MONITORING PROGRAM FOR PHARMACEUTICALS, ILLEGAL SUBSTANCES AND CONTAMINANTS IN FARMED FISH

Annual report for 2019

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Summary (English):

This report summarises the monitoring data collected in 2019 on the status of illegal substances, pharmaceuticals and contaminants in Norwegian farmed fish. A total of 13 725 fish were collected. Samples examined for illegal compounds would be collected at all stages of farming and are representative of farmed fish under production. The samples were analysed for substances with anabolic effects or unauthorized veterinary drugs. No residues of illegal compounds were detected. Samples tested for approved veterinary drugs and contaminants were collected at processing plants and are representative of Norwegian farmed fish ready for human consumption. Residues of anti-sea-lice agents were found in five samples, the levels present were below the Maximum Residue Limit (MRL) for all samples. Other veterinary drugs, like antibiotics or drugs used against internal parasites were not found. No environmental contaminants were found above the EU maximum levels.

Summary (Norwegian):

Denne rapporten oppsummerer overvåkingsresultatene fra 2019 for ulovlige stoffer, legemidler og miljøgifter i norsk oppdrettsfisk. Totalt ble det samlet 13 725 fisk. Prøver som ble analysert for ulovlige forbindelser, som stoffer med anabole effekter eller uautoriserte legemidler, ble tatt ut under alle livsstadier, og er representative for oppdrettsfisk under produksjon. Ingen rester av ulovlige forbindelser ble detektert. Prøver som ble testet for godkjente veterinære legemidler og miljøgifter ble samlet inn på slakterier, og er representativt for norsk oppdrettsfisk som er klar for markedet. Rester av lusemidler ble funnet i fem prøver, nivåene var under grenseverdien (MRL) for alle prøvene. Andre veterinære legemidler, som antibiotika eller legemidler brukt mot interne parasitter ble ikke funnet. Ingen miljøgifter ble funnet over EUs maksimumsgrenser.

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1 - Introduction

1.1 - Background

According to EU legislation (EU 96/23/EC), all food producing animals should be monitored for certain substances and residues thereof. The following residues or substance groups are monitored in Norwegian farmed fish:

Group A Substances with anabolic effects and unauthorized substances:

A1: Stilbenes, derivatives and their salts and esters

A3: Steroids

A6: Prohibited substances

Group B Veterinary drugs and contaminants:

B1: Antibacterial agents

B2a: Anthelmintics

B2d: Sedatives

B3a: Organochlorine compounds

B3b: Organophosphorus compounds

B3c: Chemical elements

B3d: Mycotoxins

B3e: Dyes

B3f: Others

1.2 - Group A, Substances with anabolic effects and unauthorized substances

Fish tested for illegal compounds were collected at the farm by official inspectors from the Norwegian Food Safety Authorities, without prior notification to the farmers. Samples were taken at all stages of farming in order to represent farmed fish during production. Substances monitored in Group A include growth promoters like steroids and stilbenes, and unauthorized drugs. Unauthorized drugs considered most relevant for aquaculture are chloramphenicol, nitrofurans, metronidazole and dyes. Since the use of the dyes malachite green, crystal violet and brilliant green is not allowed for food producing species (EU 37/2010), they are considered Group A substances and hence monitored in samples throughout the production chain. However, according to directive 96/23 these dyes belong to the group B3e. Thus, in order to fulfill criteria for group B sampling, some of the samples assigned to analysis of dyes were also collected at the slaughterhouse.

To ensure harmonized levels for the control of unauthorized substances, the analytical methods should meet a minimum required performance limits (MRPLs) set by the European Union (EU 2003/1881, EU 2004/25, CRL 2007), and European reference laboratories (EU-RLs), (EU 2003/1881, EU 2004/25, CRL 2007). Table 6.3 gives an overview of MRPLs of relevant compounds.

1.3 - Group B, veterinary drugs

In order to protect public health, current EU legislation (EU 37/2010) provisions the assignment of Maximum Residue

Limits (MRLs) for all legally applied pharmacologically active substances in products intended for human consumption. An MRL denotes the highest permitted residual concentration of a legally applied veterinary drug and is evaluated for each substance and each food product individually. Consumption of food with drug residues below the MRL should not pose a health risk to the consumer. For fish, the MRLs are set for muscle and skin in natural proportions. Samples examined for veterinary drugs were collected from fish at processing plants and the samples are representative of fish ready to be placed on the market for human consumption.

1.4 - Group B, contaminants

Samples examined for contaminants were collected from fish at processing plants and are representative of fish ready for human consumption. The EU (EU 1881/2006) has set a maximum level (ML) for some of the contaminants in fish, while for others, like the pesticides, PAH, PFC and BFR, maximum levels have not yet been established.

1.5 - Ethoxyquin

Ethoxyquin (EQ) is a synthetic antioxidant, which has widely been used as an additive (E324) in components for animal feed for pets and livestock to preserve product quality and increase shelf live. Because of its high efficacy, EQ has also been widely used by the global fishmeal industry both as a nutritional preservative, but also to avoid oxidation and self-ignition under long-distance transport.

2 - Material and methods

2.1 - Sampling

Samples were taken on fish farms or slaughterhouses in all fish-producing regions in Norway by official inspectors from the NFSA. The sampling plan was randomised according to season and region. In 2019, the following fish species were included in the monitoring program: Atlantic salmon (*Salmo salar*), rainbow trout (*Oncorhynchus mykiss*), turbot (*Scophthalmus maximus*), Atlantic halibut (*Hippoglossus hippoglossus*), and Arctic char (*Salvelinus alpinus*). Samples were transported to IMR in a frozen state. For most analyses, the Norwegian quality cut (NQC) was used (Johnsen, Hagen et al. 2011). However, both NQC and individual liver samples were collected for analysis of antibiotics. Samples to be used for analyses of substances with anabolic effects or unauthorized substances also included small fish from early life stages and in these cases, the whole fish except head, tail and gut were homogenised. The samples were analysed as pooled samples comprising five fish from the same cage/farm.

2.2 - Pre-treatment

Upon arrival at IMR the sample identification was anonymised for the analysts. A back-up sample was stored for all samples. Pooled samples of muscle from five fish from the same cage/farm were homogenised before analyses. Samples of liver were excised from the fish to be screened for residues of antimicrobial agents by the microbiological inhibition zone assay. Liver samples were examined individually, if residues were detected, the back-up sample of muscle was analysed by chemical methods. The maximum residue limits for veterinary drugs are set for muscle and skin in natural proportions (EU 37/2010). Therefore, according to the analytical protocol, any detection of drug residues in the muscle or liver was followed by a re-analysis of the back-up sample, consisting of muscle and skin in natural proportions, in duplicate.

