOXYNIVALENOL (DON): Prevalence in spelt grains and spelt-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	food grain (Norwegian)	3	32	18	24	14	49
	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	food grain (Norwegian)	2	9.0	1.4	9.0	0.0	10
	food grain (imported)	2	37	38	37	0.0	64
2010	food grain (Norwegian)	2	35	35	35	0.0	60
	food grain (imported)	2	81	100	81	0.0	152
2011	-	-	-	-	-	-	-

DEOXYNIVALENOL (DON): Prevalence in rye grains and rye-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	2	60	0.0	60	60	60
	food grain (imported)	18	26	14	20	0.0	50
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	food grain (imported)	11	25	17	15	0.0	62
1994	food grain (imported)	12	28	17	22	0.0	60
1995	food grain (imported)	11	25	15	19	0.0	57
1996	food grain (imported)	10	10	0.0	10	0.0	20
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

DEOXYNIVALENOL (DON): Prevalence in maize grains and maize-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	infant porridge	8	866	86	819	792	1022
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	3	42	7	46	34	46
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

DEOXYNIVALENOL (DON): Prevalence in sorghum grains and sorghum-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

DEOXYNIVALENOL (DON): Prevalence in mixed-cereals foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	baby food	6	11	10	8.0	0.0	31
2009	-	-	-	-	-	-	-
2010	breakfast cereals (Norwegian)	8	121	70	125	0.0	230
	breakfast cereals (imported)	21	47	44	38	0.0	167
2011	-	-	-	-	-	-	-

T2: Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	food grain (Norwegian)	28	15	0.0	15	0.0	30
	food grain (imported)	14	15	0.0	15	0.0	30
1997	food grain (Norwegian)	28	21	21	15	7.7	35
	food grain (imported)	10	15	0.0	15	0.0	30
1998	food grain (Norwegian)	35	15	0.0	15	0.0	30
	food grain (imported)	24	15	0.0	15	0.0	30
	crude grain	24	11	2.2	10	-	-
1999	crude grain	83	14	2.2	15	-	-
2000	crude grain	59	13	4.2	10	-	-
2001	crude grain	18	5.3	1.2	5.0	-	-
2002	crude grain	74	15	1.3	15	-	-
2003	crude grain	88	15	0.0	15	-	-
2004	crude grain	60	15	0.0	15	-	-
2005	crude grain	93	15	0.0	15	-	-
2006	crude grain	22	14	2.3	15	-	-
2007	crude grain	6	13	4.1	15	-	-
2008	milled flour (Norwegian)	33	2.5	0.0	2.5	0.0	5.0
	milled flour (imported)	18	2.5	0.0	2.5	0.0	5.0
	sieved flour (Norwegian)	24	2.5	0.0	2.5	0.0	5.0
	sieved flour (imported)	38	2.6	0.4	2.5	0.0	10
	bran (Norwegian)	6	3.3	1.3	2.5	0.0	10
	bran (imported)	17	7.2	9.8	5.0	0.0	45
	groat (Norwegian)	1	2.5	0.0	2.5	0.0	5.0
	infant porridge	4	4.4	1.3	5.0	0.0	10
2009	milled flour (Norwegian)	17	5.3	6.5	1.3	0.0	30
	milled flour (imported)	25	9.7	6.7	15	0.0	30
	sieved flour (Norwegian)	18	6.6	6.9	1.3	0.0	30
	sieved flour (imported)	31	8.5	6.9	15	0.0	30
	bran (Norwegian)	1	20	0.0	20	0.0	40
	bran (imported)	19	11	9.2	15	2.2	40
	groat (Norwegian)	1	1.3	0.0	1.3	0.0	2.5
	groat (imported)	1	15	0.0	15	0.0	30
2010	milled flour (Norwegian)	15	14	3.5	15	0.0	30
	milled flour (imported)	27	9.8	5.1	5.0	0.0	30
	sieved flour (Norwegian)	11	15	0.0	15	0.0	30
	sieved flour (imported)	39	12	4.6	15	0.0	30
	bran (Norwegian)	9	27	7.0	30	0.0	60
	bran (imported)	17	23	10	27	0.0	60
	groat (Norwegian)	2	15	0.0	15	0.0	30
2011	milled flour	29	15	0.0	15	0.0	30
	sieved flour	37	15	0.0	15	0.0	30
	bran	24	30	0.0	30	0.0	60

