



# The surveillance programme for feed and feed materials for terrestrial animals in Norway 2023 - Mycotoxins and fungi



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## Summary

In 2023, the surveillance programme for feed and feed materials focussed on mycotoxins of potential concern in oats, barley, rye and maize, as well as in compound feed for poultry and pigs. In addition, the weight of sclerotia from ergot (*Claviceps purpurea*) was determined in barley and rye.

Overall, the concentrations of T-2 + HT-2 toxins in oats were the only sample exceeding the recommended maximum levels for mycotoxins in feed or feed materials. Only one sample of oats had a content exceeding the recommended maximum levels for mycotoxins in feed.

In **oats**, the concentrations of deoxynivalenol (DON) was the highest since 2012 with no statistical significant differences between regions. One sample (2 %) had a sum of T-2+HT-2 concentrations exceeding the guidance level. The concentrations of T-2 and HT-2 in oats were low in 2023 compared to both previous years and compared to the recommended maximum levels. The levels of zearalenone (ZEA) were higher than the previous years, but still low compared to guidance values for feed materials for ZEA. No samples exceeded the relevant maximum levels in feed materials in 2023.

**Barley:** Trichothecenes were present in low levels in a high proportion of the samples, but in low concentrations compared to the guidance values for feed materials. ZEA was found in higher concentrations than in previous years, but no samples exceeded the guidance values for feed materials. Ergot was found in some barley samples at very low levels.

In **rye**, ergot was detected in all but one sample, several at high levels. Ergot alkaloids were also significantly elevated in several samples. Correlation between ergot and ergot alkaloids was statistically significant. As the samples were taken from the batches before the rye was cleansed for ergot, the results were not representative for rye at the market.

In **maize**, aflatoxins and ochratoxin A (OTA) were not detected in any of the six samples. . ZEA was found in most samples, but all were below the guidance level.

In compound feed for pig, DON was found in all samples, but in concentrations below the guidance values when analytical uncertainty was taken into consideration. Other trichothecenes, ZEA, fumonisins and OTA were not detected or detectable only at low concentrations. Ergot alkaloids were found in some samples at low levels.

DON and ZEA were present in low concentrations in all samples of poultry feed, but all concentrations were well below the recommended maximum guidelines.

## Sammenheng

Overvåkingsprogrammet for fôr og fôrmidler i 2023 omfattet mykotoksiner av potensiell betydning i havre, bygg, rug, mais, samt i kraftfôr til gris og fjørfe. I tillegg ble bygg og rug undersøkt for meldrøye (*Claviceps purpurea*),

Med unntak av en prøve av havre som inneholdt for høyt innhold av T-2+HT-2, ligger alle resultater innenfor fastsatte eller veiledende grenseverdier.

Konsentrasjonene av deoksynivalenol (DON) i havre var de høyeste siden 2012. Det ble ikke funnet noen geografiske forskjeller i innhold av DON i havre fra ulike geografiske områder. En prøve av havre inneholdt T-2 + HT-2 over gjeldende grenseverdi, men gjennomsnittlig var nivåene av T-2/HT-2 toksin var lavere enn i 2022 og under gjennomsnittskonsentrasjonene fra det siste tiåret. Nivåene av zearalenon (ZEA) var høyere enn tidligere år, men fortsatt langt under anbefalte maksimumsverdier for fôrmidler.

I bygg ble det påvist trichothecener i lave nivåer i en stor andel av prøvene, men nivåene var lave i forhold til anbefalte grenseverdier i fôrvarer. ZEA ble funnet i høyere konsentrasjoner enn tidligere, men ingen prøver hadde et innhold over de anbefalte grenseverdiene. Meldrøye ble funnet i noen prøver i lave nivåer.

I rug ble meldrøye påvist i alle prøver og i svært høye nivåer i flere prøver. Også meldrøyealkaloider var betydelig forhøyet i flere prøver. Korrelasjonen mellom meldrøye og meldrøyealkaloider var statistisk signifikant. Etersom prøvene ble samlet fra rugpartiene før rensing for meldrøye, er resultatene ikke representative for rug på markedet.

I mais ble det ikke påvist aflatoksiner eller OTA i noen av de seks prøvene. ZEA ble funnet i de fleste prøvene, alle under veiledende grense.

I kraftfôr til gris ble det funnet DON i de fleste prøvene, alle i ubetydelige konsentrasjoner. Andre trichothecener, samt ZEA, fumonisiner og OTA ble ikke, eller nesten ikke, påvist.

I kraftfôr til fjørfe ble det funnet lave mengder av trichothecener og zearalenone i alle prøvene. Meldrøyealkaloider ble funnet i lave konsentrasjoner i enkelte prøver. Okratoksin A ble ikke påvist. Ingen prøver av fjørfefôr overskred de anbefalte grenseverdiene for mykotoksiner i fjørfefôr.

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## Introduction

The annual surveillance programme on mycotoxins and microorganisms in feed and feed materials is a collaboration between the Norwegian Food Safety Authority (NFSA) and the Norwegian Veterinary Institute (NVI). NFSA decides the scope of the programme based on scientific advice from NVI, with NFSA responsible for collecting the samples, NVI for analysing and reporting of the results, and finally NFSA for result management. The agents for analyses usually consists of important mycotoxins and fungi (moulds, yeasts and ergot), in some years also selected bacteria. The programme gives good basis for assessments of feed quality, the impact of animal health and human exposure via animal products.

Fungi in cereals may be differentiated into field and storage fungi. Field fungi invade the seeds and produce toxins before harvest, and may affect the appearance and quality of seed or grain. Common field fungi in Norwegian cereal grain include mould species of the genera *Fusarium*, *Alternaria*, *Microdochium*, *Cladosporium*, *Acremonium*, *Epicoccum*, *Phoma* and more. In addition, *Claviceps purpurea* (ergot) is a field fungus [1]. Storage fungi usually occur in small amounts before harvest. However, under improper storage conditions, storage fungi can grow rapidly leading to significant problems. The most common storage fungi are *Penicillium*, *Aspergillus* and Mucorales. In addition, yeasts can occur in variable amounts among field and storage fungi [1].

*Fusarium* species are the most important mycotoxin-producing field fungi. They produce important mycotoxins such as the trichothecenes deoxynivalenol (DON), T-2 toxin (T-2) and HT-2 toxin (HT-2), as well as zearalenone (ZEA) and fumonisins [1].

Two decades of surveillance in Norwegian cereals have shown that DON can occur in high concentrations, particularly in oats and wheat. DON is hazardous to health if ingested by animals and humans [1]. Well-documented gastrointestinal disorders of DON exposure are reduced feed intake and stunted growth rate in pigs. In addition, DON impairs the immune system. T-2 and HT-2 are usually present in levels of concern only in oats and oat products. They have similar but potentially stronger toxic effects than DON, in causing gastrointestinal lesions as well as immune suppression [1]. Based on the limited available surveillance data, the oestrogenic mycotoxin ZEA produced by the same *Fusarium* species as DON, is usually present at insignificant levels in Norwegian cereals [1].

Data on the occurrence of the mycotoxins ergot alkaloids are of considerable interest in EU [2]. They show moderately acute neurotoxic effects, inhibition of blood circulation and interference of hormone levels. Ergot alkaloids are produced by *Claviceps purpurea* and are found mainly in rye, but may also occur in other cereal species - usually more in barley than oats [3-8].

Species of genera *Penicillium* and *Aspergillus* are the most important mycotoxin-producing storage fungi. *Penicillium* species generally grow and produce mycotoxins at lower temperatures than species of *Aspergillus*, and are therefore of main concern under the Norwegian storage conditions while *Aspergillus* species are more related to imported feed or feed materials [1].

Ochratoxin A (OTA) is an important mycotoxin produced by several species of both *Penicillium* and *Aspergillus*. The most prominent adverse effect of OTA in livestock is nephrotoxicity in pigs. It may also suppress the immune response and growth performance [1]. As far as we know, OTA has not caused problems for Norwegian husbandry. Nonetheless, active surveillance of OTA is important, particularly because of imported feed ingredients [1].

In addition, aflatoxins produced by some *Aspergillus* species may occur in imported feed ingredients [1]. These carcinogenic and liver toxic compounds must be kept at low levels to minimise human health risks via consumption of animal products as well as to ensure animal health. Aflatoxins in feed can result in, human exposure via dairy products since active metabolites are secreted into the milk.

## Aims

The aims of the programme on surveillance of feed and feed materials for terrestrial animals in Norway are to document compliance with the legislation on the occurrence of important mycotoxins and selected microorganisms, primarily fungi. The data are used to assess adverse animal health risks related to these agents in feed and to human exposure of transmissible agents via animal products.

## Materials and methods

In 2023, the surveillance programme for feed consisted of the following samples shown in Table 1.

Table 1. Samples in the surveillance programme for feed 2023.

Matrix	Planned	Sampled and analysed	Analyses
Oats	45	43	Trichothecenes, and zearalenone.
Barley	45	50	<i>Claviceps purpurea</i> , trichothecenes, zearalenone and ergot alkaloids.
Rye	20	12	<i>Claviceps purpurea</i> and ergot alkaloids.
Maize/maize products	15	6	Aflatoxins, ochratoxin A and zearalenone.
Complete compound feed for pigs	20	20	Trichothecenes, zearalenone, fumonisins, ergot alkaloids and ochratoxin A.
Complete feed for poultry	10	21	Trichothecenes, zearalenone, ergot alkaloids and ochratoxin A.

Oats, barley and rye from mills in grain production areas were sampled during autumn, representing the 2023 harvest. Batches of imported maize and maize products from third countries, compound feed for pigs and poultry from feed industries sampled throughout the year. To ensure representative samples, sampling procedures according to EU Regulation 152/2009 were followed.



## Chemical analysis of oats, barley, rye, compound feed for pigs and farm-mixed feed for pigs

A multi-mycotoxin liquid chromatography-tandem mass spectrometry (LC-MS/MS) method (ME05\_202) was used for the simultaneous determination of mycotoxins. ME05\_202 was developed in-house and validated according to the guidance document SANTE 11312/2021. Performance parameters assessed include linearity, selectivity, mean recovery, precision and limit of quantification (LOQ). According to the validation data, considerable matrix-effects, varying from 4 % to 157 %, were demonstrated for all mycotoxins across all five selected animal feed/feed products. In order to improve the accuracy of the method, stable-isotope labelled internal standards (IS) were introduced for eleven of the analysed mycotoxins including DON, its' related compounds 3-acetyl-DON (3-Ac-DON), 15-acetyl-DON (15-Ac-DON) and DON-3-glucoside (DON-3-G), as well as nivalenol (NIV), HT-2, T-2, FB1, FB2, ZEN and OTA. For quantitative analysis of ergot alkaloids, matrix-matched calibrations were prepared for each matrix. Recovery should be preferably be between 70-120%. Analyses results were corrected in cases when recoveries fall outside 70 - 120%, but not lower than 50% or above 140%. In cases when recoveries fall outside the range of 50-140%, samples were reanalysed. Statistics from proficiency tests provided for the national reference laboratories (NRLs) and appointed official control laboratories (OCLs) confirmed the applicability of this method. The accuracy of the method was assessed by determining recovery from spiking experiments and the extended measurement uncertainty ( $U'$ ) ( $U' = 2 \times u'$ , where  $u' = \text{rot}(u'_{\text{bias}}^2 + u'_{\text{precision}}^2)$ ), where  $u'_{\text{precision}} = \text{RSD} (\%)$  and  $u'_{\text{bias}}$  is the difference between the spiked and the measured concentration in percent (Table 1). Limit of quantitation (LOQs) is the lowest spiked concentration of an analyte in a tested sample which can be quantitatively determined with an acceptable level of precision and accuracy (Table 2).

