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Swedish University of Agricultural Sciences

Department of Molecular Sciences

Mycotoxins – an increasing problem?

- The effects of climate changes on *Fusarium* mould populations and the occurrence of fusarotoxins in Swedish cereals

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Mycotoxins – an increasing problem? – The effect of climate changes on *Fusarium* mould populations and the occurrence of fusarotoxins in Swedish cereals

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Abstract

Future changes in climate are expected to affect Sweden in the form of an up to 40 % increase in precipitation and a mean temperature that will be elevated by 3 - 4.5 °C by the end of the 21st century. These are conditions that presumably favour growth of *Fusarium* moulds. The infection of cereal crops by toxigenic species of this fungal genera can lead to the production of toxins with acute or chronic effects in humans and animals. This study was conducted to evaluate the potential effects of climate changes on toxigenic *Fusarium* fungi and their toxins in Swedish cereals, and how these effects might influence food safety. Different factors affecting toxin contamination were surveyed, and it was evaluated whether climatic changes could be a crucial factor. It was found that the occurrence of mycotoxins in Swedish cereals is already widespread, and that a trend of increasing toxin contents has been observed in Nordic cereals. Although the mycotoxin problem is complex and many factors, such as crop rotation and soil management, influence the toxin levels in cereals, it was found that weather conditions led to the highest increases in toxin concentrations. Thus, it was concluded that the future will bring increased problems with mycotoxins. The exposure of Swedish consumers to mycotoxins today does not exceed the TDI, and EFSA has expressed that no concern exists for the general population. However, in Sweden, the consumption of healthy foods such as nuts and vegetarian foods has increased, a trend which could result in an increased exposure to mycotoxins since these foodstuffs can be infected by moulds. In addition, Swedish consumers are exposed to mycotoxins through a variety of foods of different origin, and the import of cereal based products has increased. The expected global increase in toxin contamination of foods will thus also affect consumers and might lead to a co-exposure to several mycotoxins. This could imply risks for some consumer groups, especially since the adverse effects of exposure to a combination of toxins are not completely known. It was concluded that the mycotoxin problem should be carefully monitored.

Key words: *Fusarium* spp., fusarotoxins, climate change, Sweden, cereals, food safety

Sammanfattning

Framtida klimatförändringar förväntas påverka Sverige genom en ökning i nederbörd på upp till 40 % och en förhöjning av den genomsnittliga temperaturen med 3 – 4,5 °C vid slutet av seklet. Dessa förhållanden antas främja tillväxt av mögelsvampar inom släktet *Fusarium*. Då toxigena arter inom detta svampsläkte infekterar cerealier kan toxiner med akuta eller kroniska effekter hos djur och människor produceras. Denna studie utfördes med syftet att utvärdera klimatförändringarnas potentiella effekter på toxigena *Fusarium*-svampar och dessas toxiner i svenska cerealier, samt hur dessa effekter skulle kunna påverka livsmedelssäkerheten. Olika faktorer som påverkar toxinförekomst undersöktes, och det utvärderades huruvida förändringar i klimat skulle kunna vara en avgörande faktor. Undersökningen visade att mykotoxinförekomsten i svenska cerealier redan är utbredd, och att en trend av ökande toxinhalter har observerats i nordiska cerealier. Mykotoxinproblemet är komplext och många faktorer, såsom jordbearbetning och tillämpningen av växelbruk, påverkar toxinnivåerna i cerealier. Trots detta fann studien att väderförändringar ledde till de största ökningarna i mykotoxinhalter. Det konkluderades därför att framtiden kommer att föra med sig ökade problem med mykotoxiner. Den exponering för mykotoxiner som svenska konsumenterna utsätts för idag överskrider inte TDI, och EFSA har fastställt att ingen oro finns för den generella befolkningen. I Sverige har konsumtionen av hälsosamma produkter som nötter och vegetariska livsmedel ökat, en trend som skulle kunna innebära en ökad exponering för mykotoxiner eftersom dessa livsmedel kan infekteras av mögel. Svenska konsumenterna exponeras för mykotoxiner genom en mängd olika livsmedelsprodukter av olika ursprung, och importen av spannmålsbaserade produkter har ökat. Den förväntade globala ökningen i toxinkontaminering kommer därmed också att påverka konsumenterna, och kan leda till en samtidig exponering för flera mykotoxiner. Detta skulle kunna innebära en risk för vissa konsumentgrupper, särskilt eftersom de eventuella negativa effekterna av exponering för en kombination av mykotoxiner inte är kända. Mykotoxinproblemet bör därför övervakas noga.

Nyckelord: *Fusarium* spp., fusarotoxiner, klimatförändringar, Sverige, cerealier, livsmedelssäkerhet

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Abbreviations

15-AcDON Acetylated form of DON

3-AcDON Acetylated form of DON

BEA Beauvericin

DOM-1 Deepoxydeoxynivalenol

DON Deoxynivalenol

ENNs Enniatins

FHB *Fusarium* head blight

LOQ Limit of quantification

MON Moniliformin

NIV Nivalenol

OTA Ochratoxin A

TCTC Trichothecenes

TDI Tolerable daily intake

ZEA Zearalenone

1 Introduction

Global warming has been recognized in many studies, and quite certainly human activity plays a key role in it (Miljö- och energidepartementet, 2007). Its effects are widespread and have caused elevated temperatures in air and oceans as well as alterations in the global water cycle. The climate changes also include increased concentrations of the greenhouse gases carbon dioxide, methane and nitrous oxide in the atmosphere. Because of its dependence on climatic factors, agriculture is likely to be much affected by the global warming, leading to uncertainties in food safety as well as in food supply for the increasing world population (Medina *et al.*, 2017). One substantial concern is the expected increase in mycotoxin contamination of crops by filamentous fungi. Mycotoxins are low molecular weight, toxic secondary metabolites (Alshannaq & Yu, 2017). They are naturally occurring, and it is in principle impossible to completely avoid them. Consumption of foods contaminated with mycotoxins can cause acute or chronic symptoms. Several hundred mycotoxins have been identified, and six of these, including their metabolites and different forms, regularly contaminate food and feed. Many mycotoxins are stable against factors in food processing, such as heat and chemical and physical treatments, which makes elimination difficult. In the human food chain, cereals are the main source of mycotoxins (Medina *et al.*, 2017). Consumers can be exposed to these toxins either by direct ingestion of contaminated cereal foods or through intake of animal foods made of livestock that have been fed contaminated feed. Cereals are an important crop in Sweden. Wheat, barley, oats, rye and triticale are the most common cereals and these are cultivated on a third of the total agricultural land (Jordbruksverket, 2017b). Wheat is the most important cereal crop and is cultivated on approximately 47 % of the Swedish cereal acreage, followed by barley (31 %) and oats (16 %). Triticale is cultivated on around 2.7 % of the cereal acreage, and rye on less than 2 %.

The aim of this study was to evaluate the potential effects of climate changes on the population of toxigenic fungi and the occurrence of mycotoxin contamination of Swedish cereals, with focus on *Fusarium* fungi and mycotoxins. Another purpose was to shed light on how these changes could affect food safety for consumers. Since there are many factors that influence fungal infection and mycotoxin contamination of cereals, the aim was also to review these to investigate whether climate change could be a crucial factor.

2 Method

This literature study was based upon scientific articles found in the databases Web of Science, Scopus, Google Scholar, Food Science and Technology Abstracts and PubMed. Search words such as mycotoxins, deoxynivalenol, nivalenol, zearalenone, T2 and HT2, Swedish grains or cereals, Nordic grains or cereals, *Fusarium*, climate change and exposure were used in combination or separately to find suitable articles. Websites from authorities, like the Swedish National Food Agency, the Swedish Board of Agriculture, Statistics Sweden, EFSA and WHO have also been used for general information on mycotoxins and legislation.

3 The effects of global warming on the Swedish climate – what is the problem?

In Sweden, it is expected that the warming will be greater than the global average (Miljö- och energidepartementet, 2007). This warming will be largest during winter and affect especially north-eastern areas of the country. A yearly increase in precipitation is also likely to affect the whole country, apart from southern parts where the rainfall will probably decrease during summer. The heavy rains will also increase, as well as the most intensive rains. An increase of the latter has already been observed. The annual mean temperature could be elevated by 3 – 4.5 °C, and precipitation levels could increase by 40 % (Miraglia *et al.*, 2009). This would mean several things to agriculture. For example, it could be possible to cultivate new crops, and the growing season will be longer. However, new diseases could also appear.

4 Moulds and their toxins in Swedish cereals – what is the concern, and why are we worried?

There are several types of toxigenic fungi which can infect foods. Moulds that infect cereals are commonly divided into two groups; those that produce toxins in the field and those that produce toxins during storage of the grains (Lantbrukarnas riksförbund, 2013). Toxigenic fungi that most commonly infect cereals in the field in Sweden are species of the genus *Fusarium*, while *Penicillium* is the most common toxigenic storage fungus, responsible for the production of a variety of mycotoxins. The *Penicillium* species *P. verrucosum* is the main producer of ochratoxin A (OTA), one of the main mycotoxins, in temperate climates. *Fusarium* moulds also produce a battery of different toxins (Livsmedelsverket, 2017b). This study, as stated earlier, will focus on *Fusarium* fungi.

4.1 Trichothecenes

Some species of *Fusarium* moulds form trichothecenes (TCTC), which is a large group of toxins including deoxynivalenol (DON), nivalenol (NIV) and T2 and HT2 toxins (Livsmedelsverket, 2017c). The TCTC can be divided into two groups; type A TCTC and type B TCTC (Langseth & Rundberget, 1999). Trichothecenes are cyclic sesquiterpenes, and what separates the two types is the absence of a carbonyl group at C-8 on type A TCTC, which seems to make them more toxic. The type A group includes T2 and HT2 toxins, while DON and NIV belong to the type B group. HT2 toxin is the metabolized form of T2. The latter is the most toxic of the type A trichothecenes (Medina & Magan, 2011). All the mentioned toxins have been detected in cereals, and DON seems to be most common. The *Fusarium* species that produce trichothecenes also cause fusarium head

blight (FHB), which is a disease that destroys crops worldwide (Alshannaq & Yu, 2017).