T2: Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	crude grain	31	10	1.0	10	-	-
1999	crude grain	56	17	11	15	-	-
2000	crude grain	46	11	2.1	10	-	-
2001	crude grain	38	7.7	9.3	5.0	-	-
2002	crude grain	80	15	4.3	15	-	-
2003	crude grain	110	15	3.0	15	-	-
2004	crude grain	122	17	8.3	15	-	-
2005	crude grain	114	16	3.6	15	-	-
2006	crude grain	88	15	3.8	15	-	-
2007	crude grain	56	17	18	15	-	-
2008	crude grain	28	16	4.2	15	-	-
	infant porridge	1	2.5	0.0	2.5	0.0	5.0
2009	crude grain	13	18	12	15	-	-
	food grain (Norwegian)	3	15	0.0	15	0.0	30
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

T2: Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	food grain (Norwegian)	14	50	81	16	0.0	317
1997	food grain (Norwegian)	11	50	56	25	0.0	195
1998	food grain (Norwegian)	22	20	9.2	15	0.0	42
	crude grain	42	15	9.8	10	-	-
1999	crude grain	82	19	13	15	-	-
2000	crude grain	61	19	21	10	-	-
2001	crude grain	45	21	30	10	-	-
2002	crude grain	77	40	61	15	-	-
2003	crude grain	112	33	49	15	-	-
2004	crude grain	136	30	27	15	-	-
2005	crude grain	355	45	70	15	-	-
2006	crude grain	245	46	81	15	-	-
2007	crude grain	138	43	46	22	-	-
2008	crude grain	77	25	28	15	-	-
	oat flakes (Norwegian)	19	10	6.4	8.0	0.0	24
	oat flakes (imported)	12	9.1	3.9	10	0.0	16
	infant porridge	5	4.0	1.4	5.0	0.0	10
2009	crude grain	30	28	37	15	-	-
	oat flakes (Norwegian)	19	6.4	4.5	5.8	0.0	30
	oat flakes (imported)	15	10	6.3	15	0.0	30
2010	oat flakes (Norwegian)	14	14	2.7	15	0.0	30
	oat flakes (imported)	17	11	5.0	15	0.0	30
2011	oat flakes	31	15	0.0	15	0.0	30

T2: Prevalence in spelt grains and spelt-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	food grain (Norwegian)	3	2.5	0.0	2.5	0.0	5.0
	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	food grain (Norwegian)	2	8.1	9.7	8.1	0.0	30
	food grain (imported)	2	15	0.0	15	0.0	30
2010	food grain (Norwegian)	2	15	0.0	15	0.0	30
	food grain (imported)	2	15	0.0	15	0.0	30
2011	-	-	-	-	-	-	-

T2: Prevalence in rye grains and rye-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	food grain (imported)	4	15	0.0	15	0.0	30
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

T2: Prevalence in maize grains and maize-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	3	4.2	1.4	5.0	0.0	10
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

T2: Prevalence in sorghum grains and sorghum-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

T2: Prevalence in mixed-cereals foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	6	2.9	1.0	2.5	0.0	10
2009	-	-	-	-	-	-	-
2010	breakfast cereals (Norwegian)	8	14	3.5	15	0.0	30
	breakfast cereals (imported)	21	13	4.4	15	0.0	30
2011	-	-	-	-	-	-	-

HT-2: Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	food grain (Norwegian)	28	10	0.0	10	0.0	20
	food grain (imported)	14	11	2.7	10	0.0	20
1997	food grain (Norwegian)	28	22	77	10	25	42
	food grain (imported)	10	10	0.0	10	0.0	20
1998	food grain (Norwegian)	35	10	0.0	10	0.0	20
	food grain (imported)	24	10	0.0	10	0.0	20
	crude grain	24	11	2.1	10	-	-
1999	crude grain	83	14	2.2	15	-	-
2000	crude grain	59	13	5.1	10	-	-
2001	crude grain	18	6.1	2.6	5.0	-	-
2002	crude grain	74	9.9	0.6	10	-	-
2003	crude grain	88	10	0.0	10	-	-
2004	crude grain	60	10	0.0	10	-	-
2005	crude grain	93	10	1.2	10	-	-
2006	crude grain	22	9.9	1.2	10	-	-
2007	crude grain	6	9.2	2.0	10	-	-
2008	milled flour (Norwegian)	33	2.5	0.0	2.5	0.0	5.0
	milled flour (imported)	18	2.5	0.0	2.5	0.0	5.0
	sieved flour (Norwegian)	24	2.5	0.0	2.5	0.0	5.0
	sieved flour (imported)	38	2.8	0.4	2.5	0.0	10
	bran (Norwegian)	6	3.3	1.3	2.5	0.0	10
	bran (imported)	17	4.7	0.8	5.0	0.0	10
	groat (Norwegian)	1	2.5	0.0	2.5	0.0	5.0
	infant porridge	4	4.4	1.3	5.0	0.0	8.8
2009	milled flour (Norwegian)	17	4.4	4.0	1.3	0.0	20
	milled flour (imported)	25	7.0	4.0	10	0.0	20
	sieved flour (Norwegian)	18	4.7	4.3	1.3	0.0	20
	sieved flour (imported)	31	5.9	4.3	10	0.0	20
	bran (Norwegian)	1	20	0.0	20	0.0	40
	bran (imported)	19	13	11	10	0.0	40
	groat (Norwegian)	1	1.3	0.0	1.3	0.0	2.5
	groat (imported)	1	10	0.0	10	0.0	20
2010	milled flour (Norwegian)	15	9.3	1.8	10	0.0	20
	milled flour (imported)	27	7.4	2.5	5.0	0.0	20
	sieved flour (Norwegian)	11	10	0.0	10	0.0	20
	sieved flour (imported)	39	8.6	4.2	10	0.0	20
	bran (Norwegian)	9	21	9.0	20	0.0	40
	bran (imported)	17	21	10	20	0.0	40
	groat (Norwegian)	2	10	0.0	10	0.0	20
2011	milled flour	29	10	0.0	10	0.0	20
	sieved flour	37	10	0.0	10	0.0	20
	bran	24	20	0.0	20	0.0	40