2.3 - Analytical methods

The laboratory routines and most of the analytical methods are accredited in accordance with the standard ISO 17025 (Table 6.3). A summary of the analytical methods and their limit of detection (LOD) or limit of quantification (LOQ) is shown in Table 6.3. The LOD is the lowest level at which the method is able to detect the substance, while the LOQ is the lowest level for a reliable quantitative measurement. For all methods, a sample blank and a quality control sample (QC) with a known composition and concentration of target analyte are included in each series. The methods are regularly verified by participation in inter laboratory proficiency tests, or by analysing certified reference material (CRM), where such exist.

2.3.1 - Group A substances

A1, Stilbenes

Stilbenes were extracted by water and acetonitrile. Liquid-liquid extraction was used for sample clean up. The stilbenes were analysed by LC-MS/MS.

A3, Steroids

Steroids were extracted by water and acetonitrile. Liquid-liquid extraction followed by s olid phase extraction was used for sample clean up, before the samples were analysed by LC-MS/MS.

A6, Illegal veterinary drugs

Chloramphenicol

Chloramphenicol was extracted with ethyl acetate. Liquid-liquid extraction was used to purify the extract. The samples were analysed by LC-MS/MS.

Nitrofurans

The nitrofuran metabolites were extracted with aqueous hydrochloric acid and derivatized with nitrobenzaldehyde. Solid phase extraction was used for sample clean up. The analytes were determined by LC-MS/MS.

Metronidazole

Metronidazole and its metabolite hydroxymetronidazole were extracted by ethyl acetate. Solid phase extraction was used for sample clean up. The analytes were determined by LC-MS/MS

Malachite green (MG), crystal violet (CV), brilliant green (BG)

The analytes were extracted with acetonitrile and dichloromethane. Samples clean-up were performed by solid phase extraction. MG, CV, BG and the metabolites leuco malachite green (LMG) and leuco crystal violet (LCV), were determined by LC-MS/MS.

2.3.2 - Group B substances

B1, Antibacterial agents (antibiotics)

The presence of antibacterial agents was determined by a three-plate microbiological assay or by chemical analysis.

Microbiological assay

For the three-plate microbiological inhibition method, a specific bacterial strain was added to a plate containing growth agar. Small pieces of liver were placed on the plates before incubation. If the samples contained residues of antibacterial agents, the bacterial growth would be inhibited in a zone around each piece of liver tissue. Thus, a transparent zone with no bacterial growth surrounding the liver sample would indicate a positive sample. Any positive detection was verified by chemical analysis of muscle and skin.

Florfenicol, oxolinic acid, flumequine, enrofloxacin, ciprofloxacin and trimethoprim

The analytes were extracted with acetonitrile and water. The analysis was performed by LC-MS/MS.

Oxytetracyclin

The analyte was extracted with acetonitrile. Liquid-liquid extraction was used to purify the extract. Oxytetracyclin was analysed by LC-MS/MS.

B2a, Anthelmintics

Diflubenzuron, teflubenzuron, lufenuron, hexaflumuron and fluazuron

The analytes were extracted with acetone. Solid phase extraction was used for sample clean up. The samples were analysed by LC-MS/MS (Samuelsen, Lunestad et al. 2014).

Emamectin

Emamectin was extracted with acetonitrile, and analysed by LC-MS/MS.

Ivermectin

Ivermectin was extracted with organic solvent, and the extract were purified by solid phase extraction. The samples were analysed by LC-MS/MS

Cypermethrin and deltamethrin

Cypermethrin and deltamethrin were extracted by soxhlet extraction. The extracts were purified by gel permeation

chromatography. The samples were analysed by GC-MS/MS.

Fenbendazole and praziquantel

Fenbendazole and praziquantel were extracted from the sample by acetone, and analysed by LC-MS/MS.

B2d, Sedatives

Isoeugenol

Isoeugenol was analysed by GC coupled to a flame ionization detector (FID).

B3a, Organochlorine compounds

Dioxins, dl-PCBs, PCB-6 and PBDEs.

This is an adaptation to modern clean-up equipment of the US-EPAs (Environmental Protection Agency) methods No. 1613 and 1668. Separation and quantification were performed by high resolution gas chromatography/high resolution mass spectrometry (HRGC/HRMS). The method measures all of the 29 compounds on the WHO list: 17 PCDD / PCDF congeners, four non-ortho substituted PCBs: PCB -77, 81, 126 and 169 and eight mono-ortho substituted PCBs: PCB-105, 114, 118, 123, 156, 157, 167 and 189 (Berntssen, Julshamn et al. 2010). The PCBs included in PCB-6, PCBs no. 28, 52, 101, 138, 153 and 180, were analysed by GC-MS/MS. The PBDEs were analysed by GC/MS in a relevant solvent fraction from the EPA clean-up procedure (Pirard, De Pauw et al. 2003).

Chlorinated pesticides

Pesticides were extracted by organic solvent, and the extract were cleaned-up by column chromatography, before the pesticides were analysed by HRGC-HRMS.

B3b, Organophosphorus compounds

Azamethiphos and dichlorvos

The analytes were extracted with acetonitrile, and analysed by LC-MS/MS.

B3c, Elements

Lead, mercury, cadmium, arsenic, cobalt, chromium, copper, iron, manganese, molybdenum, nickel, selenium, silver, vanadium, and zinc

The sample was decomposed by acid treatment, assisted by heat and high pressure. The metals were analysed by inductively coupled plasma mass spectrometer (ICP-MS) (Julshamn, Maage et al. 2007).

Inorganic Arsenic

Inorganic arsenic was extracted by hydrochloric acid in hydrogen peroxide at 90 °C. Inorganic arsenic includes As (III) and As (V). As (III) was oxidised to As (V) during the extraction. Inorganic arsenic was separated from other arsenic compounds by anionic exchange HPLC, and detected by ICP-MS.

Methylmercury

Methylmercury was extracted by Tetramethylammonium Hydroxide. The pH was adjusted before derivatization and extraction by hexane. The samples were analysed by GC-ICP-MS.

Tributyltin

Tributyltin was extracted by acetic acid/methanol. The pH was adjusted before derivatization and extraction by hexane.

The samples were analysed by GC-ICP-MS.

B3d, Mycotoxins

Enniatin and beauvericin

The mycotoxins; beauvericin, enniatin A, enniatin A1, enniatin B and enniatin B1 were extracted with acetonitrile and water. Solid phase extraction was used for sample clean up. The mycotoxins were analysed by LC-MS/MS.

B3f, Others

HBCD

HBCD was extracted by a soxhlet apparatus, using a mixture of acetone and hexane. Sulfuric acid was used for purification. The extract was further cleaned up by an alumina column. The HBCD isomers were analysed by LC-MS/MS.

TBBPA

TBBPA was extracted by a soxhlet apparatus using a mixture of acetone and hexane. Sulfuric acid was used for purification. O-bis(trimethylsilyl)trifluoroacetamide (BSTFA) was used for derivatization. The extract was purified using column chromatography. TBBPA was analyzed by GC-MS using Electron Ionization (EI).

PFC

PFCs were extracted by methanol, the extract was purified by solid phase extraction. PFCs were analysed by LC-MS/MS.