DON is formed by the species *F. graminearum* and *F. culmorum* and is common in cereals grown in temperate climates (Livsmedelsverket, 2017b). DON affecting oats and wheat is the most important fusarium toxin in Sweden. The moulds producing DON can also form the toxin zearalenone (ZEA), especially in wheat but also in other cereals. This is however less common.

The species *F. langsethiae* is common in the colder climates of northern Europe (Livsmedelsverket, 2017b). It has become an important source of T2 and HT2 toxins (Medina & Magan, 2011). It mainly produces T2, which is the most toxic trichothecene, but can produce high levels of both toxins in oats, wheat and barley, and does so without causing symptoms on the grain. Toxin production has been shown at temperatures between 10 – 30 °C and might occur at even lower temperatures.

F. poae is considered the main producer of NIV in Swedish cereals (Fredlund *et al.*, 2013; Parikka *et al.*, 2012; Kosiak *et al.*, 2003). This species has also been correlated to NIV in Norwegian cereals.

The main reason for the toxicity of TCTC is the inhibition of protein synthesis by ribosomes which leads to the disruption of DNA and RNA synthesis (Alshannaq & Yu, 2017). TCTC can penetrate cell membranes and react with organelles, DNA and RNA. In humans who have been exposed to DON, the symptoms include abdominal pain, diarrhoea, nausea, vomiting, headache, dizziness and fever. A chronic exposure to DON could have effects on growth, the immune system and reproduction (Wallin *et al.*, 2013). NIV has been shown to inhibit the proliferation of human lymphocytes, and can thus be expected to have effects on the immune system (Luongo *et al.*, 2008). Toxicity studies have also reported effects on bone marrow, diarrhoea, damage to epithelial mucous membranes and changes in the gastrointestinal tract, etcetera (Nesic *et al.*, 2014). WHO has concluded that T2 toxin inhibits protein synthesis (WHO, 2002). It also affects the immune system, which is a primary target of the toxin, in several ways. Apart from

being both haematotoxic and immunotoxic, T2 toxin also has cytotoxic effects (Visconti *et al.*, 1991). Since T2 is metabolized to HT2 *in vivo*, the toxicity of these two toxins is not differentiated (WHO, 2002). The tolerable daily intake (TDI) is set for the two toxins in combination, or each of them separately, by EFSA to 0.02 µg/kg bw/day (Knutsen *et al.*, 2017c). For DON and its different forms, the group TDI is 1 µg/kg bw/day (Knutsen *et al.*, 2017a). A group TDI is set also for NIV and its different forms to 1.2 µg/kg bw/day (Knutsen *et al.*, 2017d).

4.2 Zearalenone

F. graminearum has been shown to be the most important producer of not only DON but also ZEA in cereal crops in Nordic countries (Lindblad *et al.*, 2013a). ZEA is structurally similar to oestrogen, and can bind competitively to oestrogen receptors, inducing estrogenic effects in both animals and humans (Alshannaq & Yu, 2017). ZEA has strong oestrogenic activity and has been shown to cause changes in female reproductive systems in several animal studies. Low temperature and high humidity favours formation of ZEA. Although the toxin can be eliminated partially by high temperatures, it is stable under normal cooking temperatures. The TDI for ZEA and its different forms is set by EFSA to 0.25 µg/kg bw per day (Knutsen *et al.*, 2017b).

4.3 Emerging mycotoxins in Sweden

There are some *Fusarium* toxins that have been more recently discovered, and that are considered to be an emerging problem (Logrieco & Moretti, 2008). These include moniliformin (MON), enniatins (ENNs) and beauvericins (BEA). In the Nordic countries, *F. avenaceum* seem to be the most important producer of MON and ENNs (Lindblad *et al.*, 2013b). BEA is also produced by this, and other *Fusarium* species, but *F. poae* is considered the most important producer of the toxin. *F. tricinctum* infects barley, oats and wheat and can produce MON, ENNs and BEA (Parikka *et al.*, 2012). *F. poae* infects oats, wheat and barley, but oats seem to be most susceptible. Studies have also found that it can produce the toxins fusarenon X and diacetylscirpenol. Enniatins and beauvericins are cyclic hexadepsipeptides (Logrieco & Moretti, 2008). Beauvericin is cytotoxic to mammals and

cause apoptosis of cells. Moniliformin also has toxic effects. More studies are needed to determine the toxicological effects of enniatins.

4.4 Masked/modified mycotoxins

The term masked mycotoxins has in some cases been used only for the compounds formed by plants (Freire & Sant'Ana, 2018). The term modified mycotoxins refer to all compounds derived from mycotoxins and is therefore the term used in this text.

The problem with modified mycotoxins has gained more attention from scientists as well as governments during the last years (Medina *et al.*, 2017). There are two types, or phases, of modifications¹. Phase I modifications are metabolites of the mother substance, whereas phase II modifications are *Fusarium* toxins that are conjugated with various compounds, which make them go undetected through some of the current analytical methods. These modified forms can be produced by the fungi, but can also arise due to defence mechanisms of an infected plant or during food processing (Freire & Sant'Ana, 2018). The toxic effects of these forms are not completely known. Some modified mycotoxins can be remodelled to the parent mycotoxin by gut microbiota and can in some cases become even more toxic than the parent compound due to higher bioavailability. The modified mycotoxins can occur simultaneously with the free form of the toxin, and sometimes at higher levels. This is cause for concern, since the conversion of modified mycotoxins to the free form could result in negative health effects. It also implies that an analysed food could be considered safe due to low concentrations of free mycotoxin, even though it contains high levels of modified mycotoxins which go undetected. Modified forms of DON have been observed in wheat and barley, and in smaller amounts in rye. Studies have indicated that lower incidence of FHB, as for example in resistant crops, results in higher levels of a modified form of DON. Little or no research is available on the effect climatic changes will have on these toxins (Medina *et al.*, 2017).

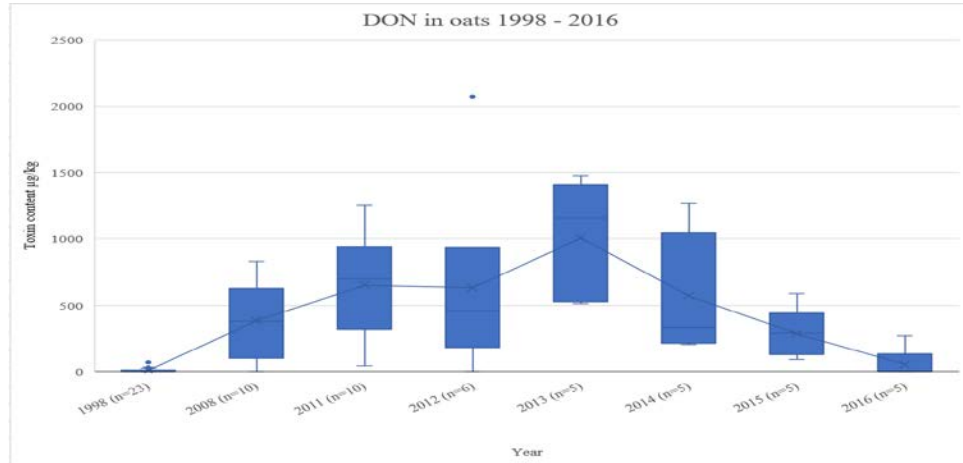
¹ Monica Olsen, National Food Agency

5 Occurrence of *Fusarium* toxins in Swedish cereals – how big is the problem?

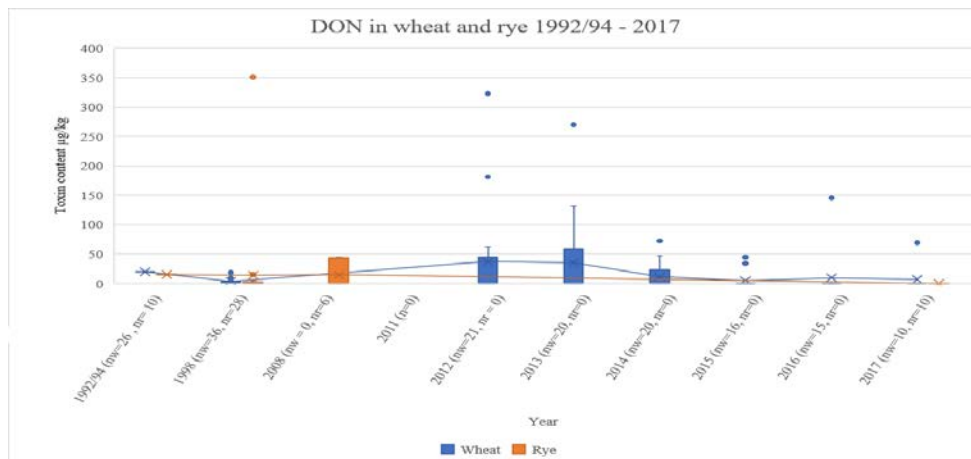
The Swedish Board of Agriculture has described oats, wheat and triticale as more susceptible to FHB, while barley and rye are less susceptible (Jordbruksverket, 2017a). In oats, presence of DON, T2 and HT2 toxins, NIV, ZEA, MON, ENNs and BEA has been reported (Fredlund *et al.*, 2013). Studies have shown that *Fusarium* moulds generally infect oats to a greater extent than wheat and barley. The species *F. poae* and *F. langsethiae* are more common in oats. Also *F. graminearum* and *F. avenaceum* have been detected. *F. poae* has been found to be the most prevalent species in a study on Swedish oats. In one study, ENNs and BEA were the most common contaminants whereas DON represented the highest toxin levels, and some samples even exceeded the legislative limits. The sample with the highest concentration of DON also contained ZEA above legislated limits. In one study on spring wheat, it was found that 90 % of samples contained DON and ENNs, about 50 % of samples contained MON whilst ZEA and NIV were less common (Fredlund, 2014).

The Swedish National Food Agency monitors mycotoxin levels in cereals and some data from previous years are available (unpublished). Data from detection of DON, ZEA, T2 and HT2 in wheat, oats and rye have shown that DON occurs in the highest levels in all cereals (Fig. 1), and that ZEA also frequently occurs in oats (Fig. 2). The data also confirm that oats are contaminated to a greater extent, regarding both contamination levels and incidence of contamination. Toxin levels fluctuate between years. T2 has not been detected in wheat and rye when samples of these crops were analysed and only a few samples of wheat contained HT2 (Fig. 3B). Oats were

significantly more contaminated by T2 and HT2 (Fig. 3B and 4). The contamination levels of all toxins in rye were low (Fig. 1B, 2B, 3B).

