Screening of veterinary medicines in agricultural areas

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REPORT

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Summary

The environmental occurrence of antibiotics and parasiticides from veterinary use in areas with livestock was investigated in a screening study. The primary goal was to investigate whether these compounds were released in a magnitude that leads to detectable levels in manure, soil, waters or sediments. The results were compared to effect levels for aquatic ecosystems, in order to assess whether the release of these compounds from veterinary use may pose an environmental risk.

Samples were taken on and downstream farms with large livestocks of fattening pigs and dairy cows, and 50 different substances were analysed. Of these substances the following groups were included:

Antibiotics: tetracyclines, aminoglycosides, sulfonamides, trimethoprim, macrolides and fluoroquinolones

Parasiticides: sulfonamides, benzimidazoles, tetrahydropyrimidines and macrocylic lactones.

In total, 63 samples of surface water, surface sediment, manure, manure treated soil, and groundwater were taken in different regions in Sweden. Most sites were investigated only on one occasion, but from a small number of sites samples were collected on two occasions.

The strategy for choosing sampling sites was: 1) identify the major uses; 2) identify large farms; 3) select all farms with a topography beneficial for runoff or leaching to adjacent streams; 4) identify farms that actually use some of these pharmaceuticals (through contact with the responsible at each farm).

No single sample contained antibiotics or parasiticides in detectable levels. With the exception of ivermectin, all EC50 and NOEC values were orders of magnitude higher than the analytical reporting levels. Therefore, the data in this study suggest that the use of antibiotics and parasiticides in livestocks of dairy cows and pigs do not pose a general risk to the aquatic ecosystems or to agricultural soils.

The use and environmental occurrence of veterinary compounds were also compared to that of pesticides, a group of chemicals that are applied in similar regions and for which ample monitoring data exists. During 2005, the total Swedish sale of pesticides was ca 100 times higher than that of veterinary antibiotics and parasiticides. Annual area doses to agricultural soils were estimated, by conservatively assuming that the applied doses all end up in soil through the use of manure as fertilizer. The area doses ranged from 0.2 to 30 g active substance per hectare and year for individual substances. This corresponds to the lower range of application rates for individual pesticides.

Sammanfattning

Förekomsten av antibiotika och antiparasitära ämnen i miljöer som kan påverkas av veterinärmedicinsk användning har undersökts som en screeningstudie. Syftet var att undersöka i vilken omfattning som den veterinärmedicinska användningen av dessa ämnesgrupper orsakar en spridning till miljön, samt om denna spridning kan orsaka toxiska effekter i miljön.

Undersökningen är inriktad på gårdar med stora besättningar av slaktsvin och mjölkkor, och omfattar mätningar i flytgödsel, gödslad jord, grundvatten, ytvatten och ytsediment. Proverna är tagna på gårdar, i deras omedelbara närhet samt i vattendrag nedströms gårdarna. Flertalet av proven härrör från fyra jordbruksintensiva regioner i Sverige, och de flesta provpunkterna har provtagits vid ett tillfälle; ett mindre antal provpunkter har undersökts vid två tillfällen.

Totalt har 63 prov analyserats på 50 olika substanser, omfattande:

Antibiotika: tetracykliner, aminoglykosider, sulfonamider, trimetoprim, makrolider och fluorkinoloner

Antiparasitära ämnen: sulfonamider, bensimidasoler, tetrahydropyrimidiner och makrocykliska laktoner.

Provtagningsområden har valts utifrån följande strategi: 1) identifiera ämnenas huvudsakliga användningsområden; 2) identifiera gårdar med stora besättningar; 3) välj gårdar med topografiska förutsättningar för spridning till ytvatten; 4) identifiera enskilda gårdar där några av ämnena används.

Inget prov innehöll något ämne över rapporteringsgränsen. Bortsett från ivermektin, som är särskilt ekotoxiskt, är EC50 och NOEC-värden för enskilda substanser en eller flera tiopotenser högre än motsvarande rapporteringsgränser. Undersökningen tyder därför på att nuvarande användning av antibiotika och antiparasitära ämnen för slaktsvin och mjölkkor inte leder till en allmän miljöpåverkan i Sverige.

Användningen och resultaten jämfördes också med motsvarande för pesticider, en grupp ämnen som används i samma regioner och för vilka omfattande miljöövervakningsdata föreligger.Under 2005 var den totala försäljningen av pesticider ca 100 gånger större än motsvarande för antibiotika och antiparasitära ämnen inom jordbruket. Arealdoser för dessa veterinärmedicinska läkemedel har även skattas, genom att konservativt anta att all användning slutligen hamnar i gödslad jord. De beräknade arealdoserna låg i intervallet 0.2-30 gram aktiv substans per hektar och år. Dessa värden är i det lägre området för motsvarande dosering av pesticider, och motsvarar doser som ges för de mer toxiska pesticiderna. Mindre toxiska pesticider doseras vanligen i betydligt högre doser.