PAH

PAHs were extracted by dichloromethane and cyclohexane using an Accelerated Solvent Extractor (ASE). The extract was purified by solid phase extraction and analysed by GC-MS/MS.

Ethoxyquin

EQ and EQDM were extracted with hexane from pooled muscle samples, after saponification in a mixture of ethanol, NaCl and NaOH. EQ and EQDM were quantified by reversed-phase high-performance liquid chromatography with fluorescence detection, using an external standard curve, as previously described by Bohne et al. (2007), with modifications described by Ørnsrud et al. (2011).

Table 2.1. Number of fish analysed for each substance.

Compounds	Fish	Atlantic salmon	Rainbow trout	Atlantic halibut	Arctic char	Turbot
A1 Stilbenes						
Zeranol 17alpha-Estradiol 17alpha-Ethinyl-estradiol 17beta-Estradiol beta-Zearalanol Dienestrol Diethylstilbestrol Estriol Estrone Hexestrol	830	775	50	5		
A3 Steroids						
16-Hydroxystanozolol 17alpha-Boldenone 17alpha-Trenbolone alpha-Nandrolone Boldenone Chlor-Testosterone Epitestosterone Methyl-Boldenone Methyltestosterone Nortestosterone Stanozolol Testosterone Testosterone propionate Trenbolone Trenbolone-acetate	830	770	60			
A6 Illegal substances						
Chloramphenicol	830	765	60	5		
Metronidazole	830	750	75	5		
Nitrofuran metabolites (AOZ, AMOZ, AHD, SEM)	830	760	65	5		
Malachite green * Crystal violet Brilliant green	835	770	60	5		
B1 Antibiotics						
Oxytetracycline Doxycycline Chlortetracycline Tetracycline	105	100	5			
Florfenicol Flumequine Oxolinic acid Enrofloxacin Ciprofloxacin Trimethoprim	450	405	40		5	
Quinolones (liver) Tetracyclines (liver) Amphenicols (liver) Sulphonamides (liver)	1600	1400	170	5	25	
B2 Other veterinary drugs						

Compounds	Fish	Atlantic salmon	Rainbow trout	Atlantic halibut	Arctic char	Turbot
Emamectin	585	515	70			
Cypermethrin Deltamethrin	610	550	55		5	
Diflubenzuron Teflubenzuron Hexaflumeron Lufenuron	560	505	50		5	
Fluazuron	340	315	25			
Ivermectin Abamectin Doramectin Eprinomectin Moxidectin	80	65	15			
Praziquantel Fenbendazole	505	470	35			
Isoeugenol Eugenol	200	185	15			
B3a Organochlorine compounds						
Pesticides	495	450	45			
Dioxin and dl-PCBs PCB-6	425	370	50	5		
B3b Organophosphorous compounds						
Azamethiphos Dichlorvos	250	235	15			
B3c Chemical elements						
Lead Cadmium Mercury Arsenic Cobalt Chromium Copper Iron Manganese Molybdenum Nickel Selenium Silver Vanadium Zinc	305	260	25	5	10	5
Inorganic arsenic Methylmercury	105	95	10			
Tributyltin	250	215	30			5
B3d Mycotoxins						
Beauvericin Enniatin	500	460	40			
B3e Dyes						
Malachite green Crystal violet Brilliant green *	455	425	30			

Compounds	Fish	Atlantic salmon	Rainbow trout	Atlantic halibut	Arctic char	Turbot
B3f Others						
PBDE	350	305	40	5		
HBCD and TBBPA	355	330	25			
PAH	355	335	15		5	
PFC	350	335	10		5	
Ethoxyquin	390	355	30		5	

Some of the samples collected have been analysed by more than one method. Therefore, the total of fish in this table will be higher than the number of fish collected.

* According to directive 96/23, malachite green, crystal violet and brilliant green belongs to the group B3e. However, these dyes are not allowed to be used for food producing animals, therefore samples analysed for dyes have been collected as both group A samples (illegal drugs) and group B samples (dyes).

3 - Results

3.1 - Substances with anabolic effects and unauthorized substances

3.1.1 - Stilbenes

A total of 166 pooled samples from Atlantic salmon, rainbow trout and Atlantic halibut were examined for presence of stilbenes. None of the included stilbenes were detected in the samples analysed.

3.1.2 - Steroids

The presence of steroids was examined in 166 pooled fillet samples of Atlantic salmon and rainbow trout. None of the substances were detected in the samples analysed.

3.1.3 - Unauthorized veterinary drugs

Totally 665 pooled samples were analyzed for unauthorized veterinary drugs, including chloramphenicol, nitrofurans, metronidazole and dyes (malachite green, crystal violet, brilliant green). No residues of the included substances were detected in any of the samples.

3.2 - Veterinary drugs

Samples analysed for veterinary drugs were collected from fish at processing plants and are representative of fish ready for human consumption. The maximum residue limit for veterinary drugs is defined for muscle and skin in natural proportions (EU 37/2010). Therefore, according to the analytical protocol, any detection of drug residues in the muscle or liver would be followed by a re-analysis of the backup sample, consisting of muscle and skin in natural proportions, in duplicate.

3.2.1 - Group B1, Antibacterial agents

The antibacterial agents were determined by a combination of the three-plate bioassay and chemical methods. The broad groups a) quinolones, b) amphenicols and tetracyclines and c) sulphonamides were measured in pooled liver samples from 1600 fish. Oxytetracyclin (21 pooled samples) and florfenicol, flumequin, oxolinic acid, enrofloxacin, ciprofloxacin and trimethoprim (90 pooled samples) were also analysed by chemical methods. No residues were detected in any of the analysed samples. The LOQs of the respective compounds are listed in Table 6.3.

3.2.2 - Group B2a, Anthelmintics

The levels of the anthelmintics; teflubenzuron, diflubenzuron, hexaflumenuron, lufenuron, fluazuron, cypermethrin, deltamethrin, emamectin, ivermectin, praziquantel and fenbendazole were determined in 508 pooled muscle samples representing 2540 fish. Emamectin was detected in three out of 117 pooled samples of Atlantic salmon, at concentrations of 3, 14 and 15 µg/kg. This concentration is below the MRL of 100 µg/kg (EU 37/2010). Residues of the anti-sea-lice agent lufenuron was found in two samples of Atlantic salmon, at concentrations of 22 and 57 µg/kg, respectively. The MRL for lufenuron is 1350 µg/kg (EU 37/2010). No residues were detected for the other anthelmintics. LOQs for the substances are specified in Table. 6.3.

3.2.3 - Group B3b, Organophosphorous compounds

The levels of the B3b substances azamethiphos and dichlorvos were determined in 47 and 3 pooled fillet samples of Atlantic salmon and rainbow trout, respectively. No residues of these agents were detected in any of the examined samples.

3.2.4 - Group B3d, Sedatives

No residues of isoeugenol were detected in any of the 40 samples analysed for this sedative.