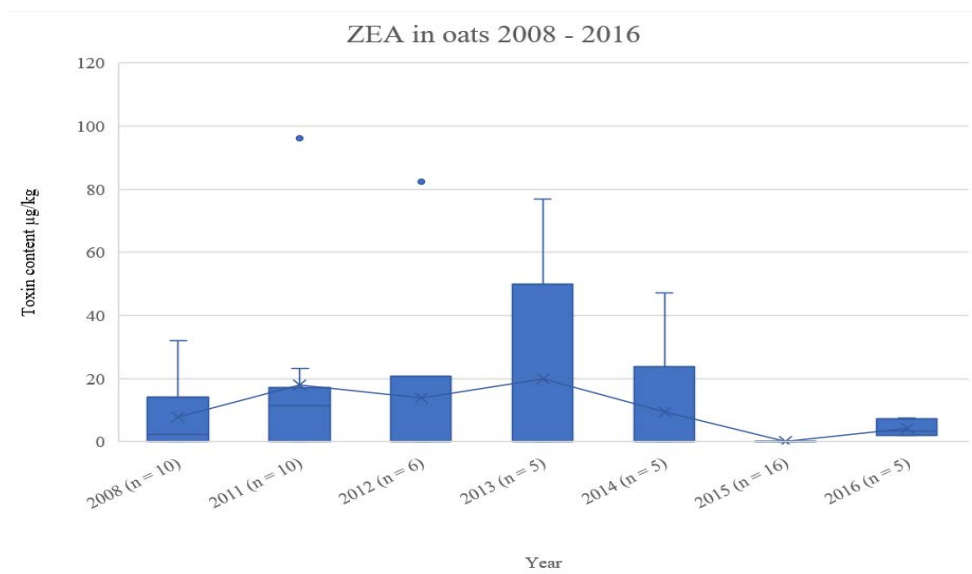


A.

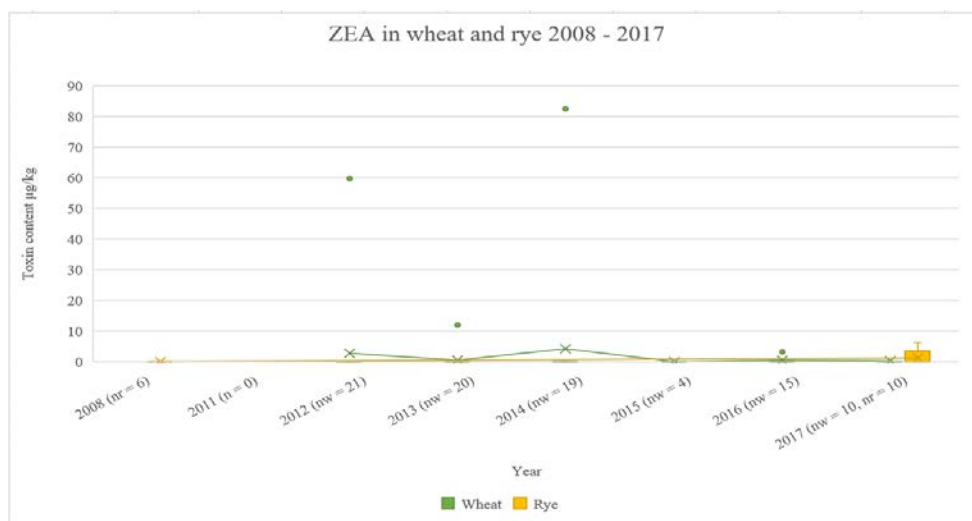


B.

Figure 1. DON levels in oat samples (A) and wheat and rye samples (B) taken by the Swedish National Food Agency from 1992-94/1998 through 2016/2017. The plots show where 75 % of the values lie in the boxes, with mean value indicated by X and median value by the line of the second quartile (Q2). Error bars show range of values. Outliers are marked by circles. LOQ varied in different sampling years as follows: for oats, <math><30\ \mu\text{g}/\text{kg}</math> in 2008, <math><60\ \mu\text{g}/\text{kg}</math> in 2011-15 and <math><100\ \mu\text{g}/\text{kg}</math> 2016, for wheat, <math><10-15\ \mu\text{g}/\text{kg}</math> in 1992/94, <math><30\ \mu\text{g}/\text{kg}</math> in 2012 – 2015, <math><100\ \mu\text{g}/\text{kg}</math> in 2016 and <math><50\ \mu\text{g}/\text{kg}</math> in 2017, and for rye <math><10-15\ \mu\text{g}/\text{kg}</math> in 1992/94, <math><30\ \mu\text{g}/\text{kg}</math> or <math><50\ \mu\text{g}/\text{kg}</math> in 2008 and 2017. No LOQ was available for samples from 1998. All samples below LOQ were set to zero. No values were available for wheat in 2008 and 2011, nor for rye in 2011 – 2016. The number of samples taken each year is indicated by n, nw and nr for oats, wheat and rye, respectively.

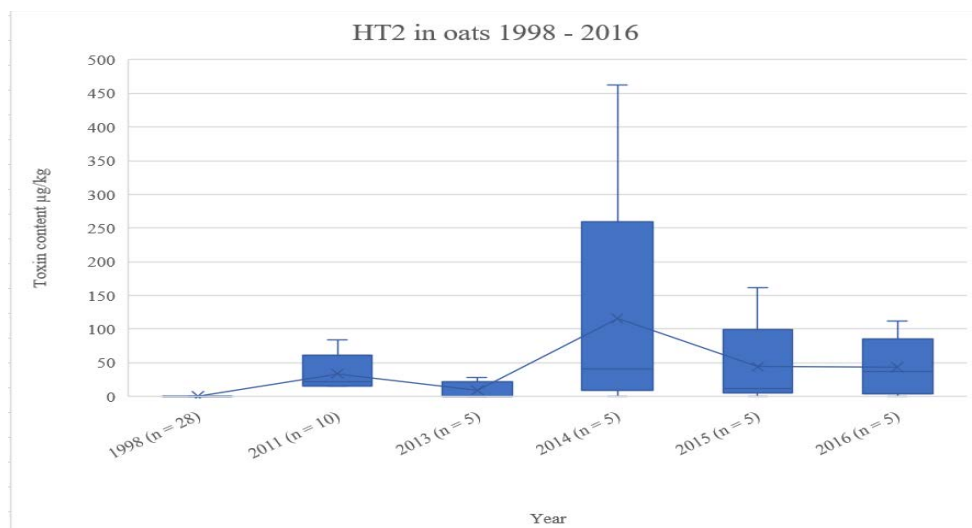


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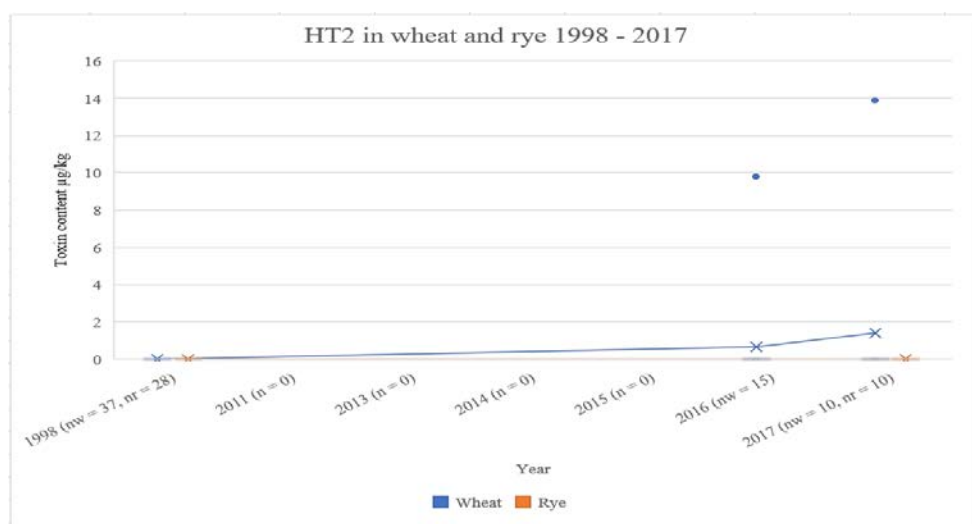


B.

Figure 2. ZEA levels in oat samples (A) and wheat and rye samples (B) taken by the Swedish National Food Agency from 2008 through 2016/2017. The plots show where 75 % of the values lie in the boxes, with mean value indicated by X and median value by the line of the second quartile (Q2). Error bars show range of values. Outliers are marked by circles. LOQ varied in different sampling years as follows: for oats, <math><4\ \mu\text{g}/\text{kg}</math> in 2008 and 2011 and <math><20\ \mu\text{g}/\text{kg}</math> in 2012-15 and <math><2</math> in 2016, for wheat, <math><4\ \mu\text{g}/\text{kg}</math> in 2012 – 15 and <math><2\ \mu\text{g}/\text{kg}</math> in 2016 and 2017 and for rye <math><4\ \mu\text{g}/\text{kg}</math> in 2008 and <math><2\ \mu\text{g}/\text{kg}</math> 2017. All samples below LOQ were set to zero. No values were available for wheat in 2008 and 2011, nor for rye in 2011 – 16. The number of samples taken each year is indicated by n, nw and nr for oats, wheat and rye, respectively.



A.



B.

Figure 3. HT2 levels in oat samples (A) and wheat and rye samples (B) taken by the Swedish National Food Agency from 1998 through 2016/2017. The plots show where 75 % of the values lie in the boxes, with mean value indicated by X and median value by the line of the second quartile (Q2). Error bars show range of values. Outliers are marked by circles. LOQ varied in different sampling years as follows: for oats, <15 µg/kg in 2011, <2 µg/kg in 2013-14, <100 µg/kg in 2015 and <5 µg/kg in 2016-17, for wheat, <5 µg/kg in 2016 and 2017 and for rye <5 µg/kg in 2016 and 2017. All samples below LOQ were set to zero. No LOQ was available for samples from 1998. No values were available for wheat in 2011 - 2015, nor for rye in 2011 - 2016. The number of samples taken each year is indicated by n, nw and nr for oats, wheat and rye, respectively.