HT-2: Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	crude grain	31	12	5.0	10	-	-
1999	crude grain	56	22	26	15	-	-
2000	crude grain	46	12	4.4	10	-	-
2001	crude grain	38	20	33	5.0	-	-
2002	crude grain	80	18	17	10	-	-
2003	crude grain	110	18	16	10	-	-
2004	crude grain	122	20	31	10	-	-
2005	crude grain	114	21	18	10	-	-
2006	crude grain	88	26	27	10	-	-
2007	crude grain	56	30	64	10	-	-
2008	crude grain	28	71	305	10	-	-
	infant porridge	1	2.5	0.0	2.5	0.0	5.0
2009	crude grain	13	16	14	10	-	-
	food grain (Norwegian)	3	10	0.0	10	0.0	20
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

HT-2: Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	food grain (Norwegian)	14	142	114	108	33	401
1997	food grain (Norwegian)	11	194	254	80	20	710
1998	food grain (Norwegian)	22	66	107	38	0.0	522
	crude grain	42	52	61	28	-	-
1999	crude grain	82	53	79	27	-	-
2000	crude grain	61	43	62	15	-	-
2001	crude grain	45	46	54	25	-	-
2002	crude grain	77	102	194	31	-	-
2003	crude grain	112	103	212	52	-	-
2004	crude grain	136	83	109	34	-	-
2005	crude grain	355	191	259	114	-	-
2006	crude grain	245	250	592	118	-	-
2007	crude grain	138	151	138	115	-	-
2008	crude grain	77	89	105	55	-	-
	oat flakes (Norwegian)	19	39	24	31	15	100
	oat flakes (imported)	12	32	12	37	0.0	49
	infant porridge	5	6.2	5.6	5.0	0.0	10
2009	crude grain	30	106	128	56	-	-
	oat flakes (Norwegian)	19	23	11	26	0.0	37
	oat flakes (imported)	15	13	10	10	0.0	45
2010	oat flakes (Norwegian)	14	11	4.9	10	0.0	24
	oat flakes (imported)	17	8.1	2.5	10	0.0	20
2011	oat flakes	31	19	10	10	0.0	36

HT-2: Prevalence in spelt grains and spelt-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	food grain (Norwegian)	3	2.5	0.0	2.5	0.0	5.0
	infant porridge	1	5.0	0.0	5.0	0.0	5.0
2009	food grain (Norwegian)	2	5.8	1.3	10	0.0	20
	food grain (imported)	2	10	0.0	10	0.0	20
2010	food grain (Norwegian)	2	10	0.0	10	0.0	20
	food grain (imported)	2	10	0.0	10	0.0	20
2011	-	-	-	-	-	-	-

HT-2: Prevalence in rye grains and rye-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	food grain (imported)	4	10	0.0	10	0.0	20
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

HT-2: Prevalence in maize grains and maize-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	3	4.2	1.4	5.0	0.0	10
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

HT-2: Prevalence in sorghum grains and sorghum-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

HT-2: Prevalence in mixed-cereals foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	6	2.9	1.0	2.5	0.0	10
2009	-	-	-	-	-	-	-
2010	breakfast cereals (Norwegian)	8	10	0.4	10	0.0	20
	breakfast cereals (imported)	21	9.0	2.0	10	0.0	20
2011	-	-	-	-	-	-	-