3.3 - Contaminants

Samples analysed for contaminants were collected from fish at processing plants and are representative of fish ready for human consumption.

3.3.1 - Group B3a, Organochlorine compounds

The levels of organochlorine compounds were determined in 254 pooled samples. The results are summarised in Table 3.1 to 3.3.

3.3.1.1 - Organochlorine pesticides

For several of the pesticides, the amount present is calculated as a sum including metabolites or transformation products (EU GD SANTE 2017). The results for these groups of pesticides are presented in Table 3.1. To calculate the sum of the components, conversion factors (Table 6.4) are used to adjust for different molecular weights (EU GD SANTE 2017). The sums in Table 3.1. were calculated according to the upper bound (UB) formula. When using UB calculations, the numerical value of LOQ is substituted for analytes with levels below LOQ. UB represents a “worst case scenario”. As an example, all measurements of endosulfan are below LOQ, however, a sum is generated based on the LOQ-values.

Table 3.1. The sum of groups of pesticides (µg/kg w.w.) in fillets of farmed fish

		Atlantic salmon	Rainbow trout
Sum	*n*	90	9
DDT	Median (UB)	5.6	6.1
	Max (UB)	24	11
Endosulfan	Median (UB)	0.85	0.85
	Max (UB)	1.3	0.87
Aldrin and dieldrin	Median (UB)	1.1	1.2
	Max (UB)	4.9	1.9
Chlordane	Median (UB)	0.87	0.83
	Max (UB)	2.6	1.4
Heptachlor	Median (UB)	0.43	0.47
	Max (UB)	1.1	0.68
Toxaphene	Median (UB)	2.2	2.3
	Max (UB)	6.0	4.8

Of the groups of pesticides calculated from a sum of several component, the highest values measured in Atlantic salmon fillet were 24 µg/kg w.w of DDT, with a contribution from mostly p,p`-DDE (15 µg/kg w.w), and 6 µg/kg w.w Toxaphene. DDT and Toxaphene were also the highest measured concentrations in rainbow trout, with 11 and 4.8 µg/kg w.w, respectively.

The results for the other pesticides are summarised in Table 3.2. Hexachlorbenzene and trans-nonachlor were present in concentrations above LOQ in most of the samples. The highest levels measured were 4.2 µg/kg w.w. hexachlorobenzene and 2.4 µg/kg w.w. of trans-nonachlor in Atlantic salmon samples. There are currently no MRLs established in fish fillet for any of the listed pesticides (EU 2014).

Table 3.2. Pesticides (µg/kg w.w.) in fillets of farmed fish.

Pesticide		Atlantic salmon	Rainbow trout	LOQ
	n	90	9	
α-Hexachlorocyclo-hexane	#Values	11	2	
	Median	LOQ	LOQ	
	Max	0.33	0.18	0.13-0.60
β-Hexachlorocyclo-hexane	#Values	4	1	
	Median	LOQ	LOQ	
	Max	0.26	0.23	0.13-0.60
γ-Hexachlorocyclo-hexane	#Values	2	0	
	Median	LOQ	LOQ	
	Max	0.18	LOQ	0.13-0.60
δ-Hexachlorocyclo-hexane	#Values	0	0	
	Median	LOQ	LOQ	
	Max	LOQ	LOQ	0.13-0.60
Hexachlorobenzene	#Values	89	9	
	Median	1.3	1.3	
	Max	4.2	2.4	0.06-1.0
Pentachlorobenzene	#Values	12	1	
	Median	LOQ	LOQ	
	Max	0.65	0.46	0.30-1.2
Trans-Nonachlor	#Values	89	9	
	Median	0.70	0.84	
	Max	2.4	1.4	0.13-0.60
Endrin	#Values	2	0	
	Median	LOQ	LOQ	
	Max	0.30	LOQ	0.15-0.71
Mirex	#Values	14	2	
	Median	LOQ	LOQ	
	Max	0.15	0.07	0.05-0.24
Octachlorstyrene	#Values	54	7	
	Median	0.09	LOQ	
	Max	0.32	0.15	0.03-0.12

3.3.1.2 - Dioxin, dl-PCBs and PCB-6

The levels of dioxin (PCDD+PCDF), dl-PCBs and PCB-6 in farmed fish are shown in Table 3.3. The data is mainly represented by Atlantic salmon, but also samples from rainbow trout and Atlantic halibut were examined.

The sums of dioxins, dioxins + dl-PCBs and PCB-6 are calculated as upper bound (EU 1259/2011). Accordingly, the numerical LOQ values were used for congeners with levels below LOQ.

The levels of dioxins and dl-PCBs are reported as ng toxic equivalents 2005 (TEQ05)/kg and represent the sum of 17 different PCDD/F and 12 dl-PCBs where each congener was multiplied by a Toxic Equivalency Factor (TEF). TEF values are determined by WHO, and the toxicity of each congener is expressed relative to the most toxic form of dioxin, 2,3,7,8-TCDD which has a TEF value of 1 (EU 1259/2011) .

For salmon, the median of the sum of dioxins was 0.25 ng TEQ/kg w.w. The maximum value was 0.39 ng TEQ/kg w.w., which is below the EU maximum level of 3.5 ng TEQ/kg w.w.

The median of the sum of all 29 PCDD/F and dl-PCBs was 0.54 ng TEQ/kg w.w for salmon and 0.55 ng TEQ/kg w.w for rainbow trout. The highest result for sum dioxin and dl-like PCBs was found in one Atlantic halibut sample at 1.5 ng TEQ/kg w.w. All measured values were below the EU maximum level of 6.5 ng TEQ/kg w.w.

The median of PCB-6 for salmon was 3.5 µg/kg w.w and 3.7 in rainbow trout. The concentration of PCB-6 in one Atlantic halibut sample was 13 µg/kg w.w, which was the highest concentration PCB-6 measured. The EUs maximum level for PCB-6 in fish is 75 µg/kg w.w..

Table 3.3. Dioxins, dl-PCBs and PCB-6 in fillets of farmed fish.

		Atlantic salmon	Rainbow trout	Atlantic halibut	Maximum level
	n	74	10	1	
Sum dioxins (ng TEQ/kg w.w.)	Median	0.25	0.26	-	
	Max	0.39	0.30	0.45	3.5
Sum dioxin + dl-PCBs (ng TEQ/kg w.w.)	Median	0.54	0.55	-	
	Max	0.90	0.76	1.5	6.5
	Samples	74	10	1	
PCB-6 (µg/kg w.w.)	Median	3.5	3.7	-	
	Max	7.7	6.3	13	75

3.3.2 - Group B3c, Chemical elements

In 2019, monitoring of the levels of chemical elements, such as arsenic (and inorganic arsenic), total mercury in addition to methylmercury, cadmium, lead and mono-, di- and tributyltin included samples of Atlantic salmon, Rainbow trout, Atlantic halibut, Atlantic char and turbot.