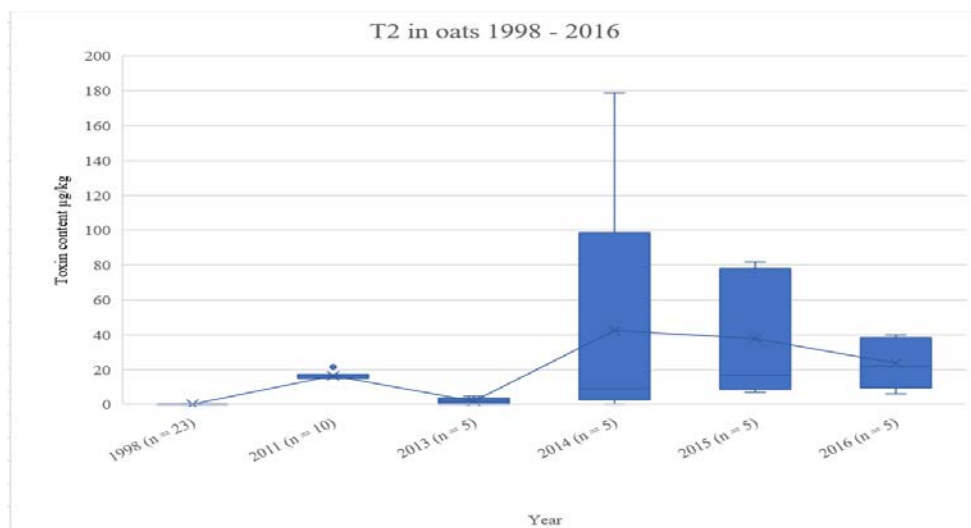


Figure 4. T2 levels in oat samples taken by the Swedish National Food Agency from 1998 through 2017. The plots show where 75 % of the values lie in the boxes, with mean value indicated by X and median value by the line of the second quartile (Q2). Error bars show range of values. Outliers are marked by circles. LOQ was <15 µg/kg in 2011, <1 µg/kg in 2013-15 and <5 µg/kg in 2016. All samples below LOQ were set to zero. No LOQ was available for samples from 1998. The number of samples taken each year is indicated by n.

6 Factors affecting mycotoxin contamination – what, other than climate, has an impact?

There are several factors apart from climate that affect fungal infection and mycotoxin contamination. Sometimes, these factors appear to be of more importance than for example weather conditions, and some variations in toxin contamination levels cannot be explained by climatic factors. It might be difficult to distinguish which factor leads to more contamination. Factors could also have a combined effect.

6.1 Agronomic factors

Different cereal species show different susceptibility to infection by *Fusarium*, and there are also differences between crop species in different countries (Edwards, 2004). There are varieties that are bred to be resistant to infection by *Fusarium*. Some studies have indicated that in wheat species bred to be resistant to FHB the total DON contamination decreased, but the ratio of a modified form of the toxin to DON increased (Lemmens *et al.*, 2016). Thus, the masked form does not appear to be affected to the same extent by plant breeding. A German study comparing different wheat cultivars found that the moderately-resistant species contained three times more DON than a species with good resistance (Obst *et al.*, 1997). However, the study did not analyse the presence of modified mycotoxins, which can be higher in resistant plants.

In Norway, it has been observed that oats seem to be the cereal most susceptible to DON contamination, something which is thought to be due to

the use of less careful cultivation methods in oat production than in the production of wheat and barley (Langseth & Elen, 1996). Oats are often grown in monoculture and in less favourable conditions causing the crop to be more susceptible to *Fusarium* infection (Langseth & Elen, 1997). It is often harvested later in the autumn which could increase the risk of fungal infection (Langseth & Elen, 1996). Apart from the crop itself, crop rotation also influences fungal infection, where maize as preceding crop to wheat has been shown to cause the highest contamination levels of DON. A study performed in Germany showed that the DON levels were at least four times higher in wheat with grain maize as pre-crop compared to when wheat or barley was used as preceding crop (Obst *et al.*, 1997). Another experiment showed that when wheat was grown with wheat as pre-crop and maize as pre-crop, the DON levels on average were 25 % and 50 % higher, respectively, compared to when wheat was grown with soybeans as pre-crop (Dill-Macky & Jones, 2000). Studies in Canada have shown similar results, where wheat grown with maize as pre-crop contained more than double the values of DON as when the pre-crop was soybean or wheat (Schaafsma & Hooker, 2007). In another study on wheat, it was shown that maize as pre-crop combined with a no-ploughing practice had more impact on toxin formation than warm and wet weather at flowering (Parikka *et al.*, 2012). Another factor which could be of importance is the level of *Fusarium* infection during the previous year, resulting in different levels of inoculum in the soil (Langseth & Elen, 1997).

Fusarium infection can be reduced by soil cultivation practices where residues of an infected crop are either destroyed, removed or buried, reducing the amount of *Fusarium* inoculum. *F. langsethiae* has been shown to infect grain to a greater extent in direct drilling practices compared to tilling (Medina & Magan, 2011). A German study found that when minimum tillage was used in combination with maize as pre-crop, the DON levels in wheat could show a 10-fold increase compared to when a ploughing practice was utilized (Obst *et al.*, 1997). Another study found that the levels of T2 and HT2 produced by *F. langsethiae* was 2.5 times higher in an oat cultivar where a minimum tillage practice was used combined with oat-straw incorporated in plots compared to when the straw was removed and ploughing was used (Imathiu *et al.*, 2017).

It has been reported that the incidence of *Fusarium* infection is lower in organically grown cereals than in those that are conventionally grown (Parikka *et al.*, 2012). Possible explanations for this are better practices of soil management and crop rotation in organic farming. One study showed that the *Fusarium* infestation was 5, 9 and 19 % lower in organically grown barley, oats and wheat, respectively, compared to the corresponding conventionally grown cereals (Bernhoft *et al.*, 2010). The study also found that the mean HT2 concentration in conventionally grown oats was 46 % higher than in oats grown in an organic practice. Mean DON levels were approximately 274 % higher in conventional oats compared to organic oats. Mean DON levels in conventional wheat were 98 % higher than in organic wheat.

Fertilizers could cause higher incidence or severity of *Fusarium* infection, for example by changing the rate of decomposition of residues or by causing a physiological stress on the plant which might render it more susceptible to infection (Miraglia *et al.*, 2009; Edwards, 2004). One study found that the levels of T2 and HT2 toxins were higher in oats for human consumption compared to those destined for animal feed. It was concluded that possible reasons for this were the utilization of more fertilizer and growth regulator in the production of oats for human consumption, both of which are factors that favour the growth of *Fusarium* and toxin contamination. A Canadian study found that the incidence of *Fusarium* infection on wheat, barley and triticale increased by up to 125 % when growth regulator or supplementary nitrogen was used (Martin *et al.*, 1991). It has been suggested that DON occurrence is more dependent on the intensity of nitrogen fertilization than ZEA (Tajnssek *et al.*, 2014).

There are some fungicides that are active against *Fusarium* and that can be used to reduce the levels of DON contamination (Edwards, 2004). However, there are also studies showing that mycotoxin production can be stimulated if too low concentrations of fungicides are used. In a Norwegian study it was found that some fungicides used to control other fungal diseases increased the *Fusarium* infection levels in spring wheat and barley by 9 – 19 %, and by similar numbers in oats (Henriksen & Elen, 2005). Studies of fungicide treatment in Norway showed that T2/HT2 producers were not affected by such a treatment (Parikka *et al.*, 2012). The Swedish Board of Agriculture states that even though the possibilities to completely prevent

fungal infection and FHB are limited, a fungicide treatment still has effects in some cases (Jordbruksverket, 2017a). The board recommends the use of fungicides when conditions are wet during bloom, especially in western Sweden, and if the crop is of a sensitive cultivar or if there are residues of a risk pre-crop on the field.

Weed control is important, since a high density of weeds has been shown to lead to increased infection by *Fusarium* (Edwards, 2004). One study found that FHB on wheat was on average twice as common in fields with large amounts of weeds (Teich & Nelson, 1984). This was thought to be due to an increased amount of water in these fields or nutrient stress caused by the weeds. Damage of the crop by insects can lead to the plant being more susceptible to infection (Edwards, 2004). Insects can also be carriers of the fungus. It has been shown that a combination of risk factors has a synergistic effect on the risk of DON contamination.

6.2 Regional and annual variations

The spread of toxins is different in different regions, but it also differs between years (Fredlund *et al.*, 2013). While DON is common and occurs globally, T2 and HT2 toxins seem to be most common in northern parts of Europe. In Sweden, surveys have shown that there are differences in the fungal communities in different regions, something which could explain regional differences in mycotoxin contamination of grain (Lindblad *et al.*, 2012). In a study by Fredlund *et al.* (2013) on Swedish oats, it was found that the western regions of the country had a higher prevalence of *F. graminearum* than other regions. *F. tricinctum* occurred in higher levels in the South than in other regions. DON and ZEA occurred in higher levels in oats from the West than from the East, and NIV was higher in samples from East and West compared to samples from the South. Samples from the South had higher levels of MON than all other regions.

According to another study on oats, the influence of year-to-year variations in weather on DON levels was low (Lindblad *et al.*, 2012). However, in wheat, this annual variation has been shown to explain a large part of the total variation. A Canadian study investigating DON levels in wheat over 4

years found that effects of year, and mainly the weather conditions, explained 48 % of the variation (Schaafsma *et al.*, 2001).

6.3 Co-occurrence of toxins

Cereals often contain a so-called species complex, composed of different *Fusarium* species, both non-pathogenic, pathogenic and opportunistic species, where the last two may produce mycotoxins (Logrieco & Moretti, 2008). The composition of species depends on agricultural practices and genetic factors, but also on climate and especially temperature and moisture. Grain that contains several different trichothecenes could be toxic to farm animals, even if the amounts of each individual toxin would not cause adverse effects (Langseth & Rundberget, 1999). In a study on cereals from northern Europe, a correlation was found between the levels of DON and ZEA in wheat, and between T2 and HT2 in oats (Van der Fels-Klerx *et al.*, 2012b). The acetylated forms of DON, 15-AcDON and 3-AcDON, occurs simultaneously with DON. It also appears that when DON is present in high concentrations, NIV concentrations are low and vice versa. The same negative correlation has been observed for T2 and NIV.