NIVALENOL: Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	138	25	0.0	25	0.0	50
	food grain (imported)	27	27	7.7	25	0.0	65
	bran (Norwegian)	10	19	13	15	5.5	33
1991	food grain (Norwegian)	107	35	28	25	0.0	170
1992	food grain (Norwegian)	112	25	0.0	25	0.0	50
	food grain (imported)	16	25	0.0	25	0.0	50
1993	food grain (Norwegian)	102	25	0.0	25	0.0	50
	food grain (imported)	29	25	0.0	25	0.0	50
1994	food grain (Norwegian)	112	25	0.0	25	0.0	50
	food grain (imported)	30	28	17	26	0.0	120
1995	food grain (Norwegian)	26	25	0.0	25	0.0	50
	food grain (imported)	13	25	0.0	25	0.0	50
1996	food grain (Norwegian)	28	10	0.0	10	0.0	20
	food grain (imported)	14	10	0.0	10	0.0	20
1997	food grain (Norwegian)	28	14	12	10	0.0	60
	food grain (imported)	10	40	81	10	0.0	270
1998	food grain (Norwegian)	35	10	0.0	10	0.0	20
	food grain (imported)	24	12	8.1	10	0.0	38
	crude grain	24	11	2.1	10	-	-
1999	crude grain	83	12	2.5	10	-	-
2000	crude grain	59	11	2.7	10	-	-
2001	crude grain	18	5.3	1.2	5.0	-	-
2002	crude grain	74	15	1.3	15	-	-
2003	crude grain	88	15	0.0	15	-	-
2004	crude grain	60	15	0.0	15	-	-
2005	crude grain	93	15	0.0	15	-	-
2006	crude grain	22	15	3.8	15	-	-
2007	crude grain	6	13	4.1	15	-	-
2008	milled flour (Norwegian)	33	2.7	1.0	2.5	0.0	8.0
	milled flour (imported)	18	4.2	5.9	2.5	0.0	27
	sieved flour (Norwegian)	24	2.5	0.0	2.5	0.0	5.0
	sieved flour (imported)	38	2.8	1.3	2.5	0.0	10
	bran (Norwegian)	6	9.5	11	2.5	0.0	25
	bran (imported)	17	8.4	6.3	5.0	0.0	20
	groat (Norwegian)	1	2.5	0.0	13	0.0	25
	infant porridge	4	4.4	1.3	5.0	0.0	10
2009	milled flour (Norwegian)	17	13	1.2	13	0.0	30
	milled flour (imported)	25	13	3.3	15	0.0	30
	sieved flour (Norwegian)	18	14	1.3	13	0.0	30
	sieved flour (imported)	31	14	2.0	15	0.0	30
	bran (Norwegian)	1	30	0.0	30	0.0	60
	bran (imported)	19	21	8.4	15	0.0	60
	groat (Norwegian)	1	13	0.0	13	0.0	25
	groat (imported)	1	15	0.0	15	0.0	30
2010	milled flour (Norwegian)	14	14	3.6	15	0.0	30
	milled flour (imported)	24	9.2	5.0	5.0	0.0	30
	sieved flour (Norwegian)	10	15	0.0	15	0.0	30
	sieved flour (imported)	36	13	5.0	15	0.0	40
	bran (Norwegian)	9	35	21	30	0.0	60
	bran (imported)	16	25	12	27	0.0	60
	groat (Norwegian)	2	15	0.0	15	0.0	30
2011	milled flour	23	15	0.0	15	0.0	30
	sieved flour	29	15	0.0	15	0.0	30
	bran	20	30	0.0	30	0.0	60

NIVALENOL: Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	10	26	2.1	25	0.0	50
	bran (Norwegian)	10	25	0.0	25	0.0	50
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	crude grain	31	12	8.6	10	-	-
1999	crude grain	56	11	3.3	10	-	-
2000	crude grain	46	11	6.9	10	-	-
2001	crude grain	38	6.1	3.8	5.0	-	-
2002	crude grain	80	16	6.6	15	-	-
2003	crude grain	110	22	21	15	-	-
2004	crude grain	122	18	18	15	-	-
2005	crude grain	114	16	5.1	15	-	-
2006	crude grain	88	16	3.3	15	-	-
2007	crude grain	56	18	21	15	-	-
2008	crude grain	28	18	17	15	-	-
	infant porridge	1	2.5	0.0	2.5	0.0	5.0
2009	crude grain	13	21	20	15	-	-
	food grain (Norwegian)	3	15	0.0	15	0.0	30
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