**Table 1. Performance validation parameters for multi-analyte mycotoxin LC-MS/MS method (ME05\_202)**

Toxin	Recovery (%) and extended uncertainty** (%)									
	Rye		Barley		Oats		Pig feed		Mais	
Ergocornine*	110%	31%	116%	33%	116%	33%	104%	25%	-	-
Ergocristine*	109%	29%	115%	35%	104%	34%	104%	32%	-	-
$\alpha$ -Ergocryptine*	90%	37%	120%	41%	95%	41%	103%	35%	-	-
Ergonovine*	116%	34%	93%	15%	85%	33%	90%	23%	-	-
Ergosine*	114%	31%	116%	32%	112%	32%	99%	25%	-	-
Ergotamine*	115%	44%	118%	36%	115%	31%	101%	27%	-	-
15-ADON	117%	43%	106%	24%	115%	48%	110%	50%	-	-
3-ADON	114%	30%	85%	43%	90%	40%	86%	36%	-	-
DON	118%	64%	74%	67%	102%	12%	109%	23%	-	-
DON-3-G	103%	18%	100%	16%	110%	32%	120%	56%	-	-
Fumonisin B1	120%	40%	118%	36%	120%	42%	98%	29%	-	-
Fumonisin B2	104%	21%	111%	31%	103%	34%	102%	21%	-	-
HT-2	116%	35%	102%	39%	114%	57%	103%	44%	-	-
NIV	115%	32%	102%	16%	75%	51%	107%	15%	-	-
OTA	105%	35%	75%	54%	110%	29%	87%	31%	-	-
T-2	97%	44%	86%	42%	116%	63%	101%	33%	-	-
ZEN	107%	26%	71%	63%	88%	25%	92%	47%	113%	34%

\* sum of epimers (-inine and -ine)

\*\*extended uncertainty  $U' = 2u' = 2 \times \text{rot}(u'_{\text{bias}}^2 + u'_{\text{precision}}^2)$



Table 2. LOQ levels of toxins in different matrices

Toxin	LOQ ( $\mu\text{g}/\text{kg}$ )				
	Rye	Barley	Oats	Pig feed	Maize
Ergocornine*	50,0	12,5	10,0	25,0	-
Ergocristine*	50,0	12,5	10,0	25,0	-
$\alpha$ -Ergocryptin	25,0	12,5	5,0	25,0	-
Ergonovine*	25,0	12,5	5,0	25,0	-
Ergosine*	25,0	12,5	10,0	25,0	-
Ergotamine*	25,0	12,5	10,0	25,0	-
15-ADON	25,3	50,6	40,4	50,6	-
3-ADON	25,0	12,5	20,2	50,3	-
DON	25,1	25,1	60,0	100,3	-
DON-3-G	50,2	25,1	20,1	25,1	-
Fumonisin B1	80,5	80,5	160,6	100,4	-
Fumonisin B2	81,1	81,1	40,6	101,4	-
HT-2	50,0	25,0	10,0	50,1	-
NIV	200,0	100,0	161,1	201,1	-
OTA	25,1	25,1	20,1	50,2	-
T-2	25,0	12,5	5,05	60,2	-
ZEN	12,5	12,5	10,0	50,2	40,28

\* sum of epimers (-inine and -ine)

Samples were grinded to fine powder and a subsample of 2.5 g ( $\pm 0.5$  %) was weighed in. The extraction methodology was based on a two-step extraction (MeCN:H<sub>2</sub>O:HCOOH, 80:19.9:0.1, v/v/v and MeCN:H<sub>2</sub>O:HCOOH, 20:79.9:0.1, v/v/v) in order to improve the extraction efficiency with respect to polar and non-polar compounds. Extracts were centrifuged, filtered and were ready for instrumental analysis.

The LC-MS/MS analyses were performed on an Agilent Triple Quadrupole LC-MS system (1290-6470), equipped with an AJS electrospray ionization (ESI) while the Agilent MassHunter workstation software was used for data acquisition and quantitative analysis. 2  $\mu\text{L}$  of sample extract was injected into the LC system and analytes were separated on a Kinetex F5 100 Å column (100 x 2.1 mm), equipped with a precolumn, under a constant flow of 0,25 mL/min. Gradient elution was performed with a 5 mM ammonium acetate/1% acetic acid aqueous mobile phase and methanol to achieve optimal separation. Due to differences in the nature of each compound, the Triple Quadrupole was operated in both positive and negative ionization mode for optimal sensitivity. Identification of target mycotoxins was performed using three compound specific MRM transitions.

## Chemical analysis of aflatoxins and ochratoxin A in maize

In order to obtain the required sensitivities, a different multi-mycotoxin LC-MS/MS method (ME05\_203), dedicated to the determination of aflatoxins and ochratoxin A (OTA), was developed in house. ME05\_203 was validated according to the guidance documents SANTE/11312/2021 and NS-EN 17641:2022.

Performance parameters assessed include linearity, selectivity, mean recovery, precision and limit of quantification (LOQ). Based on the validation data, considerable matrix-effects were observed for both aflatoxins and OTA. To correct for matrix-effects and to compensate for losses during sample preparation, stable-isotopically labelled analogues were added to the calibration standards and to each sample before extraction. Statistics from proficiency tests provided for the national reference laboratories (NRLs) and appointed official control laboratories (OCLs) confirmed the applicability of this approach.

The accuracy of the method was assessed by determining recovery from spiking experiments and the extended measurement uncertainty ( $U'$ ) ( $U' = 2 \times u'$ , where  $u' = \sqrt{u'^2_{\text{bias}} + u'^2_{\text{precision}}}$ ), where  $u'_{\text{precision}} = \text{RSD} (\%)$  and  $u'_{\text{bias}}$  is the difference between the spiked and the measured concentration in percent (Table 3). Limit of quantitation (LOQs) is the lowest spiked concentration of an analyte in a tested sample which can be quantitatively determined with an acceptable level of precision and accuracy.

**Table 3. Performance validation parameters for multi-analyte mycotoxin LC-MS/MS method (ME05\_203)**

Toxin	LOQ ( $\mu\text{g}/\text{kg}$ )	Recovery (%)		Extended uncertainty* (%)
		Maize		
AFB1	0,156	119%		50 %
AFB2	0,313	105%		14 %
AFG1	0,313	81%		42 %
AFG2	0,313	86%		32 %
OTA	1,250	111%		24 %

\*extended uncertainty  $U' = 2u' = 2 \sqrt{u'^2_{\text{bias}} + u'^2_{\text{precision}}}$

Samples were grinded to fine powder and a subsample of 2 g ( $\pm 0.5 \%$ ) was weighed in. The subsamples were subjected to extraction procedure described in EN17641:2000 with minor modifications.

The LC-MS/MS analyses were performed on an Agilent Triple Quadrupole LC-MS system (1290-6470), equipped with an AJS electrospray ionization (ESI) while the Agilent MassHunter workstation software was used for data acquisition and quantitative analysis. 1  $\mu\text{L}$  of sample extract was injected into the LC system. Analytes were separated on a EVO C18 100 Å column (100 x 1.0 mm), equipped with a precolumn, under a constant flow of 0,1 mL/min. Gradient elution was performed with a 5 mM ammonium formate/0.1 % formic acid aqueous mobile phase and 98 % methanol to achieve optimal separation. Triple Quadrupole was operated in positive ionization mode and identification of target mycotoxins was performed using three compound specific MRM transitions.

## Quantitative determination of *Claviceps purpurea* in barley and rye

*Claviceps purpurea* sclerotia in grams per kg cereal were calculated according to the method described by Vrålstad *et al.* [8]. The weighed sample was spread over a large light surface for visual inspection. Detected sclerotia of *C. purpurea* were picked out and weighed separately.

## Statistical analysis

Descriptive statistics followed by One-way Anova were used to determine significance in statistical differences between groups (e.g. geographical regions) for variables that were measured quantitatively. Box-plots provide visual analysis of differences across geographical regions. To investigate possible linear correlation between two variables in the same feed type, scatter plots and Pearson correlations with p-values were determined.

According to the regulations, the levels of not quantifiable ergot alkaloids should be set to 0 when comparing the  $\Sigma$  Ergot alkaloids with the MRL. We used this approach for ergot alkaloids for all calculations. For other mycotoxins, half of the lowest quantified levels for undetected toxins specific to a variable were used for calculation of statistical descriptors when levels were not detectable.

## Results and discussion

### Cereals

#### *Mycotoxins in oats*

In oats in 2023, DON was detectable in 80 % of the samples, with a mean concentration of 915 µg/kg, DON was detected at the highest concentrations since 2012 (Table 3; Figures 1, 2). Still, all samples had DON levels below the limit for DON in feed materials recommended by EU and Norway (8000 µg/kg) [10, 11].

The DON-related compounds included in the analysis of oats were the acetylated precursor compounds (3-Ac-DON and 15-Ac-DON) and DON-3-glucoside (DON-3-G). The latter is a plant metabolite of the mycotoxin. DON-3-G was detectable in 98 % of the samples. In samples collected in 2023 also 3-Ac-DON was detected in all but one sample while 15-Ac-DON was not detected in any sample. DON and DON-3-G were significantly positively correlated (Figure 3) and the correlation was slightly higher than in 2022 ( $r = 0.84$  vs.  $r=0.73$  in 2022, both with a  $p<0.0001$ ). There was no significant differences in DON content nor DON-3-G between regions.

**Table 3.** Concentrations (µg/kg) of deoxynivalenol (DON), 3-acetyl-DON (3-Ac-DON), 15-acetyl-DON (15-Ac-DON), DON-3-glucoside (DON-3-G), T-2 and HT-2 toxin and the sum of T-2/HT-2, nivalenol (NIV), zearalenone (ZEA), and sum ergot alkaloids ( $\Sigma$ Erg alk) in oats (N = 45) sampled in Norway in 2023.

	DON	3-Ac-DON	15-Ac-DON	DON-3-G	T-2	HT-2	T-2+HT-2	NIV	ZEA
Mean	915	134	<40	169	20	68	88	< 161	122
Median	697	72	<40	114	6	34	41	<161	20
Minimum	155	<20	<40	< 20	<5	<10	<15	<161	< 10
Maximum	4084	1062	<40	654	266	966	1232	334	2281**
SD*	824	192	1	148	41	148	189	76	362
% samples >dl*	100	98	0	100	70	84		42	86
% samples >gv*	0						1		0**

\* SD = Standard Deviation, >dl = above detectable limits, >gv = above guidance values for feed ingredients taking the uncertainty into consideration. Empty means no guidance value are set.