The EU maximum level for mercury is 0.50 mg/kg w.w. for the species analyzed in this report, except Atlantic halibut which have a EU maximum level of 1.0 mg/kg w.w. (EU 1881/2006). Total mercury levels above LOQ were found in all measured samples, with the highest concentrations of 0.07 mg/kg w.w. in rainbow trout and halibut and 0.06 mg/kg w.w. in salmon (Table 3.4). Thus, the concentrations measured in all samples were below the maximum level.

In addition to mercury, methylmercury was determined in 21 samples. The result showed that the levels of methylmercury (Table 6.1) were similar to the level of mercury in the same samples.

The concentrations of cadmium in most samples analyzed since 2002 have been lower than the LOQ. In 2019, a single sample of turbot was found to contain a cadmium concentration above the LOQ. With a level of 0.005 mg/kg w.w. in the

fillet, the measured concentration was well below EUs maximum level of 0.05 mg/kg w.w. (EU 1881/2006).

Arsenic is determined as “total arsenic”, comprising the sum of all arsenic species. The median level of total arsenic in Atlantic salmon was 0.58 mg/kg w.w., and the highest concentration measured was 2.1 mg/kg w.w. (Table 3.4). As in the previous year, none of the samples had concentrations of inorganic arsenic above the LOQ (Table 6.1), indicating that arsenic in fish is present mainly as organo-arsenic compounds of low toxicity (Shiomi 1994). There is currently no EU upper limit for neither total arsenic nor inorganic arsenic in fish fillets.

The EU maximum level for lead in muscle meat of fish is currently set at 0.30 mg/kg w.w. (EU 1881/2006). Of the 61 pooled samples analyzed, a lead concentration above LOQ was found in a single salmon sample. The measured concentration was 0.023 mg/kg w.w., and thus well below the EU-level.

In 2019, the concentrations of 11 additional chemical elements were included into the surveillance. Silver or molybdenum was not detected in any of the analysed samples (Table 3.4). Cobalt and nickel above LOQ were found in one and four of the salmon samples, respectively, with maximum concentrations of 0.023 mg cobalt/kg fillet and 0.71 mg nickel/kg fillet. Chromium was detected in less than 50% of the samples and had a maximum concentration of 1.7 mg/kg in salmon. Copper, iron, manganese, selenium, vanadium and zinc were found at levels above LOQ in almost all samples. With the exception of zinc, where the maximum concentration of 9 mg/kg was measured in one sample of turbot, the maximum concentrations of both copper, iron, manganese, selenium and vanadium were found in salmon, and were 0.7 mg/kg, 11 mg/kg, 0.27 mg/kg, 0.35 mg/kg and 0.029 mg/kg, respectively. There is currently no EU-limit established for any of the newly included elements.

Mono-, di- and tributyltin was monitored in a total of 50 samples. The monitoring included mostly salmon samples, but also rainbow trout and turbot fillet were analyzed. There is currently no EU upper limit for tin in fish fillet. Mono- and dibutyltin were detected in 31 and 39 of the 50 samples, respectively. However, none of the samples contained mono- or dibutyltin levels above the LOQ. Tributyltin was detected in 41 of the analyzed samples. A total of 15 samples contained tributyltin above the LOQ, with the highest measured level of 0.3 µg/kg w.w. found in both rainbow trout and turbot.

Table 3.4. Chemical elements in fillets of farmed fish.

Element		Atlantic salmon	Rainbow trout	Atlantic halibut	Arctic char	Turbot	LOQ	Maximum level
	n	52	5	1	2	1		
Mercury (mg/kg w.w.)	#Values	52	5	1	2	1		
	Median	0.02	0.05	LOQ	LOQ	LOQ		
	Max	0.06	0.07	0.07	0.05	0.05	0.002	0.50
Arsenic (mg/kg w.w.)	#Values	52	5	1	2	1		
	Median	0.58	1.3	LOQ	LOQ	LOQ		
	Max	2.1	2.0	2.3	2.3	2.2	0.003	n.a.
Cadmium (mg/kg w.w.)	#Values	0	0	0	0	1		
	Median	-	-	-	-	LOQ		
	Max	LOQ	LOQ	LOQ	LOQ	0.005	0.001-0.002	0.05
Lead (mg/kg w.w.)	#Values	1	0	0	0	0		
	Median	LOQ	-	-	-	-		
	Max	0.023	LOQ	LOQ	LOQ	LOQ	0.006-0.01	0.30
Cobalt (mg/kg w.w.)	#Values	1	0	0	0	0		
	Median	LOQ	-	-	-	-		
	Max	0.023	LOQ	LOQ	LOQ	LOQ	0.006-0.009	n.a.
Chromium (mg/kg w.w.)	#Values	20	2	1	1	1		
	Median	LOQ	LOQ	LOQ	LOQ	LOQ		
	Max	1.7	0.035	0.009	0.058	0.009	0.006-0.01	n.a.
Copper (mg/kg w.w.)	#Values	52	5	1	2	1		
	Median	0.4	0.4	LOQ	LOQ	LOQ		
	Max	0.7	0.4	0.2	0.4	0.2	0.1	n.a.
Iron (mg/kg w.w.)	#Values	52	5	1	2	1		
	Median	2.8	2.9	LOQ	LOQ	LOQ		
	Max	11	3.5	0.85	2.1	0.79	0.1	n.a.
Manganese (mg/kg w.w.)	#Values	52	5	1	2	1		
	Median	0.077	0.078	LOQ	LOQ	LOQ		
	Max	0.27	0.081	0.099	0.061	0.55	0.03	n.a.
Molybdenum (mg/kg w.w.)	#Values	0	0	0	0	0		
	Median	-	-	-	-	-		
	Max	LOQ	LOQ	LOQ	LOQ	LOQ	0.01-0.4	n.a.
Nickel (mg/kg w.w.)	#Values	4	0	0	0	0		
	Median	LOQ	-	-	-	-		
	Max	0.71	LOQ	LOQ	LOQ	LOQ	0.07-0.1	n.a.
Selenium (mg/kg w.w.)	#Values	52	5	1	2	1		
	Median	0.16	0.23	LOQ	LOQ	LOQ		
	Max	0.35	0.27	0.26	0.18	0.2	0.01	n.a.

Element		Atlantic salmon	Rainbow trout	Atlantic halibut	Arctic char	Turbot	LOQ	Maximum level
Silver (mg/kg w.w.)	#Values	0	0	0	0	0		
	Median	-	-	-	-	-		
	Max	LOQ	LOQ	LOQ	LOQ	LOQ	0.002-0.004	n.a.
Vanadium (mg/kg w.w.)	#Values	50	2	0	2	0		
	Median	0.007	LOQ	-	LOQ	-		
	Max	0.029	0.012	LOQ	0.003	LOQ	0.001-0.002	n.a.
Zinc (mg/kg w.w.)	#Values	52	5	1	2	1		
	Median	3.9	3.5	LOQ	LOQ	LOQ		
	Max	6.8	3.9	3.7	4.2	9	0.5	n.a.
	n	43	6			1		
Monobutyltin (µg Sn/kg w.w.)	#Values	0	0			0		
	Median	-	-			-		
	Max	LOQ	LOQ			LOQ	0.4-1	n.a.
Dibutyltin (µg Sn/kg w.w.)	#Values	0	0			0		
	Median	-	-			-		
	Max	LOQ	LOQ			LOQ	0.2-0.5	n.a.
Tributyltin (µg Sn/kg w.w.)	#Values	9	5			1		
	Median	LOQ	0.1			LOQ		
	Max	0.1	0.3			0.3	0.06-0.09	n.a.