NIVALENOL: Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	20	30	21	25	0.0	120
	oat flakes (Norwegian)	20	20	0.0	20	0.0	40
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	food grain (Norwegian)	3	40	25	25	0.0	69
1994	food grain (Norwegian)	3	25	0.0	25	0.0	50
1995	food grain (Norwegian)	25	25	0.0	25	0.0	50
1996	food grain (Norwegian)	14	10	0.0	10	0.0	20
1997	food grain (Norwegian)	11	14	6.7	10	0.0	30
1998	food grain (Norwegian)	22	10	0.0	10	0.0	20
	crude grain	42	11	61	28	-	-
1999	crude grain	82	12	79	27	-	-
	infant porridge	3	10	0.0	10	0.0	20
2000	crude grain	61	14	62	15	-	-
2001	crude grain	45	8.2	54	25	-	-
2002	crude grain	77	33	194	31	-	-
2003	crude grain	112	50	212	52	-	-
2004	crude grain	136	29	109	34	-	-
2005	crude grain	355	21	259	114	-	-
2006	crude grain	245	25	592	118	-	-
2007	crude grain	138	22	138	115	-	-
2008	crude grain	77	24	105	55	-	-
	oat flakes (Norwegian)	19	3.6	3.7	2.5	0.0	18
	oat flakes (imported)	12	2.5	0.0	2.5	0.0	5.0
	infant porridge	5	4.0	1.4	5.0	0.0	10
2009	crude grain	30	29	128	56	-	-
	oat flakes (Norwegian)	19	12	3.0	13	0.0	30
	oat flakes (imported)	15	14	1.0	15	0.0	30
2010	oat flakes (Norwegian)	13	14	2.8	15	0.0	30
	oat flakes (imported)	17	11	5.0	15	0.0	30
2011	oat flakes	28	15	0.0	15	0.0	30

NIVALENOL: Prevalence in spelt grains and spelt-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	food grain (Norwegian)	3	2.5	0.0	2.5	0.0	5.0
	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	food grain (Norwegian)	2	14	1.8	14	0.0	30
	food grain (imported)	2	15	0.0	15	0.0	30
2010	food grain (Norwegian)	1	15	0.0	15	0.0	30
	food grain (imported)	2	15	0.0	15	0.0	30
2011	-	-	-	-	-	-	-

NIVALENOL: Prevalence in rye grains and rye-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	2	25	0.0	25	0.0	50
	food grain (imported)	18	25	0.0	25	0.0	50
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	food grain (imported)	11	25	0.0	25	0.0	50
1994	food grain (imported)	12	25	0.0	25	0.0	50
1995	food grain (imported)	11	25	0.0	25	0.0	50
1996	food grain (imported)	10	10	0.0	10	0.0	20
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

NIVALENOL: Prevalence in maize grains and maize-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	infant porridge	8	18	6.9	21	0.0	26
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	3	4.2	1.4	5.0	0.0	10
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

NIVALENOL: Prevalence in sorghum grains and sorghum-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	1	5.0	0.0	5.0	0.0	10
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