\*\* Below the guidance value when uncertainty taken into consideration.

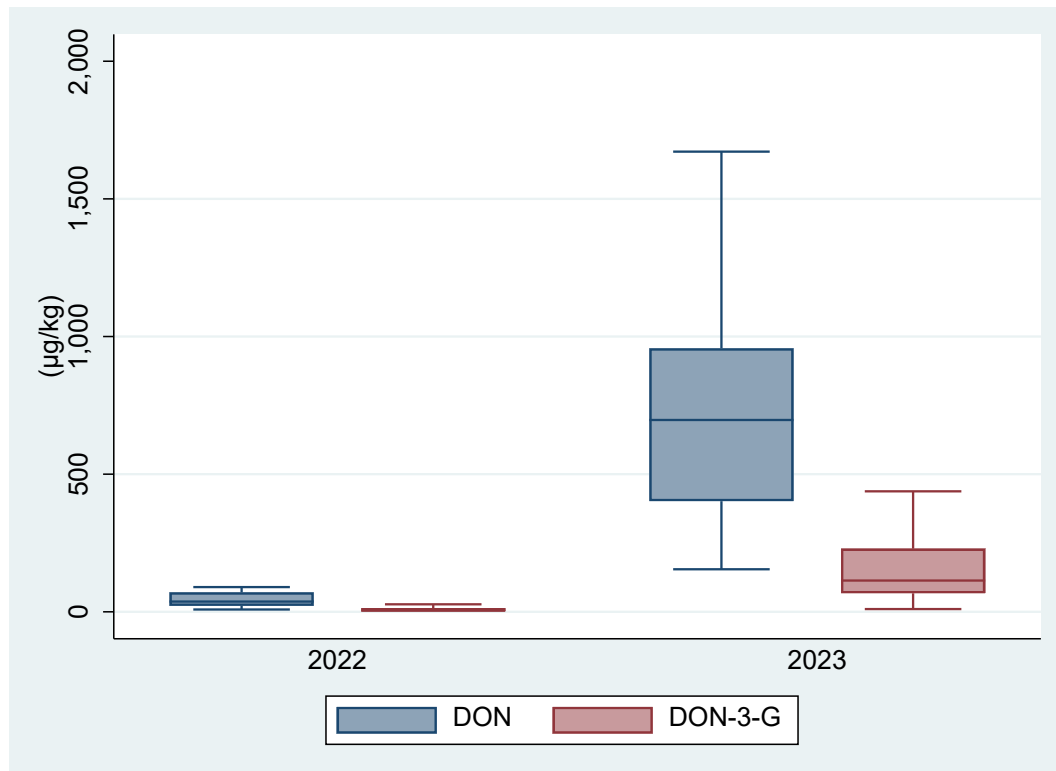


Figure 1. Concentrations of DON and DON-3 glucoside in samples from 2023 (mean values of 915 and 169 µg/kg) were significantly higher than in 2022 (83 and < 25 µg/kg).

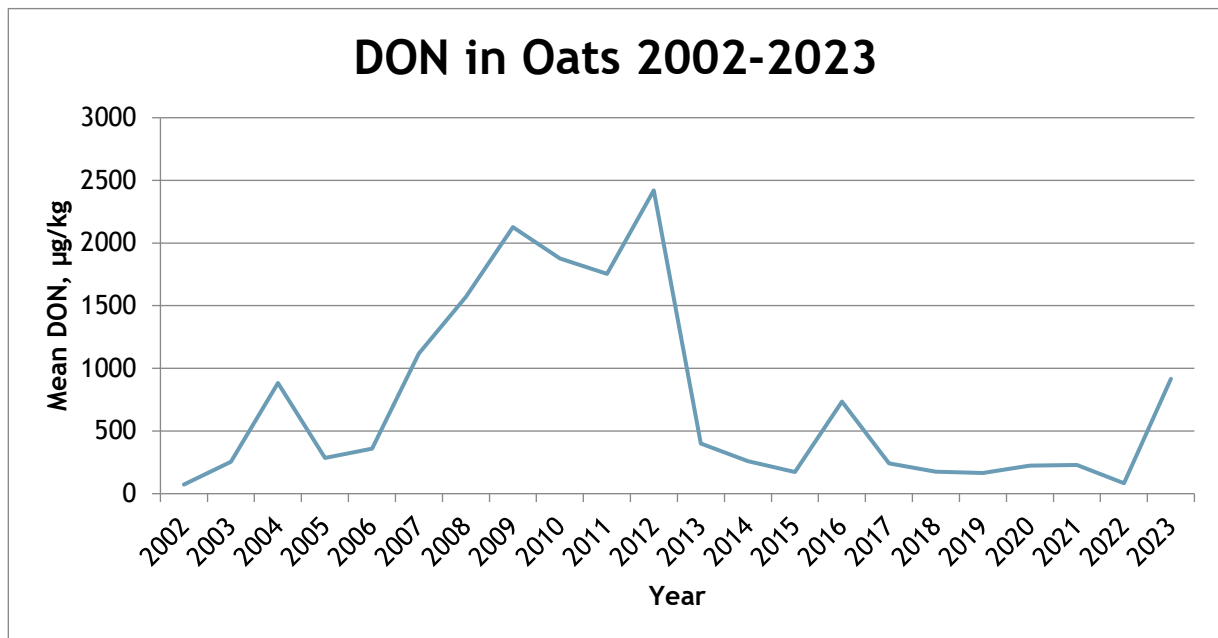
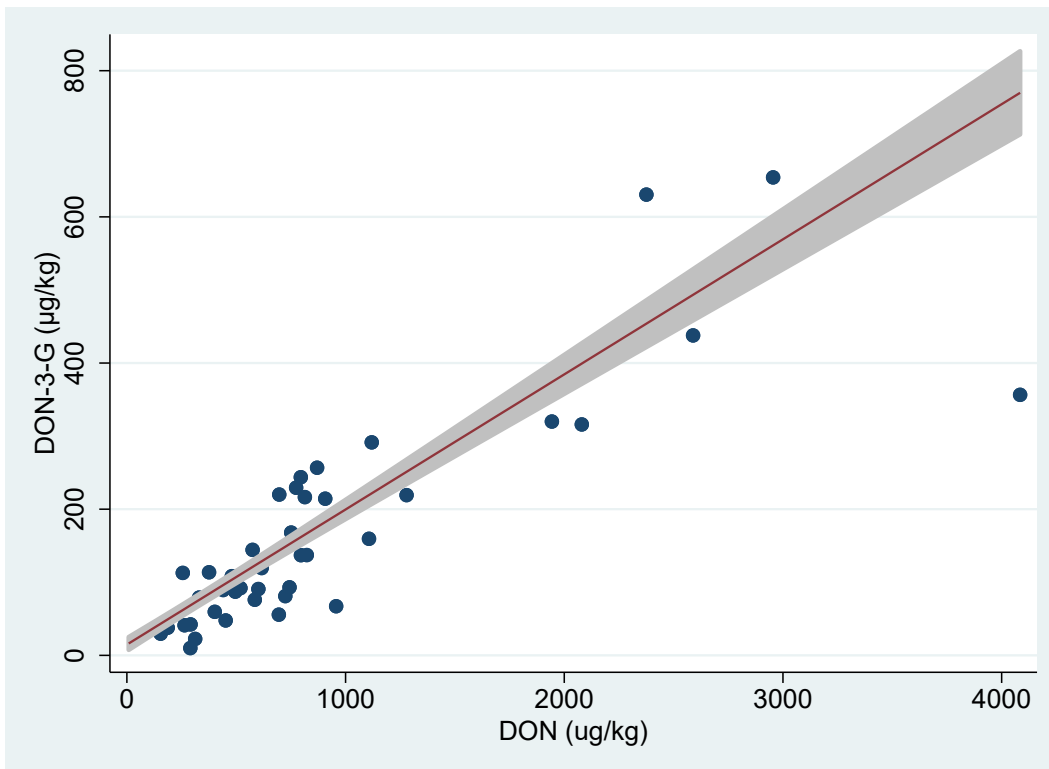


Figure 2. Mean concentration of deoxynivalenol (DON) in 30-60 samples of oats per year in the Norwegian surveillance programme for feed.



*Figure 3. The Pearson correlation between deoxynivalenol (DON) and DON-3-glucoside (DON-3-G) in oats (N=43) 2023 was  $r=0.84$ ,  $p<0.0001$ . A regression line with 95 % confidence interval fitted to the points allows predictions of levels of DON-3-G given the level of DON detected and vice versa.*

T-2 and HT-2 were detected in 70 and 84 % of the oat samples respectively. One sample (2 %) had a sum of T-2+HT-2 concentrations exceeding the guidance level of 500  $\mu\text{g}/\text{kg}$  in EU and Norway [9, 11] (Table 3). The mean concentration of T-2+HT-2 was below the average of corresponding levels from the last decade (Figure 4). In line with previous years, the levels of T-2 and HT-2 appear to be low in years with increased levels of DON. T-2 and HT-2 were highly correlated in oats (Figure 5). Despite the similar correlations between T-2 and HT-2 toxins, the relationship between the two toxins was different in 2023, compared to the previous year. For 2022, it was concluded that the concentration of HT-2 were about twice that of T-2, which was similar to previous years [3-8, 13]. In 2023 the HT-2 concentrations were on average about 5-fold higher than the T-2 concentrations.

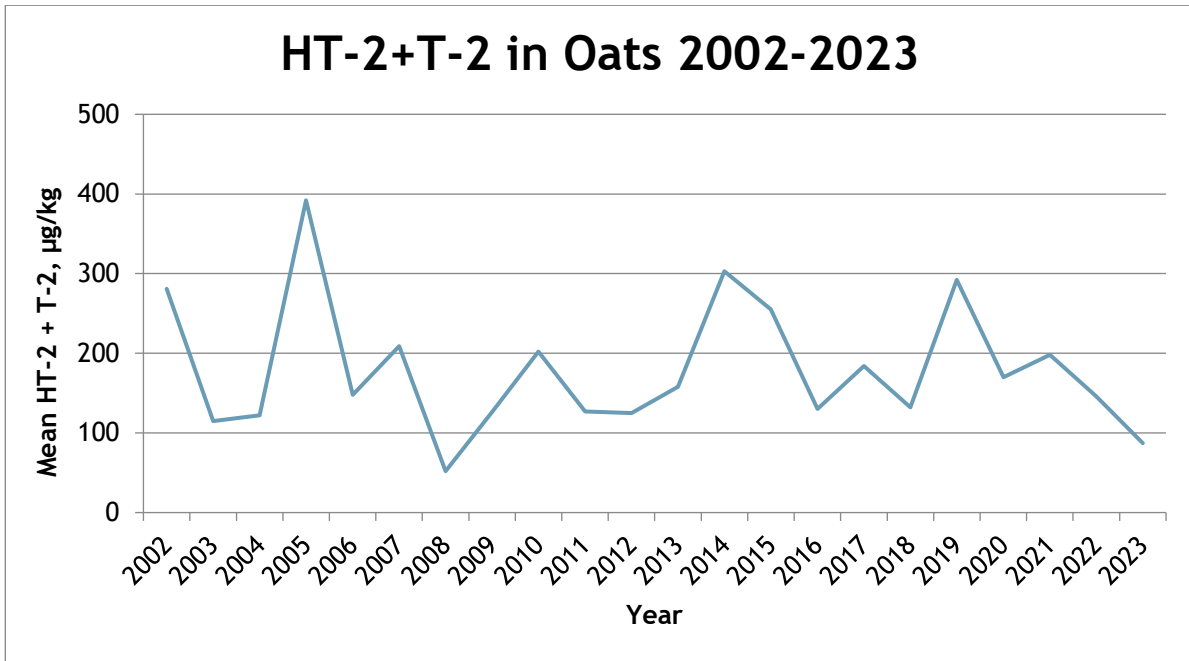


Figure 4. Mean concentration of the sum of T-2 toxin and HT-2 toxin in 30-60 samples of oats per year in the Norwegian surveillance programme for feed.