Studies have shown that DON in wheat co-occur with a number of other toxins, implying that consumers eating wheat products might be exposed to several different mycotoxins (Lindblad *et al.*, 2013b). The health effects of this are unclear.

7 Effect of climatic changes on mould populations and mycotoxin production – what can we expect in the future?

7.1 Effect of weather conditions

One prediction of climatic change is an increase in precipitation in late summer and autumn, something which favours *Fusarium* infection (Parikka *et al.*, 2012). *Fusarium* fungi are dependent on rainfall, and infection of grains occurs primarily when conditions are warm and moist during flowering (Parikka *et al.*, 2012). Different *Fusarium* species infect cereals in slightly different weather conditions, and different weather has different effects depending on the species and developmental stage of the crop. Table 1 shows some trends in the effects of different weather conditions on toxin contamination. It has been reported that mycotoxin formation by *F. graminearum* is favoured by warm and wet climates, while cool and moist environment around grain maturity favours infection by *F. avenaceum*. The wetter conditions could also lead to an increased infection by *F. culmorum*. Infection by this species is also expected to increase due to the predicted spring drought in Nordic areas. While *F. poae* infection is promoted by warm and dry conditions and increase in higher temperatures, precipitation has a negative effect on infection. Infection by *F. tricinctum* on the contrary increase with precipitation after flowering, but not at flowering. *F. langsethiae* infects in conditions of high humidity but can infect oats also when conditions are dry.

Table 1. Trends in the effect of weather conditions on contamination by different toxins summarized from Elen *et al.* (1997), Langseth & Elen (1997), Langseth & Rundberget (1999), Van der Fels-Klerx *et al.* (2012b, 2012c) and Parikka *et al.* (2012).

Toxin	Crop	Weather causing elevated toxin levels	Weather causing lower toxin levels
DON	Generally	Cold and wet Precipitation	Higher temperatures
	Wheat	High temperatures Humidity	
	Oats	High temperatures in April	Precipitation
	Barley	Warm and dry High temperatures and rainfall in April	
T2, HT2	Generally	Higher temperatures	
HT2	Wheat	Higher temperatures in June	
T2	Oats	Rainfall in May Higher temperatures at end of growing season	
ZEA	Wheat	Rainfall Higher temperatures	
	Oats	Rainfall	Higher temperatures
NIV	Wheat	High humidity in June	Rain Higher temperatures
	Oats		Rain Higher temperatures

Elevated levels of DON have been detected in cereals grown in cold and wet conditions (Parikka *et al.*, 2012; Langseth & Rundberget, 1999; Elen *et al.*, 1997; Langseth & Elen, 1997). In particular, excessive precipitation seems to cause more contamination by DON. Warmer weather appears to result in lower amounts of DON. However, one study found that dry and warm weather led to increased DON levels in barley (Langseth & Elen, 1997), indicating that climatic factors affect the interaction between grains and fungal species differently. In another study, DON concentrations were found to be higher in barley in conditions of high temperatures and rainfall during April. DON in oats increased with higher temperatures during the same month, but decreased with more precipitation. DON in wheat is increased by higher temperatures and humidity levels (Van der Fels-Klerx *et al.*, 2012b). Wheat and barley seem to be more sensitive to fungal infection in cases with elevated temperatures during blooming (Langseth & Elen, 1996).

In a Norwegian study comparing DON contamination in oats, wheat and barley in different years, it was found that the mean concentration from a year when there was little precipitation from anthesis to harvest was 35 µg DON/kg grain. In contrast, in a year with much precipitation around anthesis and a cold and wet growing season the mean levels reached 830 µg/kg (Langseth & Elen, 1997). A German survey compared the toxin content in wheat samples from nearby farms from six consecutive years (Muller *et al.*, 1997). When comparing two years, with 1.9 times more precipitation in one of the years, the mean concentration of DON was 1691.6 µg/kg grain in the rainy year whilst the mean concentration in the other year was 359.1 µg/kg. The mean concentration of ZEA was 178.0 µg/kg compared to 20.3 µg/kg; of T2 82.5 µg/kg compared to 10.0 µg/kg; and in HT2 the mean concentration in the rainy year was 8.8 µg/kg whereas no grain was infected in the other year. The opposite trend was observed in the NIV concentration, which had a mean of 9.0 µg/kg compared to 22.1 µg/kg.

In a study using model climate data it was predicted that the DON contamination levels might increase by up to three times in north-western Europe (van der Fels-Klerx *et al.*, 2012c). In most regions, these increased levels would not exceed the limit, though some regions where extreme levels of DON were predicted might be at risk of exceeding the limit.

A Norwegian study found that the level of T2 and HT2 contamination was higher in years when the weather had been warmer, suggesting that *F. langsethiae* prefers warmer weather, or has a higher toxin production in higher temperatures (Langseth & Rundberget, 1999). The presence of HT2 in wheat increased with higher temperatures in June, and T2 in oats increased with more rainfall during May and higher temperatures at the end of the growing season (Van der Fels-Klerx *et al.*, 2012b).

In wheat, the ZEA toxin levels have been shown to increase due to more rain and higher temperatures (Van der Fels-Klerx *et al.*, 2012b). However, higher temperatures resulted in lower concentrations of ZEA in oats while more rainfall was correlated to higher concentrations. Some experiments have shown an increase in ZEA contamination due to elevated CO₂ and higher temperatures (Bencze *et al.*, 2017).

Factors such as more rain and higher temperatures have been correlated to less contamination by NIV in oats and wheat, although a period of high humidity during June caused higher contamination levels in wheat (Van der Fels-Klerx *et al.*, 2012b).

The effect of weather differs in different crops in their different developmental stages and production of different toxins is not affected in the same way by the same weather conditions. Although some correlations seem to exist, it is difficult to identify definite trends.

7.2 Climate effects on agronomic factors

Climate change could have effects that indirectly affect mycotoxin contamination levels. The warmer climate will make it possible for cultivation of wheat in more northern areas (van der Fels-Klerx *et al.*, 2012c). The growing season will also be extended. Plant development will be affected, and anthesis and full maturity will occur earlier in the year.

It is expected that the cultivation of maize might increase in Nordic areas due to the warmer conditions and longer growing seasons (Parikka *et al.*, 2012). Maize residues in the field provide good substrate for *Fusarium* fungi, and

thus, the cultivation of this crop could be expected to lead to a higher incidence of fungal infections in both maize and other cereals. Maize as pre-crop has been correlated to higher DON levels in cereals. Cultivation of maize also leads to new problems with insects, which need to be controlled to avoid *Fusarium* infection – perhaps by an increased utilization of insecticides which traditionally have not been used in northern Europe. This would be a challenge to the aims of sustainable practices.

Climatic changes in the form of less snow cover during winter and increased precipitation during late summer and autumn will lead to changed demands on agricultural practices (Parikka *et al.*, 2012). Tillage and drilling will need to be minimized to avoid erosion and leaking, which leads to conditions favouring *F. graminearum* infection of grains.

It has been suggested that pests and diseases move towards the poles (Medina *et al.*, 2017). This could cause increased damage to crops, which in turn could lead to the plants being more susceptible to fungal infections. This would be a type of indirect effect of global warming on potential mycotoxin contamination.

7.3 Elevated CO₂ levels

Bencze *et al.* (2017) performed experiments comparing the effects of different CO₂ levels on *Fusarium* infestation and toxin production. The study used ambient CO₂ levels of 400 μmol mol⁻¹ corresponding to the current atmospheric CO₂ levels, and elevated CO₂ levels of 750 μmol mol⁻¹. The purpose was to reflect upcoming scenarios of elevated atmospheric CO₂ levels, which currently increase by 3 μmol mol⁻¹ per year. In experiments on the effect of elevated CO₂ levels on infection by *F. culmorum* on wheat compared to ambient levels, no significant change in fungal infection could be observed. However, the toxin contents were higher at higher CO₂ levels, suggesting that the toxin concentration is not necessarily correlated to the level of infection. Also, different varieties of wheat reacted differently to different experimental conditions. The levels of DON were higher in higher temperatures. ZEA contamination increased in elevated CO₂ levels. In one experimental setup, the concentration of DON was 350 000 ng/g in ambient CO₂ compared to 450 000 ng/g in elevated CO₂ in one of the wheat species.

The ZEA concentrations were 5 ng/g compared to 30 ng/g in the same species and experiment. The results show that the changes in temperature and CO₂ resulting from climate changes may favour infection by *Fusarium* and mycotoxin contamination of wheat.

7.4 A shift in species and increased contamination

The effects of climate change are expected to increase conditions favouring *Fusarium* infection and result in an increased risk of presence of mycotoxins in cereals in the field (Medina *et al.*, 2017). Mycotoxins are considered a major food safety hazard that is affected by the changing climate. In addition to the increased occurrence, a change in geographical distribution of toxigenic fungi is also likely, which in turn enhances the occurrence.

Since the growth of *Fusarium* moulds is related to climatic factors, future climate changes are expected to result in a higher prevalence of mould infections (Fredlund *et al.*, 2013). In the 1980s and 1990s, *F. culmorum* was the dominant species in Northern European countries, but since then, the occurrence of *F. graminearum* has increased (Lindblad *et al.*, 2012). Several studies have described a shift in dominance from *F. culmorum* to *F. graminearum* in northern Europe (Lindblad *et al.*, 2013a). Climate factors have been reported to be responsible for this change. An increased cultivation of maize, which is a major host for *F. graminearum*, and changes in tillage practice could also be causative (Fredlund *et al.*, 2008). Apart from producing DON, *F. graminearum* has also been shown to be the most important ZEA producer. One study found that the variation of DON contents in wheat was primarily caused by climate factors (Lindblad *et al.*, 2013a). These were more important than the variety of grain and the preceding crop. *F. graminearum* produces more DON than *F. culmorum*, and a shift in dominance to the first species could thus cause DON concentrations in cereals to increase (Van Der Fels-Klerx *et al.*, 2012a).

It is possible that the new conditions following climate change will be utilized differently by different *Fusarium* species, something which could lead to a change in populations of fungi (Parikka *et al.*, 2012). Changes in climate together with crop rotations with maize and small-grain cereals could also cause *Fusarium* species to spread (Bottalico & Perrone, 2002). A trend

of increasing DON levels has been observed in Norway and also partly in Sweden (Lindblad *et al.*, 2012). In one study viewing toxin levels in Nordic cereals from 1991 to 2009, an increase was observed in contamination level of 3-AcDON in oats, DON in barley and oats and of ZEA in wheat (Van der Fels-Klerx *et al.*, 2012b). However, a decrease was observed in HT2 and NIV contamination of oats.