3.3.3 - Group B3d, Mycotoxins

The mycotoxins enniatin A, enniatin A1, enniatin B, enniatin B1 and beauvericin were measured in 92 pooled samples of salmon and 8 pooled samples of rainbow trout. No residues of these mycotoxins were detected in any of the samples.

3.3.4 - Group B3e, Dyes

In addition to 167 samples analysed for residues of dyes as group A (illegal drugs) samples, dyes were measured in 91 pooled group B (contaminants, dyes) samples. No residues of malachite green, crystal violet and brilliant green were detected in any of the group B samples.

3.3.5 - Group B3f, others

The group B3f, others is a group not required for finfish products by the directive 96/23/EC, but are deemed relevant for analyses in Norwegian aquaculture by the NFSA (Mattilsynet) and IMR, because these undesirable compounds are present in the environment and may affect food safety. This group currently consist of brominated flame retardants (BFR), perfluorinated compounds (PFC), polyaromatic hydrocarbons (PAHs), and since 2018 also the technological feed additive ethoxyquin (EQ) and its main transformation product ethoxyquin dimer (EQDM).

3.3.5.1 - Brominated flame retardants

PBDE, TBBPA and HBCD are compounds previously used as flame retardants and widely spread in the environment. The summarised PBDE-7 (28, 47, 99, 100, 153, 154, 183) and in addition PBDE 66, 119 and 138 are shown in Table 3.5. The highest level of PBDE-7 was found in a sample of Atlantic halibut. Median values of PBDE-7 were 0.36 µg/kg w.w. and 0.33 µg/kg w.w for salmon and rainbow trout, respectively. Of 66 pooled Atlantic salmon samples and five

pooled rainbow trout samples, TBBPA concentrations were detected and quantified in three samples of salmon. The highest measured TBBPA value was 0.12 µg/kg w.w.. HBCD was analysed in 66 salmon fillet samples and five rainbow trout fillet samples. Median values for salmon and rainbow trout were 0.13 and 0.09 µg/kg w.w., respectively, with the highest concentration of 0.32 µg/kg w.w. found in salmon. There are currently no EU maximum levels for BFRs in food.

Table 3.5 . BFR (µg/kg w.w.) in fillets of farmed fish.

		Atlantic salmon	Rainbow trout	Atlantic halibut	LOQ
	n	61	8	1	
UB-Sum PBDE 7	Median	0.36	0.33	-	
	Max	0.83	0.56	1.2	
PBDE 66	#Values	61	8	1	
	Median	0.008	0.007	-	
	Max	0.020	0.009	0.028	0.002-0.01
PBDE 119	#Values	21	2	1	
	Median	LOQ	-	-	
	Max	0.007	0.003	0.009	0.002-0.01
PBDE 138	#Values	0	0	0	
	Median	-	-	-	
	Max	LOQ	LOQ	LOQ	0.003-0.02
	n	66	5	0	
TBBPA	#Values	3	0		
	Median	LOQ	-		
	Max	0.12	LOQ		0.03-0.14
	n	66	5	0	
UB-Sum HBCD (α,β,γ)	Median	0.13	0.09		
	Max	0.32	0.19		

3.3.5.2 - Perfluorinated compounds

A total of 70 samples were analysed for the PFCs. All results were below the LOQ (Table 6.3). EU has currently no maximum level for PFC in fish.

3.3.5.3 - Polycyclic aromatic hydrocarbons

PAH was analysed in 71 samples, of which 67 samples were from salmon, three from rainbow trout and one from Arctic char. The results for PAH are summarised in Table 3.6. There is no maximum level for PAH in fresh fish (EU 835/2011).

Table 3.6. PAH (µg/kg w.w.) in fillets of farmed fish.

PAH		Atlantic salmon	Rainbow trout	Arctic char	LOQ
	n	67	3	1	
5-methylchrysene	#Values	0	0	0	0.09 - 0.13
	Max	LOQ	LOQ	LOQ	
Benz(a)anthracene	#Values	6	0	0	0.09 - 0.13
	Max	0.6	LOQ	LOQ	
Benzo(a)pyrene	#Values	2	0	0	0.09 - 0.13
	Max	0.5	LOQ	LOQ	
Benzo(b)fluoranthene	#Values	3	0	0	0.09 - 0.13
	Max	0.5	LOQ	LOQ	
Benzo(c)fluorine	#Values	1	1	0	0.09 - 0.13
	Max	0.1	0.7	LOQ	
Benzo(ghi)perylene	#Values	3	0	0	0.09 - 0.13
	Max	0.5	LOQ	LOQ	
Benzo(j)fluoranthene	#Values	0	0	0	0.09 - 0.13
	Max	LOQ	LOQ	LOQ	
Benzo(k)fluoranthene	#Values	0	0	0	0.09 - 0.13
	Max	LOQ	LOQ	LOQ	
Chrysene	#Values	11	0	0	0.09 - 0.13
	Max	0.8	LOQ	LOQ	
Cyclopenta(cd)pyrene	#Values	5	0	0	0.09 - 0.13
	Max	0.5	LOQ	LOQ	
Dibenz(ah)anthracene	#Values	0	0	0	0.09 - 0.13
	Max	LOQ	LOQ	LOQ	
Dibenzo(a,e)pyrene	#Values	0	0	0	0.44 – 0.66
	Max	LOQ	LOQ	LOQ	
Dibenzo(a,h)pyrene	#Values	0	0	0	0.44 – 0.66
	Max	LOQ	LOQ	LOQ	
Dibenzo(a,i)pyrene	#Values	0	0	0	0.44 – 0.66
	Max	LOQ	LOQ	LOQ	
Dibenzo(a,l)pyrene	#Values	0	0	0	0.44 – 0.66
	Max	LOQ	LOQ	LOQ	
Indeno(1,2,3,-cd)pyrene	#Values	1	0	0	0.09 - 0.13
	Max	0.3	LOQ	LOQ	

3.3.5.4 - Ethoxyquin

EQ and EQDM levels were measured in 78 pooled fillet samples. The samples were mostly taken from Atlantic salmon, but also samples from rainbow trout and Atlantic char were included (Table 3.7). The fillet concentrations of EQ and EQDM were calculated as upper bound (UB), using the numerical LOQ values (0.001 and 0.005 mg/kg ww, respectively) for measurements below LOQ. The number of samples with measurements above LOQ is indicated in Table 3.7 as the number of values.