NIVALENOL: Prevalence in mixed-cereals foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	6	2.9	1.0	2.5	0.0	10
2009	-	-	-	-	-	-	-
2010	breakfast cereals (Norwegian)	8	15	0.4	15	0.0	30
	breakfast cereals (imported)	19	14	5.0	15	0.0	40
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	138	2.7	1.5	2.5	0.0	11
	food grain (imported)	27	2.5	0.1	2.5	0.0	3.0
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	food grain (Norwegian)	32	3.6	12	1.5	0.0	68
	food grain (imported)	13	1.5	0.0	1.5	0.0	20
1996	food grain (Norwegian)	28	10	0.0	10	0.0	20
	food grain (imported)	14	10	0.0	10	0.0	20
1997	food grain (Norwegian)	28	2.8	7.3	2.1	0.0	35
	food grain (imported)	10	2.0	4.0	0.5	0.0	13
1998	food grain (Norwegian)	35	1.3	1.9	1.0	0.0	13
	food grain (imported)	24	1.4	0.9	1.0	0.0	4.0
1999	infant porridge	15	0.9	0.5	1.0	0.0	1.7
2000	infant porridge	30	0.7	0.5	0.8	0.0	2.2
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	milled flour (Norwegian)	33	2.5	1.7	1.3	0.0	7.7
	milled flour (imported)	18	2.8	2.1	1.6	0.0	7.8
	sieved flour (Norwegian)	24	1.3	0.3	1.3	0.0	2.5
	sieved flour (imported)	38	1.7	2.4	2.4	0.0	16
	bran (Norwegian)	6	6.8	3.5	6.7	2.4	11
	bran (imported)	17	8.0	5.5	6.5	0.0	21
	groat (Norwegian)	1	1.3	0.0	1.3	0.0	2.5
	infant porridge	4	1.4	0.1	1.5	0.0	3.0
2009	milled flour (Norwegian)	17	5.1	5.0	3.0	0.0	15
	milled flour (imported)	25	3.0	2.8	1.3	0.0	8.5
	sieved flour (Norwegian)	18	1.5	1.1	1.3	0.0	5.4
	sieved flour (imported)	31	2.7	4.5	1.3	0.0	21
	bran (Norwegian)	1	5.1	0.0	5.1	5.1	5.1
	bran (imported)	19	6.3	3.7	6.2	0.0	9.4
	groat (Norwegian)	1	9.0	0.0	9.0	9.0	9.0
	groat (imported)	1	1.0	0.0	1.0	0.0	2.0
2010	milled flour (Norwegian)	15	4.4	4.1	3.5	0.0	15.6
	milled flour (imported)	27	1.6	0.9	1.5	0.0	4.6
	sieved flour (Norwegian)	11	2.7	2.0	1.0	0.0	5.5
	sieved flour (imported)	39	1.4	0.9	1.0	0.0	4.8
	bran (Norwegian)	9	7.1	10	2.0	0.0	26
	bran (imported)	17	4.6	5.4	2.7	0.0	23
	groat (Norwegian)	2	17	17	17	4.4	29
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	10	2.5	0.0	2.5	0.0	5.0
	bran (Norwegian)	10	3.4	2.7	2.5	0.0	11
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	1	1.3	0.0	1.3	0.0	2.5
2009	food grain (Norwegian)	3	2.1	0.9	2.5	0.0	2.7
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	20	2.5	0.0	2.5	0.0	5.0
	oat flakes (Norwegian)	20	4.5	9.1	2.5	0.0	43
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	food grain (Norwegian)	2	2.5	0.0	2.5	0.0	5.0
1995	food grain (Norwegian)	20	4.6	14	1.5	0.0	64
1996	food grain (Norwegian)	14	10	0.0	10	0.0	20
1997	food grain (Norwegian)	11	3.0	7.0	0.5	0.0	24
1998	food grain (Norwegian)	22	1.0	0.0	1.0	0.0	2.0
1999	infant porridge	10	0.9	1.5	0.3	0.0	4.8
2000	infant porridge	16	0.4	0.3	0.3	0.0	1.4
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	oat flakes (Norwegian)	19	1.4	0.8	1.3	0.0	4.7
	oat flakes (imported)	12	2.7	2.6	1.3	0.0	10
	infant porridge	5	2.0	0.6	2.0	0.0	5.0
2009	oat flakes (Norwegian)	19	7.2	11	3.0	0.0	50
	oat flakes (imported)	15	5.2	10	1.3	0.0	39
2010	oat flakes (Norwegian)	14	2.4	3.3	1.0	0.0	13
	oat flakes (imported)	17	1.1	0.3	1.0	0.0	3.0
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in spelt grains and spelt-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	infant porridge	2	0.4	0.2	0.4	0.0	0.5
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	food grain (Norwegian)	3	1.3	0.0	1.3	0.0	2.5
	infant porridge	1	2.5	0.0	2.5	0.0	5.0
2009	food grain (Norwegian)	2	1.1	0.2	1.1	0.0	2.5
	food grain (imported)	2	1.0	0.0	1.0	0.0	2.0
2010	food grain (Norwegian)	2	1.6	0.8	1.6	0.0	2.1
	food grain (imported)	2	1.0	0.0	1.0	0.0	2.0
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in rye grains and rye-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	2	2.5	0.0	2.5	0.0	5.0
	food grain (imported)	18	2.5	0.0	2.5	0.0	5.0
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	food grain (imported)	4	1.7	0.4	1.5	0.0	2.0
1996	food grain (imported)	4	10	0.0	10	0.0	10
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in maize grains and maize-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	infant porridge	22	81	104	6.1	1.2	305
2000	-	3	10	15	3.8	1.0	33
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	3	1.4	0.1	1.5	0.0	3
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in sorghum grains and sorghum-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	1	2.5	0.0	2.5	0.0	5.0
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

ZEARALENONE: Prevalence in mixed-cereals foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	infant porridge	9	0.7	0.4	0.7	0.0	1.2
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	infant porridge	6	1.5	0.5	1.3	0.0	5.0
2009	-	-	-	-	-	-	-
2010	breakfast cereals (Norwegian)	8	2.2	1.6	1.8	0.0	5.2
	breakfast cereals (imported)	21	2.6	4.5	1.0	0.0	26
2011	-	-	-	-	-	-	-