Table of Contents

L:\365x\2006\10076331 Screening Veterinärmedicinska\C-Genomförande\24-Slutversioner\Slutrapport vet_med_070531.doc

1. Introduction

The fact that pharmaceutical substances are of potential environmental concern has been widely recognized during the last ca five years, although research was performed earlier (Halling-Sørensen et al., 1998). Pharmaceutical substances are a large and chemically diverse group, but they have in common their design for being biologically active and their release pathways to the environment. To date, most studies in this field have been devoted to the environmental impact from human use. These releases mainly follow the municipal waste waters. Many pharmaceuticals have been found in sewage sludge at levels up to at least 10 mg/kg dw (e.g. Haglund and Olofsson, 2006; Andersson et al., 2006). Furthermore, certain pharmaceuticals have also been found in the recipients of municipal sewage treatment plants (e.g., Heberer, 2002; Kolpin et al., 2002; Kim and Carlson, 2007). A Swedish review of the environmental occurrence and ecotoxicity of pharmaceutical compounds was published in 2004 (MPA, 2004).

Much less is known on the environmental impact resulting from the veterinary use of pharmaceuticals. Veterinary medicines can enter the environment via several different pathways. Internationally, the major pathways are believed to be via direct discharge of aquaculture products and via the excretion of substances in urine and feces of livestock animals (Boxall et al. 2003). Other release pathways into the environment are via emissions during the manufacture, formulation, and treatment processes, and as a result of the disposal of unused medicines and their containers.

Most studies have been devoted to veterinary antibiotics, since these compounds are in large amounts in many countries (Sarmah et al., 2006). In addition to treating infections, antibiotics are widely used as growth promoters, though not in Sweden. Environmental occurrence of antibiotics due to veterinary uses has been demonstrated in surface waters, groundwaters, manure and soil and is reviewed in Sarmah et al. (2006). Occurrence in manure and manure-treated soil has been confirmed, as well as the subsequent release to groundwater and surface water due to surface runoff and leaching (e.g., Batt et al., 2006; Kay et al., 2004, 2005). For instance, levels in surface waters range from < 10 ng/l to a few extreme values in the µg/l-level (Sarmah et al., 2006; Lissemore et al., 2006; Kim and Carlson, 2007).

In Sweden, only a few aspects of the environmental impact of veterinary medicines have been investigated. The National Food Administration (www.slv.se) annually analyses medical residues in thousands of foodstuffs (e.g. Nordlander et al., 2006). In 2005, seven samples out of 15 000 contained medical residues in levels above the maximum residue limit. This is approximately equal to the number of samples exceeding the analytical detection limit. Antibiotics have never been detected in farmed fish, except for malachitegreen in a very few samples (Ingrid Nordlander, pers comm). From a pig and a cattle farm, manure as well surface water and sediment from nearby rivers were analysed for tetracyclines by Andersson et al. (2006). Only one water sample contained detectable levels (2 and 10 ng/l).

The goal of this study was to investigate the environmental occurrence of antibiotics and parasiticides in agricultural areas. Further, the aim was also to compare the results to effect levels for aquatic ecosystems, in order to assess whether the release of these compounds from veterinary use may pose an environmental risk. In total 50 different substances were studied, covering the following groups:

<u>Antibiotics</u>: tetracyclines, aminoglycosides, sulfonamides, trimethoprim, macrolides and fluoroquinolones

<u>Parasiticides</u>: sulfonamides, benzimidazoles, tetrahydropyrimidines and macrocylic lactones.

The project is an assignment from the Swedish EPA, and is part of the national environmental screening programme.

2. Use and release of veterinary medicines in Sweden

This chapter intends to describe the current Swedish use of veterinary substances. Unless specifically stated, all facts are based on Swedish board of agriculture (SBA, 2007), Hellström and Kreuger (2005) and personal contacts with Kristina Odensvik (Apoteket AB) and some district veterinary officers of the studied regions.

2.1. Overall use of veterinary medicines

On the Swedish market there are more than 1000 different active substances in 7600 different pharmaceutical products (MPA, 2004). Of these products about 7200 are used for human pharmaceuticals and about 400 for veterinary medicines. In Sweden approximately 80 tons of antibiotics is used per year (Apoteket AB, 2005). In 2003 to 2005 the total amount of antibiotics used within veterinary medicine was about 16 tons per year (Apoteket AB). There has been a drastic decline in the use of antibiotics in livestock (Apoteket AB, 2005).

According to the National Food Administration, the following groups of medicines are used for veterinary purposes in Sweden: antibiotics, parasiticides, hormones, glucocorticoids, antiinflammatory, muscle relaxing substances, pain-killers, anesthetics, and vaccines.

Of the antibacterial medicines, G- and V-penicillin’s are the most used, followed by sulfonamides, tetracyclines and macrolides and lincosamides (Apoteket AB). Of the parasiticides, tetrahydropyrimidines and bensimidazolderivate are the most used substances. Even though some compounds are used in low amounts they can have a high persistence in the environment and thereby pose a threat to the environment, such as fluoroquinolones which are lipophilic and fairly persistent compounds (e.g., Hellström and Kreuger, 2005).

Recent statistics on the veterinary use of antibiotics and parasiticides in Sweden is summarised in Table 1. It is clear that most antibiotics are used for pigs and horses, followed by cattle. For parasiticides, cattle and horses are followed by pigs. Among cattle and pigs, antibiotics are primarily given to dairy cows and fattening pigs.

According to Table 1, the amount of drugs sold by prescription for use in fish farms was almost zero in 2005. However, this does not reflect the actual use of veterinary substances in fish farms, since according to a recent report (Fiskhälsan, 2007) the use of antibiotics for farmed fish in Sweden was 17 kg in 2005 and 28 kg in 2006. In 1990 the use of antibiotics in fish farms was approximately 1000 kg, which indicates a drastic decline of the use of these substances until today.