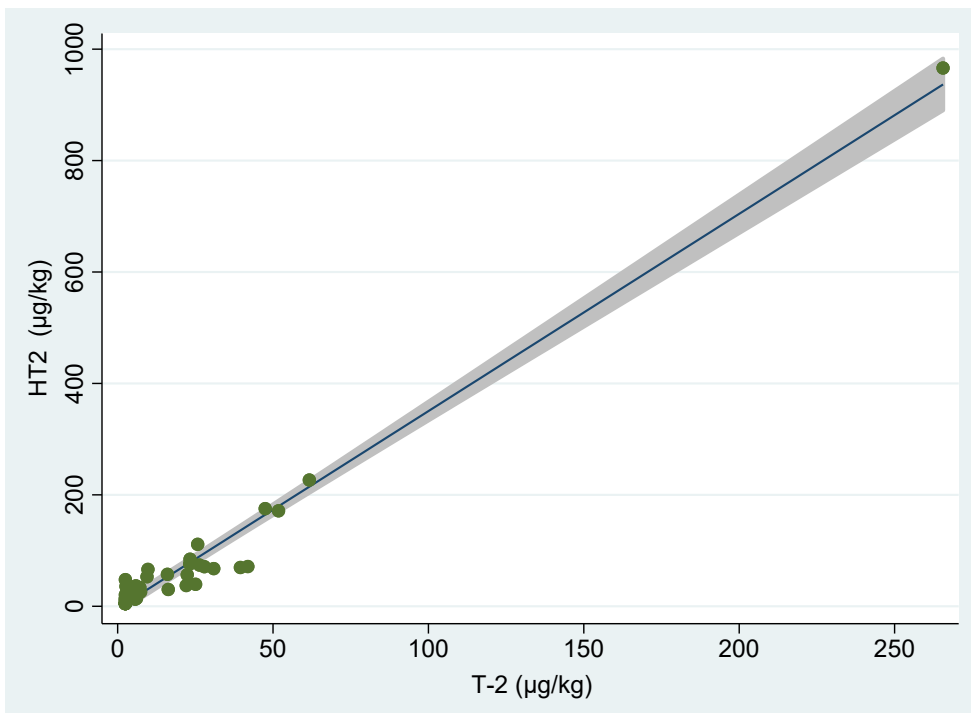


Figure 5. The Pearson correlation between T-2 toxin and HT-2 toxin in oats (N=43) 2023 was  $r=0.99$ ,  $p<0.0001$ . A regression line with 95 % confidence interval fitted to the points allows predictions of levels of HT-2 toxin given the level of T-2 toxin detected and vice versa.

NIV was detected at low levels in 42 % of the oats samples (Table 3). This was an increase from 20 % of positive samples in 2022. ZEA was detected in 86 % of the samples, compared to



0 in 2022. It should, however, be noted that the limit of quantification was lower in 2023 when comparing incidence of positive samples. Also, one sample had a very high concentration of zearalenone (2281 µg/kg) contributing to the high difference between mean and median concentration of ZEA (122 vs 20 µg/kg). When taking the measurement uncertainty into consideration all samples were below the recommended maximum limit for zearalenone in feed material (2000 µg/kg).

Mycotoxins in oats were analysed for regional differences (Table 4). Significantly higher level of DON was found in region Midt than in regions Øst and Stor-Oslo (South-Eastern Norway). No significant regional differences were found for T-2/HT-2.

**Table 4.** Survey on regional differences amongst Sør/Vest (Agder, Rogaland, Vestland), Øst (Buskerud, Vestfold, Telemark, Innlandet), Stor-Oslo (Akershus, Oslo, Østfold) and Midt (Trøndelag, Møre, Romsdal) on deoxynivalenol (DON), sum of T-2 and HT-2 toxin in oats (N = 43) sampled in Norway in 2023. Variables that were significantly different between regions are indicated by an \* ( $p < 0.05$ ).

Region		DON*	T-2+ HT-2
Sør-Vest n=1	Mean		
	St. dev.		
Øst n=14	Mean	1133	65.9
	St. dev.	1127	59.8
Stor-Oslo n=21	Mean	680	127.9
	St. dev.	487	262.9
Midt n=8	Mean	1150	25.4
	St. dev.	844	20.5

The weather during the growing season is usually a key factor for the *Fusarium* and mycotoxin contents of cereal grains. Among the mycotoxins, the influence of the weather is particularly studied for DON. The level of precipitation and humidity during flowering (usually in July), as well as precipitation up to harvest in autumn are considered particularly important in influencing *Fusarium* and DON levels, while the temperature may also play a role [1].

The weather in South-Eastern Norway (Regions Øst and Stor-Oslo) in the summer 2023 was wet in both July and August, and both Region Øst and Region Stor-Oslo was also hit by the extreme weather 'Hans' in August, leading to severe flooding. The summer in Region Midt had a normal rainfall with a few local areas with higher than normal precipitation. The temperature was warmer than normal in the grain-producing regions during the summer 2023[14].

#### *Mycotoxins and Claviceps purpurea in barley*

The pattern of lower concentrations of trichothecenes in barley compared with oats has been the same year after year [3-8, 13]. Table 5 shows the 2023 results of trichothecenes, ZEA and OTA in barley. In 2023, ZEA was quantified in 82 % of the samples, with a mean value of 127 µg/kg and median of 38 µg/kg. The maximum concentration of ZEA in barley was 2410 µg/kg in one sample. This is in contrast to 2022 when ZEA was not detected in any sample, but when the measurement uncertainty was taken into consideration still below the maximum

recommended level of 2000 µg/kg for feed materials. Trichothecenes were found in higher levels in 2023 than in 2022, but still in low concentrations compared to maximum levels in feed materials. OTA was not found in the samples.

*Claviceps purpurea* (ergot) and ergot alkaloids were also determined in barley (Table 6) as they are usually more present in this species than in oats and other cereals except rye. *C. purpurea* was detected in 54 % of the samples. The ergot had an overall mean level of 9.7 mg/kg and a maximum of 79.2 mg/kg. Thus, all samples were far below the legislated maximum concentration of 1000 mg/kg [11]. The occurrence of ergot was somewhat higher than the corresponding levels observed in barley in 2022 and similar to the levels found in 2019-2021 [3-6, 13]. As reported for 2022, most of the ergot sclerotia in barley were small in size, indicating they had been growing on other grass and randomly contaminating barley.

The ergot alkaloids were present at low levels in barley with a mean concentration of sum of alkaloids of 67 µg/kg and a maximum concentration of sum alkaloids of 1616 µg/kg. The alkaloids were detectable in 68 % of the samples. This is a higher percentage of positive samples than in previous years, but may be due to the new method able to detect and quantify lower levels of ergot alkaloids.

Ergot alkaloids have been included in the analysis repertoire of barley since 2016. They have been only sporadically present, but in some samples of significant concentrations: Except in 2018 where none was detectable, maximum sum alkaloids were between 2200 and 3000 µg/kg in 2016, -17 and -19 [4-8], which are levels of possible animal health concern if barley from these batches were used as major feed ingredients [6]. In 2020-2022, the maximum levels have been lower [13]. Knowledge on possible influence of climate or weather conditions on occurrence of ergot and ergot alkaloids is lacking.

**Table 5.** Concentrations (µg/kg) of deoxynivalenol (DON), 3-acetyl-DON (3-Ac-DON), 15-acetyl-DON (15-Ac-DON), DON-3-glucoside (DON-3-G), T-2 and HT-2 toxin and the sum of T-2/HT-2, nivalenol (NIV) and zearalenone (ZEA) in barley (n = 50) sampled in Norway in 2023.

	DON	3-Ac-DON	15-Ac-DON	DON-3-G	T-2	HT-2	T-2+HT-2	NIV	ZEA
Mean	318	32	<50.6	118	<12.5	<25	<37	<100	127
Median	261	<12.5	<50.6	91	<12.5	<25	<37	<100	38
Minimum	<25	21	<50.6	<25	<12.5	<25	<37	<100	<12.5
Maximum	1594.7	<203.1	<50.6	583.4	12.4	41.2	51.9	327.6	2409.7
SD*	345	42.3	0	128.4	3.0	7.3	9.2	55.0	364.70
% samples >dl*	84	36	0	62	26	52		24	82
% samples >gv*	0						0		0

\* SD = Standard Deviation, >dl = above detection limits, >gv = above guidance values.

**Table 6.** Concentrations of *Claviceps purpurea* sclerotia (mg/kg) and ergot toxins ( $\mu\text{g}/\text{kg}$ ) consisting of ergotamine/ergotaminine, ergocornine/ergocorninine, alpha-ergocryptine/alpha-ergocryptinine, ergocristine/ergocristinine and sum ergot alkaloids in barley ( $n = 50$ ) sampled in Norway in 2023.

	C. purpurea sclerotia	Ergo-novine/-inine	Ergo-sine/-inine	Ergot-amine/-inine	Ergo-cornine/-inine	$\alpha$ -Ergo-cryptine/-inine	Ergo-cristine/-inine	$\Sigma$ Ergot alkaloids
Mean	9.7	< 12.5	< 12.5	< 12.5	20	17	20	67
Median	0.7	< 12.5	< 12.5	< 12.5	<12.5	<12.5	<12.5	15
Minimum	0	< 12.5	< 12.5	< 12.5	<12.5	<12.5	<12.5	<12.5
Maximum	79.2	28.4	92.1	< 12.5	730	776.8	319.3	1615.7
SD*	16.7	6.9	17	1.8	103.5	109.8	50.1	234
% samples >dl*	54	7	28	1	10	12	52	68

\* SD = Standard Deviation, >dl = above detection limits.

Except for a significant correlation between ergot and ergot alkaloids in barley in 2021, lack of correlations between ergot and alkaloids were found in previous surveillance in Norway, in barley in 2019 and 2020, and in rye and wheat in 2016 and 2017 [3-5, 13, 15-16]. The low or lack of correlation indicate interactions with different variables in production of the alkaloids by ergot fungi. It could, however be pointed out that the levels of sclerotia in barley was low and dominated with very small sclerotia, indicating a potential contamination of the barley from grass.