FUMINOSIN (B1+B2): Prevalence in maize grains and maize-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	tinned fresh corn	7	10	0.0	10	0.0	20
	corn cob	10	10	0.0	10	0.0	20
	tinned kernel	11	10	0.0	10	0.0	20
2001	tinned fresh corn	11	10	0.0	10	0.0	20
	corn cob	11	10	0.0	10	0.0	20
	tinned kernel	29	10	0.0	10	0.0	20
	meal	26	321	362	174	0.0	1479
	ecological meal	4	1870	1874	1747	0.0	3967
	semolina	2	1158	1566	1158	40	2265
	starch	3	10	0.0	10	0.0	20
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

ENNIATIN (B + B1): Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	12	-	-	126	-	1590
2001	crude grain (Norwegian)	34	-	-	730	-	7400
2002	crude grain (Norwegian)	34	-	-	297	-	2230
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

ENNIATIN (B + B1): Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	19	-	-	153	-	1213
2001	crude grain (Norwegian)	23	-	-	352	-	5100
2002	crude grain (Norwegian)	33	-	-	493	-	3540
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

ENNIATIN (B + B1): Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	21	-	-	19	-	440
2001	crude grain (Norwegian)	26	-	-	49	-	223
2002	crude grain (Norwegian)	26	-	-	65	-	255
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

BEAUVERICIN: Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	min [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	12	-	-	< 3.0	0.0	3
2001	crude grain (Norwegian)	34	-	-	< 3.0	0.0	3.8
2002	crude grain (Norwegian)	34	-	-	< 3.0	0.0	4.7
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

BEAUVERICIN: Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	19	-	-	< 3.0	0.0	3.7
2001	crude grain (Norwegian)	23	-	-	< 3.0	0.0	3.7
2002	crude grain (Norwegian)	33	-	-	< 3.0	0.0	21
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

BEAUVERICIN: Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	21	-	-	< 3.0	0.0	16
2001	crude grain (Norwegian)	26	-	-	7.8	-	32
2002	crude grain (Norwegian)	26	-	-	19	-	120
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

MONILIFORMIN: Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	13	< 40	-	< 40	0.0	87
2001	crude grain (Norwegian)	35	92	-	69	0.0	420
2002	crude grain (Norwegian)	35	210	-	120	0.0	950
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

MONILIFORMIN: Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	19	< 40	-	< 40	0.0	43
2001	crude grain (Norwegian)	23	46	-	< 40	0.0	380
2002	crude grain (Norwegian)	33	48	-	< 40	0.0	230
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

MONILIFORMIN: Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	crude grain (Norwegian)	21	< 40	-	< 40	0.0	70
2001	crude grain (Norwegian)	26	< 40	-	< 40	0.0	88
2002	crude grain (Norwegian)	26	73	-	59	0.0	210
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

OCHRATOXIN: Prevalence in wheat grains and wheat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	138	0.2	0.2	0.2	0.0	1.5
	food grain (imported)	27	0.3	0.8	0.2	0.0	3.8
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	food grain (Norwegian)	7	0.8	1.7	0.2	0.0	4.7
	food grain (imported)	11	0.6	1.4	0.2	0.0	4.6
1994	food grain (Norwegian)	24	0.3	0.7	0.1	0.0	3.4
	food grain (imported)	9	0.5	1.1	0.1	0.0	3.3
1995	food grain (Norwegian)	32	0.1	0.1	0.1	0.0	0.6
	food grain (imported)	13	0.9	2.2	0.1	0.0	8.2
1996	food grain (Norwegian)	28	0.2	0.0	1.5	0.0	3.0
	food grain (imported)	14	0.7	2.0	0.2	0.0	7.5
1997	food grain (Norwegian)	28	0.2	0.7	0.0	0.0	3.5
	food grain (imported)	10	0.1	0.2	0.0	0.0	0.6
1998	food grain (Norwegian)	35	0.7	3.4	0.0	0.0	20
	food grain (imported)	24	0.1	0.1	0.0	0.0	0.5
1999	infant porridge	15	0.14	0.07	0.14	0.05	0.29
2000	infant porridge	30	0.22	0.25	0.12	0.00	0.99
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

OCHRATOXIN: Prevalence in barley grains and barley-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	10	0.2	0.0	0.2	0.0	0.3
	bran (Norwegian)	10	0.2	0.0	0.2	0.0	0.3
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	crude grain	15	1.47	5.1	0.008	0.0	19
2006	crude grain	16	0.14	0.3	0.014	0.0	0.8
2007	crude grain	16	0.18	0.5	0.008	0.0	1.7
2008	crude grain	13	0.99	3.7	0.008	0.0	13
2009	crude grain	9	4.51	14	0.008	0.0	40
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