Table 1. The amount (kg) of different veterinary drugs sold year 2005 (SBA, 2007). Substance groups included in this study is underlined.

	Amount of drugs sold by prescription					Totally (including prescription and order)	
Veterinary substance	Cattle	Pigs	Poultry	Farmed Fish	Horses	Food producing animals incl. horses	Pets and other animals
Antibacterial							
<u>Tetracyclines</u>	147	128	6,6	0	5,4	688	122
Penicillin G and V	830	759	0,01	-	212	6887	141
Aminopenicillins	5,3	90	-	-	1,5	97	840
Other betalactams	11	0,01	0,01	-	2,6	38	1116
<u>Aminoglycosides</u>	113	69	0,03	-	33	399	45
<u>Sulfonamides</u>	111	325	34	0,02	1349	2593	146
<u>Trimethoprim</u>	13	64	0,01	-	213	418	21
<u>Macrolides and Lincosamides</u>	46	701	5,4	-	6,6	835	245
<u>Fluoroquinolones</u>	19	20	0,6	-	0,6	126	59
Pleuromutilins	0	327	-	-	-	335	-
Total antibacterial	*1295*	*2483*	*47*	*0,02*	*1824*	*12416*	*2735*
Antiparasitic							
<u>Sulfonamides</u>	-	-	33	-	0,07	58	3
Triazines	0,04	4,8	-	-	0,005	5	-
Quinol derivatives and similar	0,07	-	-	-	9,3	10	109*
<u>Benzimidazoles</u>	157	94	0,4	0,01	34	573*	781*
<u>Tetrahydropyrimidines</u>	157	0,001	-	-	310	1952*	147*
Other Anthelmintic substances	-	-	-	-	-	-	17*
Pyrethrins och Pyrethroids	4	-	0,02	-	0,07	15*	219*
Other compounds against ectoparasites for external use	-	0,002	-	-	-	-	109*
Chitin inhibitors	-	-	-	-	-	-	4*
Other compounds against ectoparasites for systemic use	-	-	-	-	-	-	30
<u>Avermectines</u>	29	13	0,01	-	-	67*	10
<u>Milbemycins</u>	0,05	0	-	-	-	5*	9
Total antiparasitic	*347*	*112*	*33*	*0,01*	*353*	*2685*	*1438*

* The total amount sold of these substances also includes free merchandise or self treatment.

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2.2. Aspects of use at individual farms

2.2.1. Incitaments for use

The treatment with antibacterial medicines have drastically changed during the last ten years in Sweden due to the alarm of resistance in bacteria, demand for permits before use, more disease specific medicines etc. It used to be common that whole stocks were treated with these compounds if any animal was sick. However, today the use has decreased as well as the manner of treatment.

Now day's antibacterial veterinary medicines are mainly given at an outbreak of an illness and primarily only to those animals infected. However, at any sign of spreading of the disease the whole stock is treated. In some cases treatment with medicines are avoided if the infection can self heal. The treatment with antibacterial medicines is not restricted to certain seasons. However, the chance of infection may be larger during the winter season when animals are kept indoors tightly together. The average use of antibiotics per animal often increases at farms with larger stocks. At farms with larger stocks, antibiotics are commonly given many times per year.

Antiparasitic veterinary medicines are given to animals in preventive purpose before the animals are turned out to grass in the spring. They are almost always given to horses, swine and cattle and also to the whole stock of animals. Most commonly parasiticides are given to young cattle (ca 1 year old), which are treated before grazing and then roughly once a month during the grazing season. Parasiticides are given less frequently to pigs; however fenbandazol (Axilur) is given to pregnant sows.

During our contacts with farmers, it has become apparent that alternative strategies are used to avoid medical treatment. For instance, at one farm with a very large stock of dairy cows, the animals were never mixed with other livestock and neither were animals from other livestock bought. At a farm with fattening pigs, antiparasitic compounds were avoided through deworming of the young pigs that were bought to the stock.

2.2.2. Handling of manure

In Sweden there are no restrictions for spreading of manure from animals treated with veterinary medicines or for the keeping of treated animals. However, there are restrictions for the use of treated animals as food source.

2.3. Release pathways

Veterinary medicines used at farms can reach aquatic ecosystems through several pathways. A simple illustration of probable major routes is given in Figure 1. Following individual treatment, substances may be excreted with feces or urine in their parent form, as metabolites or as conjugates. It is possible that conjugates are deconjugated to the parent form in the environment (the process is known from municipal STPs).

Feces and urine is either applied directly to soil when the animals are grazing, or end up in manure that is generally is stored in large manure tanks at the farms. Manure is applied on fields as a fertilizer, and the medical substances in manure may adsorb to soil particles. Outflow to nearby surface waters may proceed by leaching or surface runoff.