For salmon samples, the median level of the sum of EQ & EQDM was 0.02 mg/ kg ww. Rainbow trout contained sum EQ & EQDM at a median concentration of 0.016 mg/kg ww. One sample of Atlantic char was analysed, which did not contain levels above the LOQ.

The maximum values of EQ and EQDM were 0.02 and 0.35 mg/kg ww, respectively, and were found in salmon.

Table 3.7. Ethoxyquin and ethoxyquin dimer in fillets of farmed fish.

		Atlantic salmon	Rainbow trout	Atlantic char	LOQ
	n	71	6	1	
EQ (mg/kg ww)	#Values	19	2	0	
	Median	LOQ	-	-	
	Max	0.02	0.004	LOQ	0.001
EQDM (mg/kg ww)	#Values	62	6	0	
	Median	0.02	0.02	-	
	Max	0.35	0.18	LOQ	0.005
Sum EQ&EQDM (mg/kg ww) UB					
	Median	0.02	0.02	-	
	Max	0.36	0.18	0.006	

4 - Discussion

4.1 - Unauthorized substances

No residues of unauthorized substances were detected in any of the analysed samples.

4.2 - Veterinary drugs

Most samples reviewed in this report are from fillets of farmed fish. However, as the liver has a central function in the distribution and elimination of veterinary drugs, liver samples were analysed for antibiotics. Even though the bioassay used for the antibacterial agents is less sensitive than the chemical analytical methods, the higher concentrations of antibacterial agents in liver compared to fillet enhance the ability to detect any residues. Moreover, the ability of the bioassay to detect a wider range of antibiotics than the more specific chemical methods renders the method useful for screening purposes. Any positive detection by the inhibition assay is verified by chemical analysis of the corresponding fillet sampled from the same fish. Consistent with the data gathered in the monitoring of recent years, no residues of antibiotics, endoparasitic agents or dyes were detected in any of the samples.

In 2019, residues of the anti-sea-lice agents emamectin and lufenuron were detected in three and two samples of Atlantic salmon, respectively. However, the rate of positive samples is low and the concentrations are well below the MRLs for both emamectin and lufenuron.

No residues of antibiotics, endoparasitic agents or sedatives were detected in any samples taken during 2019.

4.3 - Contaminants

Although the level of dioxins and dl-PCBs decreased from 2006 until 2012, reflecting the increased substitution of marine ingredients with vegetable ingredients in the feed, the level appears to have stabilized at approximately 0.5 ng TEQ/kg w.w. in farmed Atlantic salmon. This level has been rather stable from 2012 up to, and including, 2019.

As in the previous year, in 2019 no environmental contaminants were found above the EU maximum levels (MLs), for those contaminants where MLs have been implemented. However, the EUs MLs for food are not based on toxicological data, but derived from the ALARA (as low as reasonably achievable) principle, with the aim to prevent those commodities with the highest contaminant levels to reach the market. For all contaminants included in the programme, including brominated flame retardants (PBDEs, HBCDs and TBBP), PAHs, PCB-6 and pesticides, the concentrations were comparable to the years before.

Starting in 2019, in addition to the four heavy metals (Hg, As, Pb and Cd) traditionally included, the surveillance of chemical elements in fish fillet was extended with 11 other elements: cobalt, chromium, copper, iron, manganese, molybdenum, nickel, silver, selenium vanadium and zinc. There are currently no MLs established for any of the added elements in fish.

The levels of DDT were comparable to the years before.

4.4 - Ethoxyquin

Following the suspended authorization of the use of ethoxyquin as an anti-oxidant feed additive (EU 2017/962), specific transitional measures were settled for compound feed produced with mostly marine-derived feed material containing ethoxyquin and premixtures containing ethoxyquin (Article 3, EU 2017/962) due to a lack of suitable alternatives. These compound feeds were permitted to be placed on the marked until 31.03.2020 and used no longer than three months after this date.

The implementation of the regulation by the industry is clearly reflected the by decrease in fillet concentration of both

ethoxyquin and its main transformation product the ethoxyquin dimer. In 2018, ethoxyquin and ethoxyquin dimer were found above LOQ in 61 and 99% of the analysed salmon fillet samples, respectively. In 2019, only 27% and 87% of the sampled salmon fillets had levels of ethoxyquin and ethoxyquin dimer, respectively, above LOQ. In addition, median values declined from 0.002 and 0.11 mg/kg ww to LOQ (0.001 mg/kg ww) and 0.002 mg/kg ww for EQ and EQDM, respectively.

Due to limited data on toxicity of EQ and its metabolites, a precautional maximum residue level (MRL) at the limit of analytical quantification (0.05 mg/kg), is currently applied in the EU (EU 2014). The list of products where a MRL has been established includes several products of animal origin. The median concentration of the sum EQ & EQDM does not exceed the currently set precautional MRL value. However, no MRL has yet been defined for fish.

5 - Conclusion

No substances with anabolic effect were detected in any of the samples analysed.

None of the veterinary drugs were detected at levels exceeding the MRL established for fish. Emamectin or lufenuron were detected in a total of five samples, but the measured levels were below their respective MRLs.

For contaminants, no samples exceeded the EUs maximum levels, where such levels have been established (sum dioxins, sum dioxins and dl-PCBs, PCB-6, mercury, lead and cadmium).

6 - Tables

Table 6.1. Inorganic arsenic and methylmercury in fillets of farmed fish

		Atlantic salmon	Rainbow trout	LOQ
	n	19	2	
Inorganic arsenic (µg/kg w.w.)	#Values	0	0	
	Median	-	-	
	Max	LOQ	LOQ	2-3
Methyl-mercury (mg Hg/kg w.w.)	#Values	19	2	
	Median	0.02	-	
	Max	0.05	0.03	0.001

Table 6.2. PFCs (µg/kg w.w.) in fillets of farmed fish

Compound	Atlantic salmon	Rainbow trout	Arctic char	Max value	LOQ
PFBA	67	2	1	<LOQ	1.0
PFBS					1.0
PFDA					0.2
PFDoDA					0.2
PFDS					0.2
PFHpA					0.2
PFHxA					0.5
PFHxS					1.0
PFNA					0.2
PFOA					0.6
PFOS					0.2
PFOSA					0.5
PFTeDA					0.2
PFTrDA					0.2
PFUdA					0.2

Table 6.3. Summary of analytical methods.