Mycotoxins and ergot in barley were analysed for regional differences (Table 7). The concentrations of DON in barley from Stor-Oslo was significantly higher than in barley from the other regions ( $p = 0.001 - 0.013$ ). There were no significant differences for the other mycotoxins even if there is a tendency to a higher ZEA level in Region Stor-Oslo compared to the other regions.

**Table 7.** Survey on regional differences amongst Sør/Vest (Agder, Rogaland, Vestland), Øst (Buskerud, Vestfold, Telemark, Innlandet), Stor-Oslo (Akershus, Oslo, Østfold) and Midt (Trøndelag, Møre, Romsdal) on deoxynivalenol (DON), sum of T-2 and HT-2 toxin, nivalenol (NIV), zearalenone (ZEA) and sum of ergot alkaloider ( $\Sigma\text{Erg alk}$ ); all toxin concentrations  $\mu\text{g}/\text{kg}$ ) and *Claviceps purpurea* (mg/kg) in barley ( $N = 47$ ) sampled in Norway in 2023. There were no regional difference at the  $p=0.05$  statistical significance for all variables.

Region		DON	$\Sigma\text{T-2} + \text{HT-2}$	ZEA
Sør og Vest n=6	Mean	16.6	19.1	16.5
	St. dev.	10.0	0.9	30.2
Øst n=14	Mean	226.7	16.1	59.8
	St. dev.	221.5	4.4	61.4
Stor-Oslo n=20	Mean	553.0 <sup>1</sup>	16.9	259.5
	St. dev.	392.5	5.1	555.4
Midt n=10	Mean	162.5	21.5	23.4
	St. dev.	160.2	10.8	33.8

<sup>1</sup> Statistically significantly higher than the other regions.

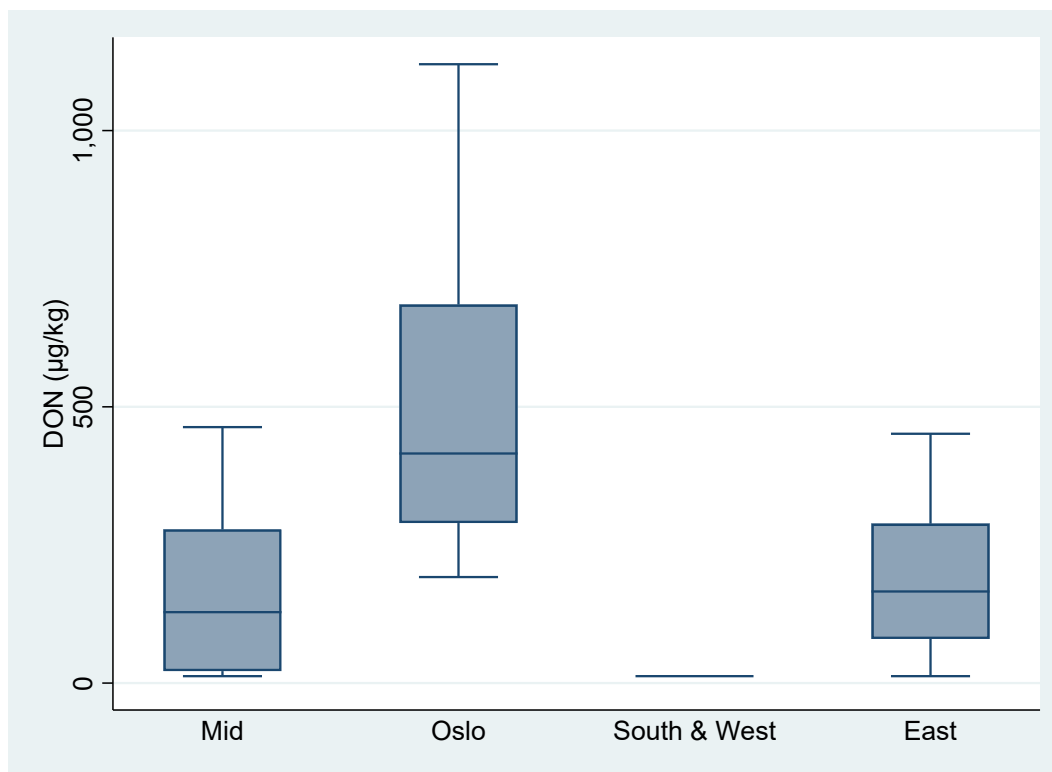


Figure 6. DON-concentrations (µg/kg) in barley collected in different geographical regions in Norway. ANOVA analysis and Bonferroni statistics showed that the DON concentrations in samples from region Stor-Oslo were significantly higher than the other regions (p = 0.001 - 0.013).

**Claviceps purpurea and mycotoxins in rye**

Claviceps purpurea was detected in 11 of the 12 rye samples with some samples containing high levels that exceeded the maximum limits for feed materials (Table 8). Still, the mean levels were considerably lower than in 2022. Also the concentrations of ergot alkaloids were lower than in 2022, with the highest concentration in 2023 being lower than the mean concentration in 2022. Correlation between ergot and ergot alkaloids was statistical significant (p=0.04; Figure 7). The samples were taken from the batches before the rye was cleansed of ergot. Thus, the results would not be representative for rye found in the market. The low number of samples of rye did not allow tests for regional variations.

Table 8. Occurrence of Claviceps purpurea sclerotia (mg/kg) and ergot toxins (µg/kg) consisting of ergotamine/ergotaminine, ergocornine/ergocorninine, alpha-ergocryptine/alpha-ergocryptinine, ergocristine/ergocristinine and sum ergot alkaloids in rye (n=12) sampled in Norway in 2023.

	C. purpurea sclerotia	Ergo-novine/-inine	Ergo-sine/-inine	Ergot-amine/-inine	Ergo-cornine/-inine	α-Ergo-cryptine/-inine	Ergo-cristine/-inine	Σ Ergot alkaloids
Mean	1558	<25	29	42,4	<50	<25	<50	144,6
Median	269	<25	<25	<25	<50	<25	<50	<25
Minimum	nd	<25	<25	<25	<50	<25	<50	<25
Maximum	7234	50,1	250,5	437,8	167,4	82,5	317,4	1157
SD*	2387	15,8	73	125,4	49,5	25,7	91,4	339
% samples >dl*	92	33	33	17	25	17	33	

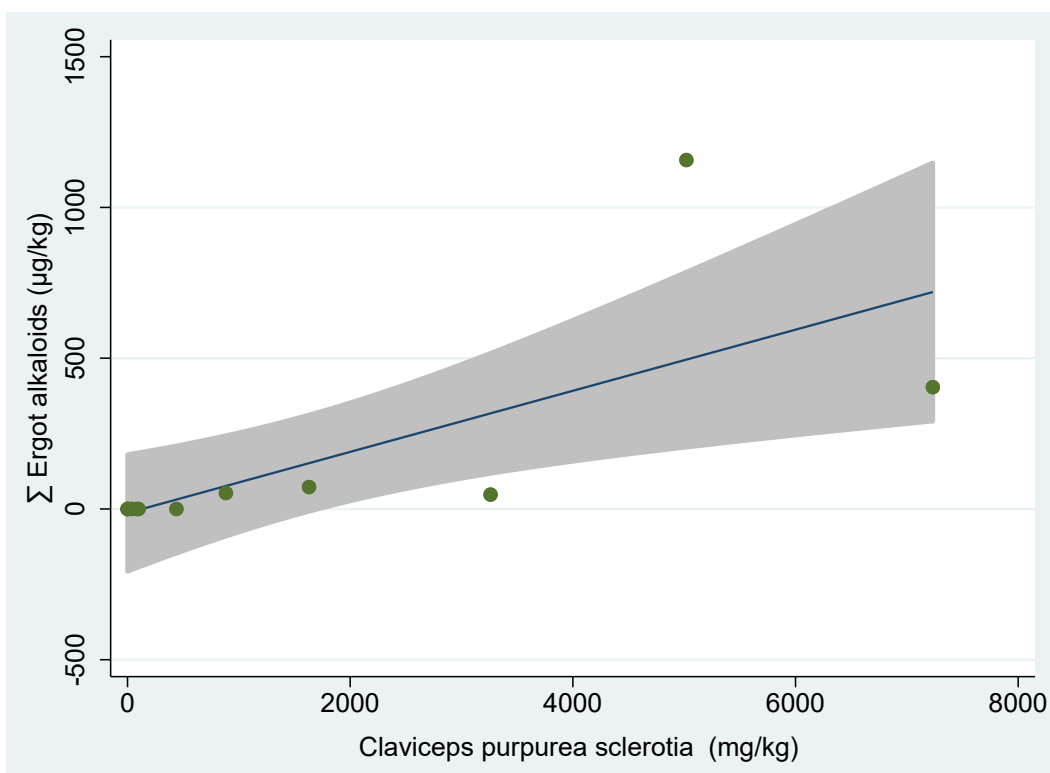


Figure 7. The Pearson correlation between *Claviceps purpurea* and sum of ergot alkaloids in samples of rye ( $n=12$ ) 2023 was  $r=0.63$ ,  $p=0.04$ . A regression line fitted to the points with 95 % confidence interval is shown.

#### Aflatoxins in maize

Aflatoxins and OTA were not detectable in any of the six analysed maize samples. (Table 9).

In addition to aflatoxins and OTA, ZEA was analysed in maize (Table 9). ZEA was detected in most samples up to 312 µg/kg, which were far below the guidance level of 3000 µg/kg [9].

Table 9. Concentrations (µg/kg) of aflatoxins (B1, B2, G1, G2), ochratoxin A (OTA) and zearalenone (ZEA) in maize ( $N = 8$ ) sampled in Norway in 2023.

	Afla B1	Afla B2	Afla G1	Afla G2	OTA	ZEA
Mean	<0,16	<0,31	<0,31	<0,31	<1,25	153
Median	<0,16	<0,31	<0,31	<0,31	<1,25	122
Minimum	<0,16	<0,31	<0,31	<0,31	<1,25	69,8
Maximum	<0,16	<0,31	<0,31	<0,31	<1,25	311,7
SD*						96
% samples >dl*	0	0	0	0	0	100
% samples >gv*	0				0	0

## Feed

### Feed for pigs

Samples of complete compound feed for pigs were analysed for trichothecenes, ZEA, OTA, fumonisins and ergot alkaloids.

The results on mycotoxins in complete compound feed for pigs in Table 10 show that DON was detected in all samples, but below the guidance level of DON for pig feed in Norway when the analytical measurement uncertainty was taken into consideration (500 µg/kg) [10]. The levels was however higher than in 2022. Co-occurrence of DON with DON-related compounds were mostly undetectable, with only trace amounts of DON-3-G in some samples. Related compounds of DON can be an additional factor to the total DON exposure and EFSA considers their toxic effects to be similar to that of DON [18].