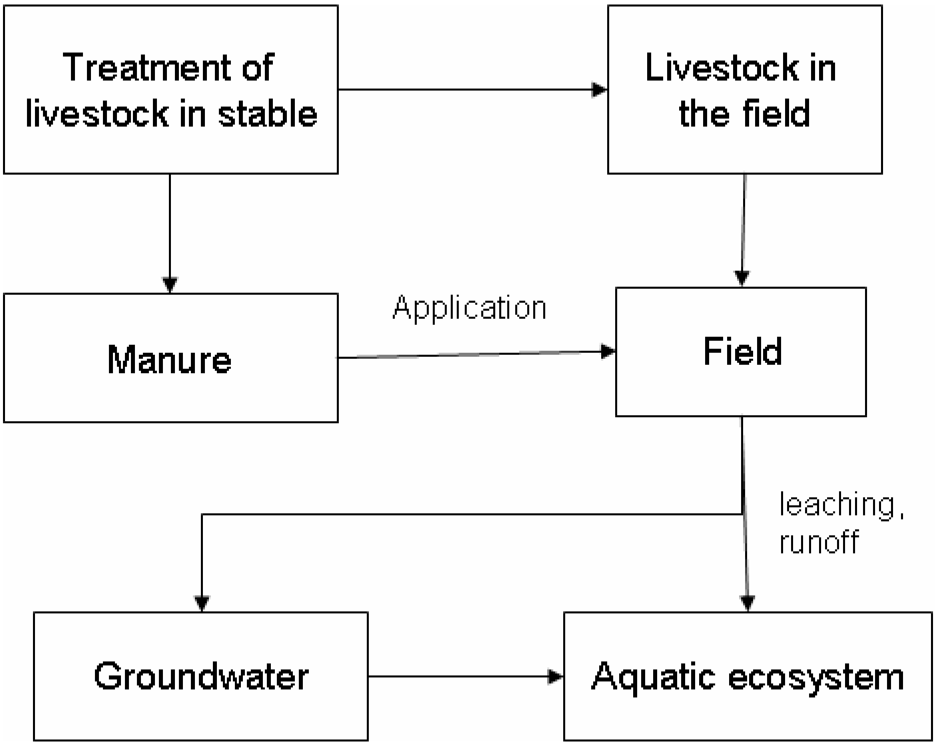

Figure 1. Conceptual release pathways for veterinary medicines used in livestock (modified after Halling-Sørensen m.fl., 2002).

3. Physico-chemical and ecotoxic properties

A recent compilation and review of physico-chemical properties of veterinary medicines is given in Hellström and Kreuger (2005). Due to the large number of compounds, these aspects are not presented in detail in this report but a brief review is presented below and in Table 2.

Table 2. Summary of physico-chemical properties (from Hellström and Kreuger, 2005). K_D values are from Sarmah et al. (2006).

Group	Excretion products	Solubility in water (mg/l)	log K_{ow}	K_D in soils (l/kg)
Antibiotics				
Tetracyclines	Unmetab. and metab.	600; 800; 630	- 0.02; - 0.49; -1.06	420-2400
Aminoglycosides	Unmetabolised	>20 000	low	
Sulfonamides	Unmetab. and metab.	2000; 2700; 273	-0.09; 0.7; 0.05	0.6-7.4
Trimethoprim	Unmetab. and metab.	400	0,91	
Macrolides	Unmetabolised	5	1.7 - 3,3	8-7700
Fluoroquinolones	Mainly unmetab., but also metab.	250; 70	-0.41; 3.1	420-5600
Parasiticides				
Sulfonamides	-	soluble	-	-
Benzimidazoles	Unmetab. and metab.	10-40; 3-5; insoluble	3.9; 1.95; 3.1; 1,53	-
Tetrahydropyrimidines	Unmetab. and metab.	insoluble	3,14	-
Macrocyclic lactones	Unmetab. and metab.	4; 0.03;0.51	3.22; 4.41;4.77	-

3.1. Antibiotics

Generally, the antibiotics are highly water soluble whereas the parasiticides are more lipophilic. Many of these substances are thought to adsorb to soil particles through electrostatic interactions rather than solely to organic carbon (Sarmah et al., 2006). Furthermore, many substances have one or several functional groups that may interact with protons, why the solubility and sorption behaviour vary with ambient pH in the soil.

Of the antibiotic substances, tetracyclines are relatively water soluble and easily photodegenerate in water. However, they bind to solid matrices and are stable in soil and sediment. Tetracyclines can be transformed into several other compounds and they are both excreted unmetabolised as well as metabolised. Sulfonamides are relatively stable compounds both in water and solid matrices, but they vary in their aquatic solubility. Sulfonamides can be metabolised and are excreted both as the parent substances as well as conjugated with acetic acid. Trimethoprim is a relatively stable compound. It is metabolized in the liver to oxides and hydrolysed metabolites, and is excreted both unchanged and metabolised. Trimethoprim is not easily photodegenerated in water.

Aminoglucosides are antibiotic substances with high water solubility. They are not metabolized and thereby they are excreted as the parent substances. Macrolides are also excreted

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unmetabolised, however they are highly lipophilic. Macrolides are relatively easily degraded in the environment. Fluoroquinolones, on the other hand, are stable compounds with low water solubility. However, they are easily photodegenerated in water. Fluoroquinolones are excreted mainly as the parent substance but also as active metabolites.

3.2. Antiparasitic compounds

Of the antiparasitic compounds, the information of the properties of the sulfonamides is limited, but they seem to be relatively water soluble. Benzimidazoles on the other hand have low water solubility and are stable compounds that are firmly bound to soil and sediment. They are excreted both as the parent substance and as metabolites. Tetrahydropyrimidines are insoluble in water and are mainly excreted as the parent substance or as inactive metabolites. Macrocyclic lactones (Avermectines) are lipofilic compounds that firmly binds to solid matrix. They are excreted both as the parent substance as well as metabolites. Some macrocyclic lactones are toxic to larva of insects that live in manure.