In addition to the increase of *F. graminearum* infection, dry years might benefit *F. poae*, and *F. langsethiae* could also increase in dry conditions during spring and summer (Parikka *et al.*, 2012). The infection of oats by *F. langsethiae* and thereby the contamination with T2 and HT2 toxins has been reported to have increased in northern Europe during the last two decades.

8 Exposure of consumers to mycotoxins – is there need for concern?

In a study on exposure of Swedish adults to mycotoxins through food, it was found that 99 % of urine samples from the study population contained at least one mycotoxin metabolite, showing that exposure to mycotoxins is common (Wallin *et al.*, 2015). The most common toxins found were, in order of the amount found, DON, OTA and ZEA. A previous study also found that the exposure of Swedish consumers to DON is common (Wallin *et al.*, 2013). The deoxynivalenol metabolite deepoxydeoxynivalenol (DOM-1) and NIV were also detected, although only in samples containing DON (Wallin *et al.*, 2015). Simultaneous exposure to several mycotoxins was common and occurred in 69 % of the samples. This concurrent exposure could lead to the toxins causing adverse effects at lower concentrations than they would individually. The cooccurrence might also lead to interactions resulting in unpredictable toxic effects.

Intake of breakfast cereals, porridge and whole grain has been especially associated with high DON levels in urine (Wallin *et al.*, 2013). Raw cereals contain the highest levels of DON, but the consumption of processed cereals containing lower levels could also contribute to the exposure since such products are usually consumed in quite large quantities.

The levels of DON in samples rarely exceeded the TDI of 1 µg DON/kg bw (Wallin *et al.*, 2013). The possible negative health effects of DON on the Swedish population could thus be considered a minor problem. However, concern has been expressed for individuals consuming large amounts of cereals since they could be exposed to DON levels far above the TDI. The intake of DON in young children has been shown to be close to the TDI in a

study on mycotoxin intake in European countries (Schothorst & van Egmond, 2004), and a Norwegian study reported that the intake of DON often exceeded the TDI in young children (Sundheim *et al.*, 2017). This has been concluded also by EFSA, and is cause for concern (Knutsen *et al.*, 2017a). The population in general were however not in the risk zone of exposure to too high levels of toxin. Intake levels of NIV in European countries has been shown to be far below the TDI (Schothorst & van Egmond, 2004).

In a study by EFSA, it was concluded that the intake of ZEA in general does not cause concern (EFSA, 2011b). Only extreme cases where an individual consumes cereal from the same batch containing high levels of ZEA for several days could pose a possible risk for consuming too high levels of the toxin. Toddlers were exposed to the highest levels of ZEA. There were also data suggesting that vegetarians might be exposed to twice as much ZEA as the average population.

When studying unprocessed grains from a number of European countries, ZEA was found in approximately 15 % of the samples, with the highest concentration in wheat bran and corn products (EFSA, 2011b). The levels in unprocessed grains were much higher than in grains destined for human consumption. It is possible that the consumption of ZEA contaminated foods in combination with other foods containing compounds with oestrogenic activity, such as soy products, could result in either antagonistic or additive effects.

In studies on T2 and HT2 toxins, higher levels have been found in unprocessed products, and also in bran since the toxins are often attached to the outer part of the grain (EFSA, 2011a). Oats and oat products contained the highest levels of T2 and HT2. Toddlers were subjected to the highest exposure. The exposure was however below the TDI in all age groups, leading to the conclusion that T2 and HT2 toxins are of no health concern in EU-countries.

There are indications that the intake of TCTC has increased during the last 10 years (Wallin *et al.*, 2013).

9 Discussion

Mycotoxins in cereals pose a real threat to food safety due to their potential acute or chronic effects. Toxigenic fungi already occur in Swedish cereals, and during the last years a group of emerging mycotoxins as well as modified mycotoxins have been identified and are considered a problem which is not yet well studied. Data from the Swedish National Food Agency confirm that oats seem to be contaminated by toxins to a greater extent than other cereals, regarding both contamination levels and incidence of contamination (more samples contained levels above LOQ). However, this does not necessarily mean that our consumption of oats poses the biggest threat of mycotoxin exposure, or that the exposure could be significantly reduced by a reduced consumption of oats. Wheat is the main cereal crop in Sweden, and we consume significantly more wheat than oats (Karlsson, 2011). Thus, wheat is probably the major source of our mycotoxin exposure. During recent years (2015/16), a trend of decreasing levels of DON and ZEA have been observed in the data collected by the National Food Agency (Fig. 1 – 2). It is unclear what causes this, but it might be due to agronomic changes resulting from better communication to the farmers and more widespread application of management strategies for mycotoxins (e.g. timing of fungicides), as recommended in the Board of Agriculture guidelines which are revised and published yearly (personal communication). Levels of HT2 in wheat and rye have been higher during recent years. Van der Fels-Klerx et al. (2012b) surveyed mycotoxin contamination in northern Europe over a 20-year period and observed an increased occurrence of several toxins. It is therefore considered likely that mycotoxin contamination has increased in Sweden. The data presented in Fig. 1 – 4 are too few and collected over too short a timespan to show any robust trends. The values do however show how toxin contamination can fluctuate and how large the deviations can be,

pointing to the complexity of the problem, and they are to be viewed as a baseline against which possible changes in the future can be compared. It is thus difficult to draw any conclusions from the data presented in Figures 1 – 4.

Many factors apart from climate affect toxin contamination. However, some factors seem to be of greater importance and cause higher toxin levels. In Table 2, the effects of some different factors on toxin contamination levels are shown. Although several agronomic factors are of importance, especially soil management, it is the weather conditions that seem to have the largest effect on toxin levels and that cause the highest increases. It is evident from the table that a combination of factors, such as maize as pre-crop and minimum tillage, can lead to a manifold increase in toxin levels. It might be difficult to distinguish which factors have larger effects, since several factors are inevitably combined in the field. The mycotoxin contamination of cereals is complex, and though some toxins seem to increase due to one factor, it is not certain that the same development will be observed in other toxins. The production of different toxins is also affected in different ways by the same factor, and the causative factor is often difficult to identify. However, based on what has been found in the present study, it is concluded that the contamination by mycotoxins is complex and depend on many factors, but weather conditions seem to have the largest effects on toxin contamination, although results of more studies than those reviewed in the current study should be considered to state any definite trends. The importance of weather conditions also implies that the problem with mycotoxins will be more pronounced in the future due to climate changes. This has been stated also in several scientific studies.

Something that is important to address are the effects that future agriculture might have on toxin contamination. The extended cultivation of wheat might lead to a spread of *Fusarium* fungi to areas where they have not earlier been a problem. What could be considered to pose the biggest potential threat is however the cultivation of maize which might be possible in larger parts of Sweden due to climatic changes, since this crop has been correlated to higher DON levels in cereals.

The shift in dominance from *F. culmorum* to *F. graminearum* indicates that fungi indeed are affected by climate changes and that some species might be established in areas where they earlier did not occur. The occurrence of additional species could lead to an increased simultaneous exposure to several mycotoxins, which in turn could cause unpredictable adverse effects on consumers.

Table 2. Examples of the effects of different factors on toxin contamination.

	Cereal	Toxin	Factor (a)	Compared to (b)	Percentage increase in contamination of (a)*	Citation
Agronomic factors	Wheat	DON	Moderately-resistant species	Good resistance species	200 %	Obst <i>et al.</i> (1997)
			Grain maize as pre-crop	Wheat or barley as pre-crop	300 %	Obst <i>et al.</i> (1997)
			Maize as pre-crop	Soybeans as pre-crop	50 - 100%	Dill-Macky & Jones (2000), Shaafsma & Hooker (2007)
			Wheat as pre-crop	Soybeans as pre-crop	25 %	Dill-Macky & Jones (2000)
			Maize as pre-crop + minimum tillage	Ploughing	Up to 900 %	Obst <i>et al.</i> (1997)
			Conventionally grown	Organically grown	98 %	Bernhoft <i>et al.</i> (2010)
Oats	DON	Conventionally grown	Organically grown	274 %	Bernhoft <i>et al.</i> (2010)	
		HT2	Conventionally grown	Organically grown	46 %	Bernhoft <i>et al.</i> (2010)
		T2 and HT2	Minimum tillage + oat straw incorporated in plots	Ploughing + removal of oat straw	150 %	Imathiu <i>et al.</i> (2017)
Climate factors	Oats, wheat and barley	DON	Large amounts of precipitation around anthesis + cold and wet growing season	Small amounts of precipitation from anthesis to harvest	2271 %	Langseth & Elen (1997)
	Wheat	DON	Large amounts of precipitation	Approximately half as much precipitation	371 %	Muller <i>et al.</i> (1997)

	Elevated CO ₂	Ambient CO ₂	28 %	Bencze <i>et al.</i> (2017)
ZEA	Large amounts of precipitation	Approximately half as much precipitation	777 %	Muller <i>et al.</i> (1997)
	Elevated CO ₂	Ambient CO ₂	800 %	Bencze <i>et al.</i> (2017)
T2	Large amounts of precipitation	Approximately half as much precipitation	725 %	Muller <i>et al.</i> (1997)
NIV	Large amounts of precipitation	Approximately half as much precipitation	-246 %	Muller <i>et al.</i> (1997)

* As cited in the references, converted from *x*-fold to percentage when necessary, or calculated from cited mean values.