T-2 and HT-2 were hardly present in samples of the compound feed for pigs and at concentrations below the guidance level (250 µg/kg) in all samples [10, 12]. NIV and OTA were not detected in any sample. However, it has to be commented that the detection limit for OTA in the multi-toxin method is higher (24 µg/kg) than the guidance level of OTA in feed for pigs at 10 µg/kg [10]. ZEA and fumonisins were present at trace concentrations in single samples.

Ergot alkaloids were present in several samples of the compound feed (Table 11). All concentrations were considered not to constitute any health risk for the pigs.

**Table 10.** Concentrations (µg/kg) of deoxynivalenol (DON), DON-3-glucoside (DON-3-G), sum of T-2 and HT-2 toxin, nivalenol (NIV), zearalenone (ZEA) fumonisin B1 and B2, and ochratoxin A (OTA) in complete compound feed for pigs (n = 20) sampled in Norway in 2023.

	DON	DON-3-G	T-2 + HT-2 <sup>1</sup>	NIV	ZEA	FUMB1	FUMB2	OTA
Mean	136.4	83.3	33.7	<201	<50	<100	<101	<50
Median	<100	<25	25.3	<201	<50	<100	<101	<50
Minimum	<100	<25	<15.2	<201	<50	<100	<101	<50
Maximum	661.7	288.0	111.0	<201	68.8	<100	<101	<50
SD*	187.9	110.9	27.8		21.7	10	0	0
% samples >dl*	100	20	75	0	65	0	0	0
% samples >gv*	0		0		0			0

\* SD = Standard Deviation, >dl = above detection limits, >gv = above guidance values.

<sup>1</sup> % samples above the dl for at least one of the toxins.

**Table 11.** Concentrations of ergot toxins ( $\mu\text{g}/\text{kg}$ ) consisting of ergotamine/ergotaminine, ergocornine/ergocorninine, alpha-ergocryptine/alpha-ergocryptinine, ergocristine/ergocristinine and sum ergot alkaloids in complete compound feed for pigs ( $n = 20$ ) sampled in Norway in 2023.

	Ergo- novine/- inine	Ergo- sine/- inine	Ergot- amine/- inine	Ergo- cornine/- inine	$\alpha$ -Ergo- cryptine/- inine	Ergo- cristine/- inine	$\Sigma$ Ergot alkaloids
Mean	7.5	9.2	0.7	0.7	1.5	11.0	22.0
Median	1.3	3.9	<6.6	<	<0.9	2.5	16.9
Minimum	< 0.5	<2.0	<6.6	<0.3	<0.9	<2.1	
Maximum	26.8	47.4	7.7	<7.2	22.1	60.8	93.7
SD*	10.0	13.1	2.2		5.0	17.1	25.5
% samples >dl*	60	70	10	1.8	25	60	85

### Feed for poultry

Trichothecenes and ZEA were present in low concentrations in all samples and all samples were well below the recommended maximum limits for the two toxins. Ergot alkaloids were found at low levels in some samples. OTA was not detected in any sample.

**Table 12.** Concentrations ( $\mu\text{g}/\text{kg}$ ) of deoxynivalenol (DON), DON-3-glucoside (DON-3-G), sum of T-2 and HT-2 toxin, nivalenol (NIV), zearalenone (ZEA) fumonisin B1 and B2, and ochratoxin A (OTA) in poultry feed ( $n = 21$ ) sampled in Norway in 2023.

	DON	DON-3-G	T-2 + HT-2	NIV	ZEA	OTA
Mean	243.6	22,7	54.1	<201	28.9	<11.8
Median	207.6	< 22.7	43.7	<201	25.1	<11.8
Minimum	34.1	<22.7	< 5.8	<201	2.9	<11.8
Maximum	559.6	57.4	194.7	<201	71.1	11.8
SD*		28.01	46.1		20.4	36.3
% samples >dl*	100	3	90	0	100	10
% samples >gv*	0		0		0	0

\* SD = Standard Deviation, >dl = above detection limits, >gv = above guidance values.

**Table 13.** Concentrations of ergot toxins ( $\mu\text{g}/\text{kg}$ ) consisting of ergotamine/ergotaminine, ergocornine/ergocorninine, alpha-ergocryptine/alpha-ergocryptinine, ergocristine/ergocristinine and sum ergot alkaloids in poultry feed ( $n = 21$ ) sampled in Norway in 2023.

	Ergo- novine/- inine	Ergo- sine/- inine	Ergot- amine/- inine	Ergo- cornine/- inine	$\alpha$ -Ergo- cryptine/- inine	Ergo- cristine/- inine	$\Sigma$ Ergot alkaloids
Mean	0.6	1.6	1.3	2.5	2.7	1.4	11.1
Median	<1.2	<0.5	<1.7	<0.4	<0.3	<4.0	2.4
Minimum	<1.2	<0.5	<1.7	<0.4	<<0.3	<4.0	
Maximum	3.4	11.6	10.8	41.7	50.2	11.1	148.6
SD*	1.1	2.8	3.0	9.0	10.9	3.2	32.0
% samples >dl*	0	43	19	38	24	19	67



## Conclusions

### Feed materials

- **Oats:** In 2023, the concentration of DON was the highest since 2012, probably due to a wet summer. There were no regional differences in DON levels. T-2/HT-2 toxin were lower than in 2022 and also lower than average concentrations in the last decade. Although with one exception still at low concentrations compared to guidance values for feed materials, oats had a higher ZEA concentrations in 2023 than in 2022.
- **Barley:** Trichothecenes were present in low levels in a high proportion of the samples, in concentrations lower than the guidance values for feed materials. ZEA was found in higher concentrations than in previous years, but no samples exceeded the guidance values for feed materials. Ergot was found in some samples, all at very low levels. No significant correlation between ergot and ergot alkaloids was present.
- **Rye:** Ergot was detected in all but one rye sample, in several at very high levels. Also ergot alkaloids were significantly elevated in several samples. Correlation between ergot and ergot alkaloids was statistically significant. As the samples were taken from the batches before the cleansing of ergot, the results were not representative for rye distributed at the market.
- **Mycotoxins in maize:** Aflatoxins and OTA were not detected in any of the six samples of maize. ZEA was found in all samples, all below the guidance levels.

### Feed

- **Compound feed for pig:** DON was found in all samples, but no sample exceeded the maximum guidance value when the uncertainty was taken into consideration. Other trichothecenes, ZEA, fumonisins and OTA were not detectable or present in low concentrations. Ergot alkaloids were found at low levels in some samples.
- **Compound feed for poultry:** Trichothecenes and ZEA were present in low concentrations in all samples. Ergot alkaloids were found at low levels in some samples. OTA was not detected.

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## Appendix

Appendiks Table 1. Results on mycotoxins (all in µg/kg) in 45 individual samples of oats from different regions in 2023. NIV=nivalenol, DON=deoxynivalenol, DON-3-G=DON-3-glucoside 3-Ac-DON=3-acetyl-DON, 15-Ac-DON=15-acetyl-DON, HT-2=HT-2 toxin, T-2=T-2 toxin, ZEA=zearalenone

ID nr.	NIV	DON	DON-3G	3-Ac-DON	15-Ac-DON	HT-2	T-2	ZEA
<b>Oats, Region Stor-Oslo</b>								
2023-23-146	334,2	290,3	0,0	44,9	0,0	966,2	265,6	11,7
2023-23-147	0,0	330,3	79,0	71,9	0,0	175,4	47,5	14,5
2023-23-149	52,2	519,3	92,1	83,2	0,0	20,6	0,0	46,4
2023-23-150	115,8	495,9	87,1	44,7	0,0	56,6	22,4	12,9
2023-23-151	147,3	480,1	108,4	67,5	0,0	111,2	25,8	697,8
2023-23-153	64,7	751,4	168,0	73,2	0,0	76,0	23,2	35,1
2023-23-154	33,5	291,5	42,4	20,3	0,0	26,2	4,2	28,1
2023-23-164	196,9	585,1	76,1	77,6	0,0	0,0	0,0	6,7
2023-23-165	46,0	440,5	89,4	28,6	0,0	12,7	5,7	0,0
2023-23-166	72,4	601,3	90,8	69,3	0,0	17,4	4,6	13,9
2023-23-179	0,0	2589,0	437,8	419,7	0,0	226,7	61,7	259,7
2023-23-180	0,0	574,9	144,4	46,4	0,0	84,5	23,3	48,3
2023-23-181	69,1	773,9	229,4	222,4	0,0	66,2	9,8	84,4
2023-23-182	0,0	696,8	220,0	64,6	0,0	71,1	27,9	10,0
2023-23-183	106,7	814,0	216,6	132,4	0,0	35,8	2,7	107,6
2023-23-188	77,0	724,4	81,1	72,2	0,0	30,5	3,7	52,1
2023-23-191	0,0	616,3	119,5	90,8	0,0	25,9	7,4	15,5
2023-23-192	0,0	451,9	47,8	28,0	0,0	33,7	7,1	9,1
2023-23-193	0,0	263,9	41,2	47,8	0,0	13,2	0,0	19,9

2023-23-195	0,0	1119,7	291,5	113,9	0,0	57,4	16,0	37,5
2023-23-196	0,0	869,9	256,7	98,4	0,0	0,0	0,0	129,1
<b>Oats, Region Øst</b>								
2023-23-158	0,0	796,1	137,1	101,2	0,0	71,4	41,9	2,5
2023-23-159	0,0	695,0	55,7	67,2	0,0	69,7	39,5	6,5
2023-23-160	89,4	4084,4	356,5	652,6	0,0	171,4	51,8	271,2
2023-23-161	0,0	1943,5	319,9	225,0	0,0	53,1	9,4	26,1
2023-23-162	0,0	1107,2	159,5	115,8	0,0	47,9	0,0	115,1
2023-23-163	0,0	401,9	59,6	34,9	0,0	37,3	22,1	0,0
2023-23-168	0,0	186,7	37,8	0,0	0,0	0,0	0,0	0,0
2023-23-175	87,9	822,8	137,2	65,4	0,0	9,7	0,0	5,3
2023-23-176	0,0	177,2	38,2	19,7	0,0	23,1	5,2	4,6
2023-23-205	159,9	907,1	214,4	160,2	0,0	67,7	30,9	443,4
2023-23-208	0,0	743,9	93,1	78,2	0,0	36,7	5,9	6,5
2023-23-227	98,1	2954,9	654,0	1061,7	0,0	0,0	0,0	2281,2
2023-23-227	0,0	255,7	112,8	26,7	0,0	0,0	0,0	141,5
2023-23-227	0,0	795,5	243,8	77,6	0,0	74,0	26,3	99,0
<b>Oats, Region Midt</b>								
2023-23-232	0,0	375,8	113,7	21,7	0,0	39,5	25,1	0,0
2023-23-232	0,0	957,0	67,2	360,9	0,0	14,2	6,1	21,0
2023-23-265	0,0	1279,0	219,3	345,2	0,0	25,6	3,0	69,8
2023-23-266	0,0	1671,6	311,7	114,0	5,9	0,0	0,0	7,1
2023-23-267	0,0	2080,2	315,9	61,4	0,0	0,0	0,0	15,2
2023-23-268	295,4	2375,6	630,4	203,5	0,0	14,5	0,0	82,9
2023-23-268	0,0	154,6	29,6	17,0	0,0	30,2	16,3	6,8
2023-23-277	201,4	312,3	22,5	15,9	0,0	8,6	0,0	0,0

Appendix Table 2. Results on mycotoxins ( $\mu\text{g}/\text{kg}$ ) and *Claviceps purpurea* ( $\text{mg}/\text{kg}$ ) in barley based on 45 individual samples from different regions in 2023. NIV=nivalenol, DON=deoxynivalenol, DON-3-G=DON-3-glucoside, 3-Ac-DON=3-acetyl-DON, 15-Ac-DON=15-acetyl-DON, T-2=T-2 toxin, HT-2=HT-2 toxin, ZEA=zearalenone. All alkaloids are given as a sum of two epimers, the -ine and -inine (eks. ergovine= ergovine + ergovinine).