3.3. Ecotoxicity

Antibiotics have different modes of actions. Some are bacteriostatics (tylosin, oxitetracycline, sulfadoxin, trimethoprime) and inhibit protein synthesis therefore inhibiting growth and reproduction of the bacteria, whereas others are bactericides (dihydrostreptomycin and enrofloxacin) and kill the bacteria. Antiparasitic compounds also have different modes of actions and act against different kinds of parasites. Avermectines e.g. ivermectin mode of actions includes chloride influx into nerve cells hence disrupting the nerve impulse and decreases the nerve transmission (Kolar et al. 2006).

Some of the agents evaluated in this study have been classified as environmentally hazardous. According to a report from the Swedish Medical Products Agency (MPA, 2004) oxitetracycline, tylosin, dihydrostreptomycin and ivermectin are classified as very toxic to aquatic organisms. Ivermectin is also a potential bioaccumulator.

A compilation of NOEC and EC50 values is presented in Table 3. Except for ivermectin, all substances reviewed display ecotoxic properties at levels in the µg/l-mg/l range. Ivermectin is much more ecotoxic and the lowest NOEC found was 10 ng/l.

Table 3. Ecotoxic properties of selected veterinary medicines. The table contains data on those substances that were confirmed to be used at the investigated farms.

Agent	Organism	Test	Concentration	Reference
	ANTIPARASITIC			
Febantel	*Daphnia Magna*	EC50, 48 h	216 µg/l (48h)	Oh et al 2006
Ivermectin	*Daphnia magna*	NOEL, 48 h	0.01 µg/l	MPA 2004 (review)
	Salmo gairdneri (rainbow trout)	LC50, 96 h	3000 µg/l	Kolar and Erzen. 2006 (review)
	Lepomis macrochirus (freshwater fish)	LC50, 96 h	5.3 µg/l	MPA 2004 (review)
	Eisenia foetida (earthworm)	NOEL, 28 days	12 mg/kg soil	MPA 2004 (review)
	Haematobia irritans (dung dwelling organism)	LC50, 88 h	0.0032-0.0066 mg/kg	Kolar and Erzen 2006 (review)
	Scatophaga scercoraria (dung dwelling organism)	EC50	0.001-0.051 mg/kg	Kolar and Erzen 2006 (review)
	Bacteria	NOEC	2 mg/kg	MPA 2004 (review)
Fenbendazol	*Daphnia Magna*	EC50, 48 h	16.5 µg/l 1.25-4.1 µg/l (Chronic toxicity)	Oh et al 2006
	Salmo gairdneri (rainbow trout)	LC50, 96 h	40 µg/l	Kolar and Erzen 2006 (review)
	Onthophagus gazelle (dungdwelling organism)	NOEC, 7 days	770 ng/g	Kolar and Erzen 2006 (review)
	Earthworm	NOEC, 28 days	56 mg/kg	Kolar and Erzen. 2006 (review)
Oxfendazole	*Daphnia magna*	EC50, 48 h	1168 µg/l	Oh et al 2006
	Eubacteria (8 different genera of soil-dwelling bacteria)		no significant difference in highest tested concentration (9 ng/g)	Kolar and Erzen (review)
	Earthworm	28 days	no significant difference in highest tested concentration (971 ng/g)	Kolar et al. 2006 (review)
	Lepomis macrochirus (freshwater fish)	LC50, 96 h	>2700 µg/l (highest tested concentration)	Kolar and Erzen 2006 (review)
	ANTIBACTERIAL			
	Daphnia magna	EC50, 48 h	487 000 µg/l	Hellström-Kreuger 2005 (review)
	Microsystis aeruginosa	MIC	300 µg/l	Hellström-Kreuger 2005 (review)
Tylosin	*Microcystis aeruginosa* (freshwater cyanobacteria)	EC50, 7 days	34 µg/l	MPA 2004 (review)
	Lemna gibba (macro algae)	NOEC, 7 days	300 – 1 000 µg/l	Brain et al. 2004
	Daphnia magna	NOEC 21 days	45 000 µg/l	MPA 2004 (review)
	Selenastrum capricornutum (green algae)	EC50, 72 h	950 µg/l	Hellström-Kreuger 2005 (review)
	Aporrectodea caliginosa (earth worm)	EC50, 21 days	4530 mg/kg dw	MPA 2004 (review)

Agent	Organism	Test	Concentration	Reference
Oxitetracycline	Cyanobacteria	EC50	32-7000 µg/l	Ando et al. 2006
	Selenastrum capricornutum (green algae)	EC50	4500 µg/l	MPA 2004 (review)
	Panneus vannmei (crustacea)	LOEC, 24 h	161 µg/l	Hellström-Kreuger 2005 (review)
	Morone saxatilis (stripped bass)	LC50 96 h	75 000 µg/l	MPA 2004 (review)
Trimethoprim	cyanobacteria	EC50	11 000 - >200 000 µg/l	Ando et al. 2006
	Lemna gibba (macro algae)	LOEC, 7 days	>1 000 µg/l	Brain et al. 2004
	R. salina (algae)	EC50	160 µg/l	Hellström-Kreuger 2005 (review)
Dihydrostrep-tomycin*	*Lemna minor* (macro algae)	EC50	>1 000 µg/l	MPA 2004 (review)
Enrofloxacin	*Microsystis aeruginos* (cyanobacteria)	EC50, 24 h	49 µg/l	Robinson et al. 2005
	Psedokirchneriella subcapitata	EC50, 24 h	3100 µg/l	Robinson et al. 2005
	Lemna minor (macro algae)	EC50, 24 h	114 µg/l	Robinson et al. 2005
	Rainbow trout	LC50, 96 h	>10 000 µg/l	MPA, 2004 (review)
	Daphnia magna	LC50, 24 h	>10 000 µg/l	MPA, 2004 (review)
Sulfadoxin	No ecotoxicological data found			

* = Values presented are valid for streptomycin.