OCHRATOXIN: Prevalence in oat grains and oat-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	20	0.4	1.3	0.2	0.0	5.8
	oat flakes (Norwegian)	20	0.3	0.3	0.2	0.0	0.9
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	food grain (Norwegian)	3	0.2	0.1	0.2	0.0	0.3
1994	food grain (Norwegian)	3	3.5	5.8	0.2	0.0	10
1995	food grain (Norwegian)	20	0.3	0.9	0.1	0.0	4.2
1996	food grain (Norwegian)	14	0.2	0.0	0.2	0.0	0.3
1997	food grain (Norwegian)	11	0.1	0.1	0.0	0.0	0.2
1998	food grain (Norwegian)	22	0.1	0.1	0.0	0.0	0.5
1999	infant porridge	10	0.03	0.02	0.02	0.00	0.08
2000	infant porridge	16	0.12	0.12	0.07	0.00	0.43
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	crude grain	17	0.16	0.4	0.020	0.0	1.5
2006	crude grain	16	0.21	0.5	0.008	0.0	0.8
2007	crude grain	16	0.07	0.2	0.008	0.0	0.5
2008	crude grain	16	0.13	0.5	0.008	0.0	1.9
2009	crude grain	12	0.20	0.03	0.008	0.0	2.1
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

OCHRATOXIN: Prevalence in spelt grains and spelt-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	infant porridge	2	0.04	0.02	0.04	0.03	0.06
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

OCHRATOXIN: Prevalence in rye grains and rye-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	food grain (Norwegian)	2	0.2	0.0	0.2	0.0	0.3
	food grain (imported)	18	0.2	0.0	0.2	0.0	0.3
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	food grain (imported)	4	0.4	0.4	0.3	0.0	0.9
1994	food grain (imported)	4	0.4	0.4	0.2	0.0	0.9
1995	food grain (imported)	4	0.7	1.1	0.2	0.0	2.5
1996	food grain (imported)	4	0.2	0.0	0.2	0.0	2.5
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

OCHRATOXIN: Prevalence in maize grains and maize-based foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	infant porridge	22	0.01	0.01	0.01	0.00	0.06
2000	infant porridge	4	0.01	0.00	0.01	0.00	0.01
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

OCHRATOXIN: Prevalence in mixed-cereals foods

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	infant porridge	9	0.45	0.44	0.23	0.08	1.32
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	-	-	-	-	-	-	-
2006	-	-	-	-	-	-	-
2007	-	-	-	-	-	-	-
2008	-	-	-	-	-	-	-
2009	-	-	-	-	-	-	-
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-

AFLATOXIN (B1 + B2): Prevalence in maize grains and maize-based feeds

year	sample	number of samples	mean [µg/kg]	SD [µg/kg]	median [µg/kg]	min [µg/kg]	max [µg/kg]
1990	-	-	-	-	-	-	-
1991	-	-	-	-	-	-	-
1992	-	-	-	-	-	-	-
1993	-	-	-	-	-	-	-
1994	-	-	-	-	-	-	-
1995	-	-	-	-	-	-	-
1996	-	-	-	-	-	-	-
1997	-	-	-	-	-	-	-
1998	-	-	-	-	-	-	-
1999	-	-	-	-	-	-	-
2000	-	-	-	-	-	-	-
2001	-	-	-	-	-	-	-
2002	-	-	-	-	-	-	-
2003	-	-	-	-	-	-	-
2004	-	-	-	-	-	-	-
2005	feed maize grain	19	0.7	1.1	0.13	0.0	4.3
	feed maize gluten	14	0.8	1.2	0.50	0.0	4.8
2006	feed maize grain	20	0.3	0.4	0.13	0.0	1.9
	feed maize gluten	20	1.5	1.1	1.42	0.0	3.9
2007	feed maize grain	12	0.2	0.2	0.18	0.0	1.4
	feed maize gluten	5	0.9	0.9	0.50	0.0	2.2
2008	feed maize grain	4	0.5	0.7	0.18	0.0	1.5
	feed maize gluten	6	1.0	0.6	1.21	0.0	1.6
2009	feed maize grain	14	0.2	0.1	0.18	0.0	0.4
	feed maize gluten	7	0.5	0.3	0.52	0.0	1.0
2010	-	-	-	-	-	-	-
2011	-	-	-	-	-	-	-