Group of substances	Analyte	Method	LOD (µg/kg w.w.)	LOQ (µg/kg w.w.)	Level of action (µg/kg w.w.)	Laboratory
A1 Stilbenes	Diethylstilbestrol	LC-MS/MS	1		Presence	Eurofins
	Dienestrol		1			
	Hexestrol		1			
	B-Estradiol		1			
	α-Estradiol		1			
	Estriol		1			
	Estrone		1			
	Ethinyl estradiol		1			
A3 Steroids	α-nandrolon	LC-MS/MS	1		Presence	Eurofins
	β-nandrolon		1			
	α-trenbolon		1			
	β-trenbolon		1			
	Trenbolone-acetate		2			
	16-Hydroxy stanozolol		1			
	α -Boldenone		1			
	Boldenone		1			
	Chlor-Testosterone (Clostebol)		1			
	Epitestosterone		1			
	Methyl-Boldenone (Dianabol)		1			
	Methyltestosterone		1			
	Nortestosterone/ Nandrolone		1			
	Stanozolol		1			
	Testosterone		1			
	Testosterone-propionate		2			
A6 Annex IV substances	Chloramphenicol	LC-MS/MS	0.25		Presence (MRPL = 0.3)	IMR
	Metronidazole	LC-MS/MS	0.3		Presence (MRPL = 3.0)	
	Hydroxy-metronidazole		2.0			
	Nitrofuran AOZ	LC-MS/MS	0.5		Presence (MRPL =1.0)	
	Nitrofuran AHD		0.6		Presence (MRPL =1.0)	
	Nitrofuran AMOZ		0.4		Presence (MRPL =1.0)	
	Nitrofuran SEM		0.5		Presence (MRPL= 1.0)	

Group of substances	Analyte	Method	LOD (µg/kg w.w.)	LOQ (µg/kg w.w.)	Level of action (µg/kg w.w.)	Laboratory
B1 Antibacterial Substances Micro-biological method	Quinolones	3-plate Screening Method2	200		100-600	IMR
	Tetracyclines		200		100	
	Amphenicols		200		1000	
	Sulfonamides		400		100	
B1 Antibacterial substances Chemical method	Oxolinic acid	LC-MS/MS		40	100	IMR
	Flumequine			40	600	
	Enrofloxacin			10	100	
	Ciprofloxacin			10	100	
	Trimethoprim			2	50	
	Oxytetracycline	LC-MS/MS		30	100	Eurofins
	Florfenicol	LC-MS/MS		0.5	1000	IMR
B2a Anthelmintics	Praziquantel	LC-MS/MS		1	n.a.	IMR/ Eurofins
	Fenbendazole	LC-MS/MS		1	n.a.	
	Emamectin	LC-MS/MS		2-10	100	
	Diflubenzuron	LC-MS/M		1-10	10	
	Teflubenzuron			1-50	500	
	Hexaflumuron			1-50	500	
	Lufenuron			1-50	1350	
	Ivermectin	LC-MS/M		2	n.a.	Eurofins
	Cypermethrin	GC-MS		5	50	
	Deltamethrin			10	10	
	Isoeugenol	GC-FID		50	6000	
B3a Organo-chlorine compounds	Dioxins and dlPCB	HRGC-HRMS		0.0001-0.1 ng TEQ/kg	6.5 ng TEQ/kg	IMR
	PCB-6	GC-MS GC-MS/MS		0.004 – 0.5	75	
	Pesticides	HRGC-HRMS		0.003-0.8	n.a.	Eurofins
B3b Organo-phosphorus compounds	Azametiphos	LC-MS/MS		10	n.a.	Eurofins
	Dichlorvos					
	Lead	ICP-MS		0.005- 0.01 mg/kg	0.3 mg/kg	
	Cadmium			0.001- 0.002 mg/kg	0.05 mg/kg.	
	Arsenic			0.003 mg/kg	n.a.	
	Mercury			0.002 mg/kg	0.5 mg/kg	
	Cobalt			0.006-0.009 mg/kg	n.a.	
	Chromium			0.006- 0.01 mg/kg	n.a.	
	Copper			0.1 mg/kg	n.a.	
	Iron			0.1 mg/kg	n.a.	

Group of substances B3c Chemical elements	**Analyte**	**Method**	**LOD (µg/kg w.w.)**	**LOQ (µg/kg w.w.)**	**Level of action (µg/kg w.w.)**	**Laboratory** IMR
	Manganese			0.03 mg/kg	n.a.	
	Molybdenum			0.01- 0.4 mg/kg	n.a.	
	Nickel			0.07- 0.1 mg/kg	n.a.	
	Selenium			0.01 mg/kg	n.a.	
	Silver			0.002- 0.004 mg/kg	n.a.	
	Vanadium			0.001- 0.002 mg/kg	n.a.	
	Zinc			0.5 mg/kg	n.a.	
	Inorganic arsenic	LC-ICP-MS		4-6	n.a.	
	Methylmercury	GC-ICP-MS		1	n.a.	
	Tributyltin	GC-ICP-MS		0.3-0.5	n.a.	
B3d Mycotoxins	Beauvericin, Enniatin A, A1, B and B1	LC-MS/MS		10	n.a.	Eurofins
B3e, dyes	Malachite green	LC-MS/MS	0.15		Presence (MRPL=2)	IMR
	Leuco malachite green		0.15			
	Crystal violet		0.30		Presence	
	Leuco crystal violet		0.15		Presence	
	Brilliant green		0.15		Presence	
B3f, others	PBDE	GC-MS		0.003-0.009	n.a.	IMR
	HBCD	LC-MS/MS		0.006-0.01	n.a.	Eurofins
	TBBPA	GC-MS		0.03-0.2	n.a.	Eurofins
	PAH	GC-MS/MS		0.5-1.0	n.a.	IMR
	PFC	LC-MS/MS		0.5-13	n.a.	IMR
	Ethoxyquin	HPLC-FLD		0.001	n.a.	IMR
	Ethoxyquin dimer			0.005	n.a.	

1 All methods used muscle as sample matrix except for microbiological methods for antibacterial substances (B1), were liver was used 2 Only screening method, positive results have to be confirmed by a chemical method.

Table 6.4. Calculation of sums for certain pesticides.

Sum	Substances included in the sum	Conversion factor
DDT (sum of p,p-DDT, o,p-DDT, p,p-DDD, o,p-DDD, p,p-DDE,and o,p-DDE expressed as DDT)	op-DDT	1
	pp-DDT	1
	op-DDD	1.108
	pp-DDD	1.108
	op-DDE	1.115
	pp-DDE	1.115
Endosulfan (sum of alpha- and beta-isomers and endosulfan-sulphate expressed as endosulfan)	alpha-endosulfan	1
	beta-endosulfan	1
	endosulfan sulphate	0.962
Aldrin and dieldrin (Aldrin and dieldrin combined expressed as dieldrin)	dieldrin	1
	aldrin	1.044
Chlordane (Sum of cis- and trans-isomers and oxychlordane expressed as chlordane)	trans-chlordane	1
	cis-chlordane	1
	oxychlordane	0.967
Heptachlor (sum of heptachlor and heptachlor epoxide expressed as heptachlor)	heptachlor	1
	trans-heptachlor epoxide	0.959
	cis-heptachlor epoxide	0.959
Toxaphene (sum of toxaphene 26, toxaphene 50 and toxaphene 62)	Toxaphene 26	1
	Toxaphene 50	1
	Toxaphene 62	1

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