4. Sampling program

This study consists of a national program financed by the Swedish EPA, a larger regional program financed by the County Administration Board in Skåne, and two point samples from the County Administration Boards in Örebro and Gotland. In all of the 63 samples 50 substances were analysed. As in all screening studies, all aspects of chemical use cannot be investigated.

4.1. Sampling strategy

The strategy for choosing sampling sites was:

1. identify the major uses
2. identify large farms
3. select all farms with a topography beneficial for runoff or leaching to adjacent streams
4. identify farms that actually use some of these pharmaceuticals (through contact with the responsible at each farm).

The first step was to exclude the following release pathways:

- Fish farms, because the current medical use is low. The release via sewage water was not included due to the probable predominance of substances from human use.
- Horses and pets, since the livestock's of horses is generally small and pets are widely distributed throughout Sweden. Releases from horses and dogs will thus be difficult to track.

Focus was instead set on spreading of veterinary substances from treatment of pigs and cattle, since large amounts of antibiotics and parasiticides are used for these livestock. Further, because the manure from these animals are used for fertilization of arable land, farms with these common and widespread animals were considered to represent important sites with regard to environmental release of veterinary medicines. Large farms were considered suitable because of generally larger medical use per animal. Such farms were identified with the aid of the so-called "EMIR"-database that is part of the regional authorities enforcement of industrial activities.

The next step was to select large pig and cattle farms with topographical and hydrological conditions favorable for release to the aquatic environment, and where some of the substances to be analysed where used. This was achieved through large number of direct contacts with farmers, after initial identification of good release potential through map studies.

In addition, sampling was focused to areas where the manure from these animals is spread onto farmland. At each farm, the use of veterinary drugs was registered through personal contacts with the farmer. There was no official registry from where we could obtain such information. In as high extent as possible, sampling was performed in periods when these substances were used and following application of manure to soils. However, for project administrative reasons we were not able to sample during springtime when many antiparasitic compounds are used in the largest amounts. All samples were taken in 2006.

Collection of samples were performed both on a *local level*, i.e. sampling on farms of manure, soil, groundwater, surface water and/or sediment, and on a *regional level*, i.e. sampling of sediment and surface water in larger waters affected by several farms/sources (Table 4). In total 63 samples were collected from approximately twelve different water catchments areas (Table 4; Figure 2). One of these locations was a background area. The majority of samples were taken in four regions:

- Eastern Skåne
- Västra Götaland (close to Falköping)
- Östergötland (Mjölby-Boxholm)
- Södermanland (close to Nyköping)

These regions were selected mainly because they hold the major livestocks of pigs and cattle in Sweden and have a large part of arable land (SCB, 2006).

4.2. Areas sampled

In Skåne, samples were collected in June and August from four different water catchments areas, two areas in the vicinity of Kristianstad in northeastern Skåne and two areas in the southeastern part of the region. In each of the sampling areas water and sediment was collected in a dyke or minor stream, representing a local catchments area, as well as a in a somewhat larger water course representing a catchment on a regional level. Well water, manure and soil were sampled on 3 farms in the two northern areas (Table 5). The areas sampled in Skåne are intensively used for agricultural purposes, with a high density of cattle and pigs, but also sheep and horses. The landscape is relatively open and flat.

In Örebro, samples were collected by the county administration board in June from one water catchments area, located in the vicinity of the city Örebro in the eastern parts of the region. Water was collected in a larger water course, representing a regional catchments area. In Gotland, samples were also collected by the county administration board in June from one water catchments area, located in the central parts of the region. Water was collected in a larger water course, representing a regional catchments area.

In Östergötland, samples were collected in August from two water catchments area located in the western parts of the region. One of these areas is located in the vicinity of the city Mjölby and is characterized of a relatively hilly terrain. The agricultural area is mainly concentrated around a larger water course, where water and sediment were collected representing a regional catchments area. Animals handled within this part of the region are mainly cattle, but also pigs and fowls. Manure and soil were sampled on one cattle farm (Table 5). The other area, located in the vicinity of the city Vadstena, is relatively open and flat and intensively used for agricultural purposes. Water and sediment were collected from a lake and a stream, representing a regional catchments area. Within this area large pig farms and fowls farm dominate.

Table 4. Sampling areas and distribution of samples. Level B: background; L: local; R: regional.