Swedish consumers are not only exposed to mycotoxins through domestic foods. The climate changes will have global effects, which means that changes in mycotoxin contamination and a probable increase is likely also in other countries. For example, increased contamination by DON and aflatoxin B1 is expected in some European regions (Medina *et al.*, 2017). About half of the Swedish food supply comes from imported products (Chamber Trade Sweden, 2015). Some primary import food commodities are, among others, coffee, nuts, cocoa and spices. These are foods that all can contain mycotoxins (Livsmedelsverket, 2017c). The import of cereal goods has increased (Statistics Sweden, 2016), and Chamber Trade Sweden (2015) reports that Swedish food consumption has been influenced by an awareness of health and environmental matters during the latest years. The consumption of nutritious products such as cereal grains and nuts has increased. Thus, Swedish consumers are likely exposed to mycotoxins through a variety of food commodities from several different countries. An increased interest in vegetarian food has also been identified (Nielsen, 2017). If this development of a shift to a more vegetarian diet continues, it could lead to an increased consumption of soy-based products. Since these contain phytoestrogens (Livsmedelsverket, 2017a), the combined effect with an increased intake of ZEA from cereal based products could mean that consumers would be at risk of a too high exposure to compounds with oestrogenic activity, leading to adverse effects. Additionally, vegetarian diets often contain legumes, which also can be contaminated by fusarotoxins, thus leading to consumer exposure (Embaby *et al.*, 2013). If these trends of

healthy and vegetarian eating continue to develop in the same direction, coupled with the effects of climate changes on toxins, there should be concern about the safety of consumers. Especial concern should be directed to young children and those who are high consumers of food products that are often contaminated with mycotoxins, since these groups have already been identified as in the risk zone of exceeding the TDI for several mycotoxins, and especially since the adverse effects of the co-occurrence of several mycotoxins are not completely known. It is also difficult to foresee the effects of a long term, chronic exposure to mycotoxins. Therefore, the mycotoxin problem should be carefully monitored. More studies should be performed on the combined effects of different climate factors, crops and fungi. A large part of the research on the subject is focused on DON and wheat. More research is needed on other grains and toxins, and perhaps especially on the emerging mycotoxins and the modified mycotoxins which are not yet well studied.

10 Conclusion

Contamination by mycotoxins is complex and depends on many factors. However, considering all the aforementioned, it can be expected that *Fusarium* toxin contamination of Swedish cereals will increase in the future due to climate effects in the form of larger amounts of precipitation, higher temperatures, a decreased use of tillage, an increase in damage of crops by insects and the cultivation of maize. All these factors have been shown to affect the population of toxigenic fungi and/or toxin levels in the grain. Climate changes seem to be a crucial factor to toxin contamination of cereals, since weather has been shown to have the largest effect on toxin levels. Furthermore, a shift in dominating species of *Fusarium* fungi has been associated to climate changes, also pointing to the impact of this factor.

The combination of increased consumption of imported vegetable-based foods, trends of healthy eating possibly leading to higher consumption of cereal grains and nuts, and the likely increase in mycotoxin contamination of many food stuffs is a cause for concern. The problem with mycotoxins should be taken seriously, and monitoring of mycotoxins in our cereals as well as the factors leading to fungal infection should be continued in order to maintain the health of consumers and to minimise future risks from increased exposure.

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References

- Alshannaq, A. & Yu, J.H. (2017). Occurrence, toxicity, and analysis of major mycotoxins in food. *International Journal of Environmental Research and Public Health*, 14(6).
- Bencze, S., Puskas, K., Vida, G., Karsai, I., Balla, K., Komaromi, J. & Veisz, O. (2017). Rising atmospheric CO₂ concentration may imply higher risk of Fusarium mycotoxin contamination of wheat grains. *Mycotoxin Research*, 33(3), pp. 229-236.
- Bernhoft, A., Clasen, P.E., Kristoffersen, A.B. & Torp, M. (2010). Less Fusarium infestation and mycotoxin contamination in organic than in conventional cereals. *Food Additives and Contaminants Part a-Chemistry Analysis Control Exposure & Risk Assessment*, 27(6), pp. 842-852.
- Bottalico, A. & Perrone, G. (2002). Toxigenic Fusarium species and Mycotoxins Associated with Head Blight in Small-Grain Cereals in Europe. *European Journal of Plant Pathology*, 108(7), pp. 611-624.
- Chamber Trade Sweden (2015). *Market report food - focus on the Swedish Market*.
- Dill-Macky, R. & Jones, R.K. (2000). The effect of previous crop residues and tillage on fusarium head blight of wheat. *Plant Disease*, 84(1), pp. 71-76.
- Edwards, S.G. (2004). Influence of agricultural practices on fusarium infection of cereals and subsequent contamination of grain by trichothecene mycotoxins. *Toxicology Letters*, 153(1), pp. 29-35.
- 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 feed. *EFSA Journal*, 9(12), p. 2481.
- EFSA (2011b). Scientific Opinion on the risks for public health related to the presence of zearalenone in food. *EFSA Journal*, 9(6), p. 2197.
- Elen, O., Langseth, W., Liu, W., Haug, G., Skjinner, H., Gullord, M. & Sundheim, L. (1997). The content of deoxynivalenol and occurrence of Fusarium spp. in cereals from field trials in Norway. *Cereal Research Communications*, 25(3 II), pp. 585-586.
- Embaby, E.M., Reda, M., Abdel-Wahhab, M.A., Omara, H. & Mokabel, A.M. (2013). Occurrence of toxigenic fungi and mycotoxins in some legume seeds. *International Journal of Agricultural Technology*, 9(1), pp. 151-164.
- Fredlund, E., Gidlund, A., Olsen, M., Borjesson, T., Spliid, N.H. & Simonsson, M. (2008). Method evaluation of Fusarium DNA extraction from mycelia and wheat for down-stream real-time PCR quantification and correlation to mycotoxin levels. *Journal of Microbiological Methods*, 73(1), pp. 33-40.

- Fredlund, E., Gidlund, A., Sulyok, M., Borjesson, T., Krška, R., Olsen, M. & Lindblad, M. (2013). Deoxynivalenol and other selected Fusarium toxins in Swedish oats - Occurrence and correlation to specific Fusarium species. *International Journal of Food Microbiology*, 167(2), pp. 276-283.
- Fredlund, E., Lindblad, M. (2014). *Fusariumsvampar och dess toxiner i svenskodlad vete och havre-rapport från kartläggningstudie 2009-2011* (Livsmedelsverkets rapportserie).
- Freire, L. & Sant'Ana, A.S. (2018). Modified mycotoxins: An updated review on their formation, detection, occurrence, and toxic effects. *Food and Chemical Toxicology*, 111, pp. 189-205.
- Henriksen, B. & Elen, O. (2005). Natural Fusarium Grain Infection Level in Wheat, Barley and Oat after Early Application of Fungicides and Herbicides. *Journal of Phytopathology*, 153(4), pp. 214-220.
- Imathiu, S.M., Ray, R.V., Back, M., Hare, M.C. & Edwards, S.G. (2017). Agronomic practices influence the infection of an oats cultivar with *Fusarium langsethiae*. *Acta Phytopathologica et Entomologica Hungarica*, 52(1), pp. 15-28.
- Jordbruksverket (2017a). *Fusarium*.
- Jordbruksverket (2017-10-17). *Use of agricultural land 2017*. Available at: http://www.jordbruksverket.se/webdav/files/SJV/Amnesomraden/Statistik,%20fakta/Arealer/JO10/JO10SM1703/JO10SM1703_ikortadrag.htm [2018-14-15].
- Karlsson, A.-M. (2011). Så här mycket mjöl konsumerar vi per år. *Jordbruket i siffror* [Blog]. <https://jordbruketsiffror.wordpress.com/2011/10/25/sa-har-mycket-mjol-konsumerar-vi-per-ar-2/>.
- Knutsen, H.K., Jan, A., Lars, B., Margherita, B., Beat, B., Sandra, C., Bruce, C., Michael, D., Bettina, G.K., Christer, H., Laurentius, H., Stefano, N.C., P, O.I., Annette, P., Martin, R., Alain-Claude, R., Tanja, S., Christiane, V., Günter, V., Heather, W., Sarah, D.S., Sundstøl, E.G., Peter, F., Jean-Marc, F., Yun, G.Y., Karsten, M., Hanspeter, N., Dominique, P.M., Ivonne, R., Hans, v.E., Andrea, A., Mari, E., Petra, G., Luisa, R.B., Bistra, B., Barbara, D., Athanasios, G., Nicklas, G., Mathijs, v.M. & Lutz, E. (2017a). Risks to human and animal health related to the presence of deoxynivalenol and its acetylated and modified forms in food and feed. *EFSA Journal*, 15(9), p. e04718.
- Knutsen, H.K., Jan, A., Lars, B., Margherita, B., Beat, B., Sandra, C., Bruce, C., Michael, D., Lutz, E., Bettina, G.K., Christer, H., Laurentius, H., Stefano, N.C., Annette, P., Martin, R., Alain-Claude, R., Tanja, S., Christiane, V., Günter, V., Heather, W., Chiara, D.A., Sven, D., Gunnar-Sundstøl, E., Andrea, A., Ruth, R.T. & P, O.I. (2017b). Risks for animal health related to the presence of zearalenone and its modified forms in feed. *EFSA Journal*, 15(7), p. e04851.
- Knutsen, H.K., Lars, B., Margherita, B., Beat, B., Sandra, C., Bruce, C., Michael, D., Lutz, E., Bettina, G.K., Christer, H., Laurentius, H., Stefano, N.C., Isabelle, O., Annette, P., Martin, R., Alain-Claude, R., Tanja, S., Christiane, V., Günter, V., Heather, W., Chiara, D.A., Arno, G., Manfred, M., Isabelle, O., Dominique, P.M., Marco, B., Hans, S. & Jan, A. (2017c). Appropriateness to set a group health based guidance value for T2 and HT2 toxin and its modified forms. *EFSA Journal*, 15(1), p. e04655.
- Knutsen, H.K., Lars, B., Margherita, B., Beat, B., Sandra, C., Bruce, C., Michael, D., Lutz, E., Bettina, G.K., Christer, H., Laurentius, H., Stefano, N.C., P, O.I., Annette, P., Martin, R., Alain-Claude, R., Tanja, S., Christiane, V., Günter, V., Heather, W., Chiara, D.A., C, G.A., Manfred, M., Dominique, P.M., Marco, B., Hans, S. & Jan, A. (2017d). Appropriateness to set a group health based guidance value for nivalenol and its modified forms. *EFSA Journal*, 15(4), p. e04751.
- Kosiak, B., Torp, M., Skjerve, E. & Thrane, U. (2003). *The Prevalence and Distribution of Fusarium species in Norwegian Cereals: a Survey*(53).