ID-No	NIV	DON	DON-3G	3-Ac-DON	T-2	HT-2	Ergonovine	Ergosine	Ergotamine	Ergocornine	Alfa Ergocryptine	Ergocristine	ZEA	15-Ac-DON	Ergot-sclerotia ( $\text{mg}/\text{kg}$ )
<b>Region Stor-Oslo</b>															
2023-23-146	107,2	472,2	185,5	44,6	0,0	8,3	0,0	0,0	0,0	0,0	0,0	0,0	87,7	0,0	0,0
2023-23-147	0,0	292,9	140,5	32,7	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	70,9	0,0	0,7
2023-23-149	146,5	1594,7	583,4	203,1	4,5	13,2	0,9	6,5	0,0	0,0	0,0	0,0	392,7	0,0	0,0
2023-23-150	68,5	343,8	169,9	36,6	1,1	6,4	0,0	0,0	0,0	0,0	0,0	0,0	74,8	0,0	0,0
2023-23-151	73,0	463,8	200,9	41,7	3,9	10,2	28,4	74,0	0,0	730,0	776,8	6,4	62,3	0,0	0,0
2023-23-153	40,9	256,7	90,1	21,7	9,2	12,0	0,0	5,1	0,0	0,0	6,0	6,9	13,6	0,0	0,8
2023-23-154	136,6	503,7	214,2	65,0	0,0	2,7	0,0	0,0	0,0	0,0	0,0	0,0	39,6	0,0	0,0
2023-23-164	53,0	367,2	213,8	26,7	0,0	9,4	0,0	5,0	0,0	0,0	0,0	0,0	49,3	0,0	10,8
2023-23-165	0,0	201,7	165,1	22,3	0,0	6,6	0,0	0,0	0,0	0,0	0,0	0,0	37,0	0,0	0,0
2023-23-166	105,3	542,5	116,3	45,2	4,1	8,8	0,8	0,0	0,0	0,0	0,0	0,0	42,7	0,0	0,7
2023-23-179	0,0	1120,1	376,7	85,9	0,0	14,0	18,4	23,7	0,0	0,0	0,0	48,2	2409,7	0,0	25,0
2023-23-180	0,0	287,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	24,1	0,0	5,2
2023-23-181	0,0	658,2	203,4	64,3	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	231,0	0,0	0,0
2023-23-182	63,8	878,1	322,1	88,3	0,0	5,5	0,0	8,7	0,0	12,7	13,4	11,0	259,2	0,0	0,0
2023-23-183	0,0	711,9	213,9	46,7	0,0	16,4	0,0	0,0	0,0	0,0	0,0	0,0	145,1	0,0	0,0
2023-23-189	0,0	1319,1	369,9	165,4	12,4	17,5	0,0	12,1	0,0	0,0	0,0	24,2	1028,1	0,0	20,5
2023-23-191	0,0	191,9	124,9	12,7	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	21,0	0,0	0,0
2023-23-192	105,0	262,9	137,9	9,8	0,0	12,2	18,4	9,2	0,0	0,0	0,0	9,7	14,1	0,0	0,0
2023-23-193	0,0	292,2	126,2	19,6	0,0	13,0	18,9	16,0	0,0	0,0	0,0	16,7	79,0	0,0	3,6
2023-23-195	0,0	300,2	104,9	25,1	0,0	8,7	0,0	12,2	0,0	0,0	0,0	21,4	108,1	0,0	33,9
<b>Region Øst</b>															
2023-23-158	0,0	284,9	93,2	22,9	0,0	2,2	0,0	5,6	0,0	0,0	0,0	8,3	60,8	0,0	16,4
2023-23-159	34,0	323,3	82,6	65,2	4,3	16,6	0,0	0,0	0,0	0,0	0,0	0,0	63,5	0,0	2,9

2023-23-160	0,0	853,0	408,6	118,1	0,0	5,8	13,0	4,6	0,0	0,0	0,0	9,9	70,8	0,0	24,6
2023-23-161	0,0	451,3	233,4	58,9	2,6	3,8	0,0	16,4	0,0	0,0	0,0	26,5	189,8	0,0	51,9
2023-23-162	0,0	288,4	113,9	33,2	9,4	7,6	0,0	7,9	0,0	0,0	0,0	14,3	108,7	0,0	14,8
2023-23-163	0,0	90,3	38,7	28,3	0,0	9,4	0,0	0,0	0,0	6,5	0,0	0,0	175,1	0,0	18,6
2023-23-168	0,0	116,0	0,0	0,0	0,0	0,0	0,0	5,8	0,0	73,4	40,7	0,0	18,3	0,0	50,5
2023-23-168	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
2023-23-168	0,0	69,7	0,0	0,0	4,7	0,0	0,0	20,7	0,0	0,0	11,3	51,1	0,0	0,0	79,2
2023-23-169	0,0	14,8	24,4	0,0	0,0	0,0	0,0	0,0	0,0	0,0	6,3	0,0	35,0	0,0	10,9
2023-23-175	0,0	79,9	0,0	0,0	9,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	35,4	0,0	0,0
2023-23-176	0,0	258,8	53,2	0,0	0,0	9,5	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
2023-23-205	22,5	198,8	104,4	14,1	2,6	0,0	0,0	0,0	0,0	0,0	0,0	0,0	70,6	0,0	0,0
2023-23-208	0,0	132,8	91,9	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	9,5	0,0	0,0
<b>Region Sør og Vest</b>															
2023-23-171	0,0	0,0	0,0	0,0	0,0	0,0	0,0	5,3	0,0	0,0	0,0	11,5	2,3	0,0	2,7
2023-23-171	0,0	0,0	0,0	0,0	0,0	0,0	0,0	13,4	0,0	0,0	0,0	24,7	3,7	0,0	0,0
2023-23-171	0,0	0,0	0,0	0,0	0,0	0,0	0,0	3,4	0,0	0,0	0,0	8,6	0,0	0,0	0,0
2023-23-171	0,0	0,0	0,0	0,0	0,0	0,0	0,0	8,2	8,5	0,0	0,0	12,4	0,0	0,0	0,0
2023-23-172	0,0	0,0	0,0	0,0	0,0	0,0	0,0	4,9	0,0	0,0	0,0	34,4	16,0	0,0	0,0
2023-23-172	327,6	37,0	0,0	0,0	0,0	14,7	18,4	19,4	0,0	0,0	0,0	35,4	76,9	0,0	0,0
<b>Region Midt</b>															
2023-23-203	0,0	463,3	299,3	58,7	0,0	0,0	0,0	15,4	0,0	0,0	0,0	26,6	88,4	0,0	0,7
2023-23-204	0,0	127,9	0,0	18,6	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
2023-23-206	0,0	278,0	0,0	55,8	0,0	0,0	0,0	16,0	0,0	0,0	0,0	24,3	17,2	0,0	6,9
2023-23-207	0,0	23,4	0,0	0,0	0,0	6,9	0,0	92,1	10,1	0,0	17,7	319,3	0,0	0,0	14,5
2023-23-232	0,0	21,7	0,0	0,0	10,7	41,2	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0
2023-23-265	0,0	0,0	0,0	0,0	0,0	0,0	0,0	12,9	0,0	0,0	0,0	19,3	0,0	0,0	6,2
2023-23-267	0,0	194,9	0,0	16,9	0,0	0,0	0,0	15,7	0,0	0,0	0,0	35,3	28,5	0,0	9,8
2023-23-268	0,0	128,8	0,0	10,1	0,0	0,0	19,0	17,1	0,0	13,9	0,0	36,6	12,9	0,0	29,1
2023-23-278	0,0	0,0	0,0	0,0	0,0	0,0	0,0	41,0	0,0	0,0	0,0	162,1	5,5	0,0	41,7
2023-23-279	0,0	361,8	83,4	21,9	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	81,5	0,0	3,3



Appendix Table 3. Results on mycotoxins ( $\mu\text{g}/\text{kg}$ ) and *Claviceps purpurea* ( $\text{mg}/\text{kg}$ ) in rye based on 12 individual samples from different regions in 2023. All alkaloids are given as a sum of two epimers, the -ine and -inine (eks. ergovine= ergovine + ergovinine).

PJS nr.	Ergonovin	Ergosine	Ergotamine	Ergocornine	Alfa Ergocryptine	Ergocristine	Meldrøye- sklerotier ( $\text{mg}/\text{kg}$ )
<b>Region Stor-Oslo</b>							
2023-23-147	50,08	250,54	437,80	59,45	42,03	317,37	5019,90
2023-23-153	0,00	0,00	0,00	0,00	0,00	0,00	3,20
2023-23-190	27,50	69,27	46,79	167,35	82,53	10,63	7233,50
2023-23-239	0,72	24,70	0,00	27,57	0,00	0,00	884,10
<b>Region Øst</b>							
2023-23-176	0,00	0,00	0,00	0,00	0,00	0,00	37,40
2023-23-176	0,00	0,00	0,00	0,00	0,00	0,00	82,70
2023-23-176	0,00	0,00	0,00	0,00	0,00	72,74	1630,60
2023-23-176	0,00	0,00	0,00	0,00	0,00	0,00	0,00
2023-23-208	0,13	4,33	24,04	0,00	0,00	19,21	3261,00
2023-23-208	0,00	0,00	0,00	0,00	0,00	0,00	99,10
2023-23-208	0,00	0,00	0,00	0,00	0,00	0,00	3,30
2023-23-208	0,00	0,00	0,00	0,00	0,00	0,00	439,70

Appendix Table 4. Results on mycotoxins ( $\mu\text{g}/\text{kg}$ ) *rea* ( $\text{mg}/\text{kg}$ ) in six individual samples of maize 2023. AflaB1= aflatoxin B1, AflaB2=aflatoxin B2, AflaG1= aflatoxin G1, AflaG2=aflatoxin G2, Ochra= chratoxin A.