Description	Surface water	Ground water	Sediment	Manure	Soil	Level	Month
<u>**BACKGROUND AREAS**</u>							
Lake 1 (Rotehogstjärn)	1		1			B	November
<u>**AGRICULTURAL AREAS**</u>							
Area 1 Skåne							
Farm 1		1		1	1	L	June
Vinne å	2		2			R	June, August
Bockebäck, minor feeder stream to Vinne å	2		2			L	June, August
Area 2 Skåne							
Farm 2		1		1	1	L	June
Farm 3				1	1	L	June
Rambrobäcken, minor feeder stream to Vramsån	2		2			L	June, August
Vramsån, feeder stream to Helge å	1					R	June
Helge å	2		2			R	June, August
Area 3 Skåne							
Tommarpsån	1		1			R	June
Tommarpsån	2		1			L/R	June, August
Area 4 Skåne							
Nybroån	1		1			R	June
Örupsån, feeder stream to Nybroån	2		2			L/R	June, August
Area 5 Örebro							
Täljeån	1					R	June
Area 6 Gotland							
Gothemsån	1					R	June
Area 7 Östergötland							
Farm 4				1	1	L	August
Svartån	3		3			R	August
Svartån	1					R	August
Feeder stream to Svartån	1					R	August
Area 8 Östergötland							
Tåkern	2		1			R	August
Lorbybäcken feeder stream to Tåkern	1					R	August
Area 9 Västergötland							
Farm 5	1			1	1	L	August
Ätran	1					R	August
Area 10 Södermanland							
Farm 6	1			1	1	L	September
Area 11 Södermanland							
Runnviken	1		1			R	September

In Västergötland, samples were collected in August from one water catchments area in the eastern parts of the region, in the vicinity of the city Falköping. This area is characterised of cattle farms and it is relatively intensively used for agricultural purposes. The landscape is relatively open and flat. Water was collected in a somewhat larger water course representing a regional catchments area samples. Manure, soil and water in a minor stream were sampled on a farm with dairy cows, representing local samples (Table 5).

In Södermanland, samples were collected in September from two different water catchments area, one on a farm with fattening pigs, representing local samples, and the other in a lake, representing regional samples. These areas are located in a hilly terrain and farms are few. There is mainly poultry and pigs handled within these areas.

Figure 2. Overview of the investigated regions.

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4.3. Farms sampled

In total samples from 3 dairy cow farms and 3 fattening pig farms where sampled (Table 5). The veterinary drugs and the active substances of these used by the farms where manure and soil were sampled are shown in Table 6. Of the 50 substances analysed, at least 10 were confirmed to be used at these farms.

Table 5. Farms that were sampled.

Study area	Municipal-ity	Animals	Manure last applied before sampling	Regional recipient
Farm 1	Kristianstad	Fattening pigs, ca 2800 individuals	April, 2 months	Vinne å
Farm 2	Kristianstad	Dairy cows, ca 164 individuals	During the whole summer season (pasture land)	Vramsån, Helge å
Farm 3	Kristianstad	Fattening pigs, ca 270 individuals	April, 2 months	Vramsån, Helge å
Farm 4	Mjölby	Dairy cows, ca 150 individuals	June, 2 months	Svartån
Farm 5	Falköping	Dairy cows, ca 180 individuals	June, 2 months	Ätran
Farm 6	Katrineholm	Fattening pigs, ca 7000 individuals	September, 1 week	-

Table 6. The drugs used at the six farms sampled and the active substance of the drugs (the minimum number of used drugs). Substances not analysed in this study are shown in italics.

Drugs used	Active substance		Number of farms
	Antiparasitic	Antibacterial	
Baytril		Enrofloxacin	4
Ethacilin		*Bensylpenicillinprokain*, Dihydrostreptomycin	4
Bimotrim		Sulfadoxin, Trimethoprim	2
Engemycin		Oxitetracycline	2
Tylan		Tylosin	2
Ultra Pen		*penicillinprokain*	2
Penovet		*Bensylpenicillinprokain*	2
Siccalactin		Dihydrostreptomycin, *Bensylpenicillin*	1
Geepenil		*Bensylpenicillin*	1
Systamex	Oxfendazol		2
Axilur	Fenbendazol		1
Ivomec	Ivermectin, *Prazikvantel*		1
Rintal	Febantel		1

5. Methods

5.1. Sampling

Manure was collected from open manure tanks at the farms. Sampling of soil was performed from different farmlands where manure had been applied one week, or at the most two months, before sampling. In one case, soil was also collected from a pasture land. The samples where collected as pooled samples from the topsoil (5-15 cm).

Groundwater was sampled at two farms from shallow wells (< 50 m depth). None of the other farms sampled had shallow wells, why groundwater was not collected at those farms.

Most surface waters were sampled with a Ruttner-collector and most surface sediments were collected with a gravity corer and sliced for 0-2 cm in the field. In Skåne, however, surface waters were collected directly in the bottle. Each sample contained four liters of surface waters or 500 ml of sediments /manure/ soil. All samples were kept cold and dark in glass bottles until analyses where performed. The samples were sent to the laboratory within two days after sampling.

5.2. Chemical analysis and quality assurance

All samples were analysed by Analytica AB (GALAB Laboratories, Germany). The analytical procedure was initalised within ca 4-5 days after sampling. All compounds were detected with LC-MS-MS. Details of the analytical procedure is given in appendix 1 and reporting limits are shown in appendix 2. Detection limits were 3-5 times lower than the reporting limits. Quality assurance was performed according to the following procedures:

- Every sample set contain a blank samples throughout the whole process.
- An internal standard is added prior to extraction of the samples, in order to check the extraction efficiency
- Certified reference materials (CRM) were used when commercially available. In other cases, synthetic standards were used.
- One sample in every sample set was run in duplicate, to assure reproducibility within the laboratory.
- The laboratory successfully participate in round robin tests regarding pharmaceuticals in various matrices.
- The entire analytical procedure follows EN ISO/IEC 17025.