- Langseth, W. & Elen, O. (1996). Differences between barley, oats and wheat in the occurrence of deoxynivalenol and other trichothecenes in Norwegian grain. *Journal of Phytopathology*, 144(3), pp. 113-118.
- Langseth, W. & Elen, O. (1997). The occurrence of deoxynivalenol in Norwegian cereals—differences between years and districts, 1988–1996. *Acta Agriculturae Scandinavica Section B: Soil and Plant Science*, 47(3), pp. 176-184.
- Langseth, W. & Rundberget, T. (1999). The occurrence of HT-2 toxin and other trichothecenes in Norwegian cereals. *Mycopathologia*, 147(3), pp. 157-165.
- Lantbrukarnas riksförbund (2013). *Nationella branschriktlinjer för livsmedels- och fodersäkerhet vid produktion av spannmål, oljeväxter och trindsäd*.
- Lemmens, M., Steiner, B., Sulyok, M., Nicholson, P., Mesterhazy, A. & Buerstmayr, H. (2016). Masked mycotoxins: does breeding for enhanced Fusarium head blight resistance result in more deoxynivalenol-3-glucoside in new wheat varieties? *World Mycotoxin Journal*, 9(5), pp. 741-754.
- Lindblad, M., Borjesson, T., Hietaniemi, V. & Elen, O. (2012). Statistical analysis of agronomical factors and weather conditions influencing deoxynivalenol levels in oats in Scandinavia. *Food Addit Contam Part A Chem Anal Control Expo Risk Assess*, 29(10), pp. 1566-71.
- Lindblad, M., Gidlund, A., Sulyok, M., Borjesson, T., Krska, R., Olsen, M. & Fredlund, E. (2013a). Deoxynivalenol and other selected Fusarium toxins in Swedish wheat - Occurrence and correlation to specific Fusarium species. *International Journal of Food Microbiology*, 167(2), pp. 284-291.
- Lindblad, M., Gidlund, A., Sulyok, M., Börjesson, T., Krska, R., Olsen, M. & Fredlund, E. (2013b). Deoxynivalenol and other selected Fusarium toxins in Swedish wheat — Occurrence and correlation to specific Fusarium species. *International Journal of Food Microbiology*, 167(2), pp. 284-291.
- Livsmedelsverket (2017-09-20). *Hormonstörande ämnen*. Available at: https://www.livsmedelsverket.se/livsmedel-och-innehall/oonskade-amnen/hormonstörande-amnen?_t_id=1B2M2Y8AsgTpgAmY7PhCf%3d%3d&_t_q=fyto%3B6strogen&_t_tags=language%3asv%2csiteid%3a67f9c486-281d-4765-ba72-ba3914739e3b&_t_ip=130.238.98.89&_t_hit.id=Livs Common Model PageTypes ArticlePage/f89f0d2e-4896-4089-b1c7-fa2edd7b49a2_sv&_t_hit.pos=1 [06-05-2018].
- Livsmedelsverket (2017-08-31). *Mykotoxinbildande mögelsvampar*. Available at: <https://kontrollwiki.livsmedelsverket.se/artikel/160/mykotoxinbildande-mogelsvampar>.
- Livsmedelsverket (2017-08-18). *Mögelgifter*. Available at: <https://www.livsmedelsverket.se/livsmedel-och-innehall/oonskade-amnen/mogelgifter>.
- Logrieco, A.F. & Moretti, A. (2008). Between emerging and historical problems: An overview of the main toxigenic fungi and mycotoxin concerns in Europe. In: *Mycotoxins: Detection Methods, Management, Public Health and Agricultural Trade*, pp. 139-153. Available from: <https://www.scopus.com/inward/record.uri?eid=2-s2.0-84864551531&partnerID=40&md5=cc04d9cd52243b66cb76a6816dc70af7>.
- Luongo, D., De Luna, R., Russo, R. & Severino, L. (2008). Effects of four Fusarium toxins (fumonisin B1, α -zearalenol, nivalenol and deoxynivalenol) on porcine whole-blood cellular proliferation. *Toxicon*, 52(1), pp. 156-162.
- Martin, R.A., MacLeod, J.A. & Caldwell, C. (1991). Influences of production inputs on incidence of infection by Fusarium species on cereal seed. *Plant Dis.*, 75(8), pp. 784-788.
- Medina, A., Gonzalez-Jartin, J.M. & Sainz, M.J. (2017). Impact of global warming on mycotoxins. *Current Opinion in Food Science*, 18, pp. 76-81.

- Medina, A. & Magan, N. (2011). Temperature and water activity effects on production of T-2 and HT-2 by *Fusarium langsethiae* strains from north European countries. *Food Microbiology*, 28(3), pp. 392-398.
- Miljö- och energidepartementet (2007). *Sverige inför klimatförändringarna - hot och möjligheter*.
- Miraglia, M., Marvin, H.J.P., Kleter, G.A., Battilani, P., Brera, C., Coni, E., Cubadda, F., Croci, L., De Santis, B., Dekkers, S., Filippi, L., Hutjes, R.W.A., Noordam, M.Y., Pisante, M., Piva, G., Prandini, A., Toti, L., van den Born, G.J. & Vespermann, A. (2009). Climate change and food safety: An emerging issue with special focus on Europe. *Food and Chemical Toxicology*, 47(5), pp. 1009-1021.
- Muller, H.M., Reimann, J., Schumacher, U. & Schwadorf, K. (1997). Fusarium toxins in wheat harvested during six years in an area of southwest Germany. *Natural Toxins*, 5(1), pp. 24-30.
- Nesic, K., Ivanovic, S. & Nesic, V. (2014). Fusarial Toxins: Secondary Metabolites of Fusarium Fungi. In: Whitacre, D.M. (ed. *Reviews of Environmental Contamination and Toxicology Volume 228*. Cham: Springer International Publishing, pp. 101-120. Available from: https://doi.org/10.1007/978-3-319-01619-1_5.
- Nielsen (31-03-2017). *Försäljningen av vegetariskt ökar mest*. Available at: <http://www.niel-sen.com/se/sv/press-room/2017/foersaeljningen-av-vegetariskt-oeakar-mest.html> [06-05-2018].
- Obst, A., Lepschy-Von Gleissenthall, J. & Beck, R. (1997). On the etiology of Fusarium head blight of wheat in South Germany - Preceding crops, weather conditions for inoculum production and head infection, proneness of the crop to infection and mycotoxin production. *Cereal Research Communications*, 25(3 II), pp. 699-703.
- Parikka, P., Hakala, K. & Tiilikkala, K. (2012). Expected shifts in Fusarium species' composition on cereal grain in Northern Europe due to climatic change. *Food Additives and Contaminants: Part A -- Chemistry, Analysis, Control, Exposure and Risk Assessment*, 29(10), pp. 1543-1555.
- Schaafsma, A.W. & Hooker, D.C. (2007). Climatic models to predict occurrence of Fusarium toxins in wheat and maize. *International Journal of Food Microbiology*, 119(1), pp. 116-125.
- Schaafsma, A.W., Ilinic, L.T., Miller, J.D. & Hooker, D.C. (2001). Agronomic considerations for reducing deoxynivalenol in wheat grain. *Canadian Journal of Plant Pathology*, 23(3), pp. 279-285.
- Schothorst, R.C. & van Egmond, H.P. (2004). Report from SCOOP task 3.2.10 "collection of occurrence data of Fusarium toxins in food and assessment of dietary intake by the population of EU member states". Subtask: trichothecenes. *Toxicol Lett*, 153(1), pp. 133-43.
- Statistics Sweden (2016). *Jordbruksstatistisk sammanställning 2016 med data om livsmedel - tabeller*.
- Sundheim, L., Lillegaard, I.T., Faeste, C.K., Brantsaeter, A.L., Brodal, G. & Eriksen, G.S. (2017). Deoxynivalenol Exposure in Norway, Risk Assessments for Different Human Age Groups. *Toxins*, 9(2), p. 8.
- Tajnssek, L., Simcic, M. & Tajnssek, A. (2014). The impact of wheat production on the occurrence of mycotoxins DON (deoxynivalenol) and ZEA (zearalenone) on wheat grains (*Triticum aestivum* L.). *Acta Agriculturae Slovenica*, 103(2), pp. 245-262.
- Teich, A.H. & 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(1), pp. 11-13.
- Wallin, S., Gambacorta, L., Kotova, N., Warensjö Lemming, E., Nälsén, C., Solfrizzo, M. & Olsen, M. (2015). Biomonitoring of concurrent mycotoxin exposure among adults in Sweden through urinary multi-biomarker analysis. *Food and Chemical Toxicology*, 83, pp. 133-139.
- Wallin, S., Hardie, L.J., Kotova, N., Lemming, E.W., Nalsen, C., Ridefelt, P., Turner, P.C., White, K.L.M. & Olsen, M. (2013). Biomonitoring study of deoxynivalenol exposure and association with typical cereal consumption in Swedish adults. *World Mycotoxin Journal*, 6(4), pp. 439-448.

- Van Der Fels-Klerx, H.J., Goedhart, P.W., Elen, O., Börjesson, T., Hietaniemi, V. & Booij, C.J.H. (2012a). Modeling deoxynivalenol contamination of wheat in northwestern Europe for climate change assessments. *Journal of Food Protection*, 75(6), pp. 1099-1106.
- Van der Fels-Klerx, H.J., Klemsdal, S., Hietaniemi, V., Lindblad, M., Ioannou-Kakouri, E. & Van Asselt, E.D. (2012b). Mycotoxin contamination of cereal grain commodities in relation to climate in North West Europe. *Food Additives and Contaminants Part a-Chemistry Analysis Control Exposure & Risk Assessment*, 29(10), pp. 1581-1592.
- Van der Fels-Klerx, H.J., Olesen, J.E., Madsen, M.S. & Goedhart, P.W. (2012c). Climate change increases deoxynivalenol contamination of wheat in north-western Europe. *Food Addit Contam Part A Chem Anal Control Expo Risk Assess*, 29(10), pp. 1593-604.
- WHO (2002). Evaluation of certain mycotoxins in food. Fifty-sixth report of the Joint FAO/WHO Expert Committee on Food Additives. *World Health Organization technical report series*, 906, pp. i-viii, 1-62.
- Visconti, A., Minervini, F., Lucivero, G. & Gambatesa, V. (1991). CYTOTOXIC AND IMMUNOTOXIC EFFECTS OF FUSARIUM MYCOTOXINS USING A RAPID COLORIMETRIC BIOASSAY. *Mycopathologia*, 113(3), pp. 181-186.