PJS nr.	Afla B1	Afla B2	Afla G1	Afla G2	Ochra	Zea
2023-21-67	IP	IP	IP	IP	IP	163,01
2023-21-82	IP	IP	IP	IP	IP	212,56
2023-21-101	IP	IP	IP	IP	IP	82,56
2023-21-224	IP	IP	IP	IP	IP	80,12
2023-21-253	IP	IP	IP	IP	IP	69,83
2023-23-265	IP	IP	IP	IP	IP	311,74

Appendix Table 5. Results on mycotoxins ( $\mu\text{g}/\text{kg}$ ) in pig feed (20 samples) from different regions in 2023. NIV=nivalenol, DON=deoxynivalenol, DON-3-G=DON-3-glucoside, 3-Ac-DON=3-acetyl-DON, 15-Ac-DON=15-acetyl-DON, T-2=T-2 toxin, HT-2=HT-2 toxin, ZEA=zearalenone. All alkaloids are given as a sum of two epimers, the -ine and -inine (eks. ergovine= ergovine + ergovinine).

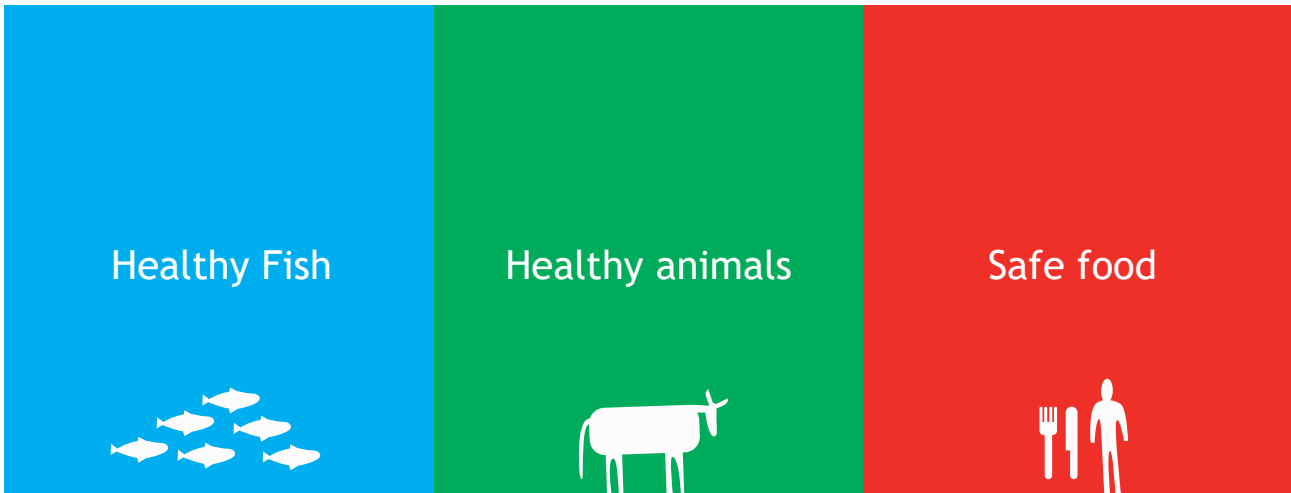
ID-no.	NIV	DON	DON-3G	Ergonovine	3-Ac-DON	HT-2	OTA	Ergosine	Ergotamine	T-2	Ergocornine	Alfa Ergocryptine	Ergocristine	Fum B1	Fum B2	ZEA	15-Ac-DON
<b>Region Stor-Oslo</b>																	
2023-21-56	IP	70,68	IP	IP	IP	23,29	IP	12,24	IP	11,62	IP	5,29	23,51	IP	IP	IP	IP
2023-21-62	IP	70,64	IP	4,10	IP	48,65	IP	14,47	IP	26,25	IP	0,91	49,33	IP	IP	23,42	IP
2023-21-66	IP	33,47	IP	IP	IP	24,04	IP	7,24	IP	13,88	7,20	2,36	IP	IP	IP	3,65	IP
2023-21-160	0,00	54,32	0,00	0,66	0,00	53,77	0,00	3,51	0,00	11,44	0,00	0,00	2,70	0,00	0,00	0,00	0,00
2023-21-249	0,00	361,08	112,78	19,52	13,81	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	34,20	0,00
<b>Region Øst</b>																	
2023-21-83	IP	21,08	IP	IP	IP	15,36	IP	IP	IP	8,56	IP	IP	5,90	IP	IP	1,94	IP
2023-21-217	0,00	389,89	128,06	0,00	14,30	12,52	0,00	0,00	0,00	3,48	0,00	0,00	0,00	0,00	0,00	64,96	29,74
2023-21-248	0,00	0,00	0,00	20,16	0,00	17,49	0,00	0,00	0,00	0,00	0,00	0,00	14,55	0,00	0,00	0,00	0,00
<b>Region Midt</b>																	
2023-21-98	IP	63,48	IP	IP	IP	41,17	IP	IP	IP	24,96	IP	IP	2,30	IP	IP	8,56	IP
2023-21-255	0,00	29,19	0,00	24,12	0,00	0,00	0,00	47,41	0,00	0,00	0,00	22,14	60,84	0,00	0,00	0,00	0,00

2023-21-255	0,0 0	0,00	0,00	26,78	0,00	0,00	0,00	34,74	0,00	0,00	0,00	0,00	22,65	0,00	0,00	11,5 2	0,00
2023-21-256	0,0 0	458,1 7	220,8 0	18,48	29,5 2	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	2,56	0,00	17,3 2	0,00
2023-21-256	0,0 0	661,6 6	287,9 9	23,06	58,0 8	0,00	0,00	28,27	0,00	0,00	0,00	0,00	0,00	0,00	0,00	68,8 5	19,20
<b>Region Sør og Vest</b>																	
2023-21-107	IP	IP	IP	0,53	IP	22,7 6	IP	1,99	IP	6,41	IP	IP	4,28	IP	IP	1,66	IP
2023-21-107	IP	IP	IP	5,82	IP	21,1 1	IP	6,30	IP	5,50	IP	IP	14,10	IP	IP	IP	IP
2023-21-161	0,0 0	0,00	0,00	4,77	0,00	35,5 7	11,7 6	2,08	0,00	12,9 5	0,00	0,00	2,06	0,00	0,00	0,00	0,00
2023-21-32	IP	36,62	IP	IP	IP	67,1 3	IP	3,58	IP	43,9 1	IP	IP	IP	IP	IP	IP	IP
2023-21-32	IP	IP	IP	IP	IP	16,0 5	IP	4,28	7,70	IP	1,69	0,05	IP	45,2 0	22,4 4	3,76	IP
2023-21-33	IP	154,1 8	IP	IP	IP	30,5 8	IP	13,50	6,60	14,0 5	4,07	IP	18,09	IP	IP	8,19	IP
2023-21-33	IP	260,5 6	IP	IP	IP	12,1 6	IP	4,41	IP	4,78	0,26	IP	IP	IP	IP	4,83	IP

Appendix Table 6. Results on mycotoxins ( $\mu\text{g}/\text{kg}$ ) in poultry feed (21 samples) from different regions in 2023. NIV=nivalenol, DON=deoxynivalenol, DON-3-G=DON-3-glucoside, 3-Ac-DON=3-acetyl-DON, 15-Ac-DON=15-acetyl-DON, T-2=T-2 toxin, HT-2=HT-2 toxin, ZEA=zearalenone. All alkaloids are given as a sum of two epimers, the -ine and -inine (eks. ergovine= ergovine + ergovinine).

ID-no.	NIV	DON	DON-3G	Ergovine	3-Ac-DON	HT-2	OTA	Ergosine	Ergotamine	T-2	Ergocornine	Alfa - Ergo-cryptine	Ergocristine	Fum B1	Fum B2	ZEA	15-Ac-DON
<b>Region Stor-Oslo</b>																	
2023-21-63	IP	47,08	IP	IP	IP	23,04	IP	2,22	IP	16,95	IP	0,32	IP	46,43	49,63	55,66	IP
2023-21-64	IP	34,54	IP	3,37	IP	9,35	IP	11,57	IP	5,78	41,74	50,19	11,11	IP	IP	52,49	IP
2023-21-160	0,00	340,79	0,00	0,00	0,00	25,42	0,00	0,87	0,00	12,27	1,12	0,00	0,00	0,00	0,00	18,70	49,79
2023-21-162	0,00	559,57	57,28	1,23	20,97	0,00	0,00	3,68	0,00	0,00	3,98	0,00	0,00	0,00	0,00	61,60	52,35
<b>Region Øst</b>																	
2023-21-71	IP	78,57	IP	2,60	IP	13,91	IP	3,85	6,94	IP	IP	IP	8,04	IP	IP	8,19	IP
2023-21-71	IP	403,53	21,59	IP	28,12	97,36	IP	0,45	IP	17,06	IP	IP	5,94	57,33	43,81	71,12	58,18
2023-21-71	IP	56,78	IP	2,55	IP	9,43	IP	3,75	IP	IP	IP	IP	4,00	IP	15,50	23,28	IP
2023-21-100	IP	207,61	IP	IP	13,25	13,95	IP	IP	IP	10,14	IP	IP	IP	24,23	20,95	30,54	32,14
2023-21-100	IP	227,48	IP	IP	12,66	16,00	IP	IP	IP	IP	IP	IP	IP	IP	IP	32,51	31,44
<b>Region Sør og Vest</b>																	

2023-21-218	0,00	374,76	0,00	0,00	0,00	144,62	81,17	0,00	0,00	50,04	0,00	0,00	0,00	98,75	35,86	38,29	69,01
242023-21-218	0,00	206,32	0,00	0,00	0,00	37,85	0,00	1,40	0,00	10,16	0,00	0,00	0,00	179,61	51,67	13,59	28,70
2023-21-218	0,00	488,52	57,42	0,00	0,00	55,27	0,00	0,00	0,00	30,70	0,00	0,00	0,00	21,91	0,00	47,41	48,77
2023-21-32	IP	191,68	IP	IP	IP	89,97	IP	IP	IP	28,10	IP	IP	IP	IP	3,40	10,36	IP
2023-21-33	IP	194,36	IP	IP	IP	42,60	IP	IP	7,05	IP	1,28	0,34	IP	IP	IP	3,99	IP
2023-21-33	IP	250,92	IP	IP	IP	34,27	IP	5,36	10,78	9,39	1,29	IP	IP	IP	IP	3,62	IP
<b>Region Midt</b>																	
2023-21-77	IP	147,64	IP	2,40	IP	12,45	IP	IP	IP	IP	IP	IP	IP	IP	IP	7,82	IP
2023-21-93	IP	373,35	IP	IP	IP	61,02	IP	IP	IP	23,41	0,44	4,43	IP	IP	3,06	23,81	30,59
2023-21-94	IP	38,34	IP	IP	IP	40,87	IP	IP	IP	11,80	IP	IP	IP	IP	IP	2,87	IP
2023-21-95	IP	198,79	IP	IP	IP	57,47	IP	IP	IP	19,51	IP	IP	IP	IP	IP	34,60	IP
2023-21-96	IP	376,89	IP	IP	IP	20,81	IP	IP	1,73	8,83	1,18	2,35	IP	20,99	12,45	41,73	43,57
2023-21-97	IP	318,96	IP	IP	IP	41,93	IP	IP	IP	21,17	0,58	IP	IP	17,54	IP	25,15	30,53



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