The method does not include conjugated species. The total uncertainty in the results is also influenced by natural heterogeneity. As an indicative measure of the total uncertainty in the results, sediments and surface waters were sampled in triplicate from one site in Östergötland.

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6. Results and Discussion

None of the analysed veterinary substances were detected in any of the collected samples. This result does not exclude that these substances are present; however, it shows that the compounds does not exist in levels above the prevailing detection limit. The analysed substances and the detection limits for the different matrices are shown in appendix 2.

6.1. Relation to other studies of veterinary medicines

There are several investigations from other countries that can be compared to this study. When different studies are compared, it must be emphasized that different study areas differ widely with respect to the amounts of medicines used, the distribution processes and hydrological conditions.

For instance, surface waters from an agricultural watershed in Canada were sampled biweekly for six months. Of the 28 different pharmaceuticals analysed, most were never or only rarely detected. Median levels of the others were generally less than 10 ng/l (Lissemore et al., 2006). Substances with human use were detected more frequently than those with only veterinary use.

The presence of tetracyclines, sulfonamides and macrolides in surface waters and sediments was investigated in regions with intensive agricultural activities (including large animal farms) in Colorado, USA (Kim and Carlson, 2007). The following levels were detected in surface waters:

- Tetracyclines: <10 - 1800 ng/l; most samples were below 50 ng/l
- Sulfonamides: <10 – 80 ng/l; most samples were below 20 ng/l
- Macrolides: < 10 – 240 ng/l; most samples below 50 ng/l.

In sediments, the highest levels were ca 70 µg/kg but most substances occurred at levels less than 10 µg/kg.

These two studies both show levels that occassionally are higher than the analytical reporting limits in this study, but they also show strongly time-dependent levels. At least in the USA, antibiotics are routinely used as growth promoters (Sarmah et al., 2006). This use is not permitted in Sweden, why it can be anticipated that the Swedish veterinary use of antibiotics per animal is much lower. Accordingly, lower levels in the environment might be expected in Sweden than in USA.

6.2. Metabolic transformation

Certain pharmaceuticals may partly be excreted as metabolites or as conjugated species (Table 2). Such species are not included in the analytical methods used. Therefore, we can neither confirm nor deny the possibility that the veterinary use of these substances influences the aquatic environment through these altered chemical species.

6.3. Reporting limits compared to toxic effect levels

In our study, no substance occurred above the analytical reporting limits, that ranged from 10 – 100 ng/l for different substances in surface water, and from 10-100 µg/kg dw in soil and manure (Appendix 2). Neither were any traces detected (no results above the detection limits). If this dataset is to be used to assess possible ecotoxicological impact, these reporting limits can be compared to effect and no-effect levels. The EC50 and NOEC for aquatic organisms are shown in Table 3 for those medicines that we could verify were used in the study areas. Because exposure can be expected to be intermittent, effects due to chronic exposure may though be less relevant in most cases.

With the exception of ivermectin, all EC50 and NOEC values were orders of magnitude higher than the reporting levels. The toxicity data covers both acute and chronic toxicity. For dung-dwelling organisms, however, EC50 values as low as 1 µg/kg dw have been reported for ivermectin. Because our reporting limit was 100 µg/kg for ivermectin, the possible risks of this compound cannot be assessed.

With the exception of ivermectin, the data in this study suggest that the use of antibiotics and parasiticides in livestocks of dairy cows and pigs do not pose a general risk to the Swedish aquatic ecosystems or to agricultural soils.

Similar conclusions were drawn in the Medical Products Agency report (MPA, 2004), where they concluded that measured levels of pharmaceuticals in the environment were at least thousand times lower than the acute toxic levels identified for aquatic organisms.

6.4. The potential size of the problem: veterinary drugs compared to pesticides

In order to assess the potential size of the problem, the use of veterinary medicines can be compared to that of pesticides. Both groups of chemicals are used in agricultural areas, although the exact location and timing of their use differ.

6.4.1. Total amounts used

Figures on the amounts of veterinary products sold in Sweden have previously been rare or even lacking. However, information recently compiled by the Swedish Board of Agriculture (Table 1) constitutes the first comprehensive information of this group of medical products (SBA, 2007). Overall, sales were greatly dominated by antibacterial substances amounting to a total of ca 15 metric tons of active ingredients sold during 2005. Corresponding figure for antiparasitic substances was 4 tons.

Comparing these figures with sold amounts of plant protection products (pesticides) demonstrates that there are large differences. During 2005 a total of ca 1 700 metric tons of pesticides were sold for agricultural and horticultural purposes (KemI, 2006). Active ingredients are used for many different purposes and under a variety of conditions throughout the cropping season. Sales are dominated by herbicides (1 300 tons), followed by fungicides (240 tons), seed dressings (42 tons), insecticides (22 tons) and plant growth regulators (20 tons).

6.4.2. Use on an areal basis

As a rough indicator of the source strength, it can be anticipated that the levels in adjacent surface water are proportional to the levels in soils in the water catchments area. The levels in soils will be related to the levels in manure and the rate of manure application to soil. Therefore, we will try to express the use of veterinary medicines in grams of active substance per hectare. This measure can be directly compared to that of pesticides, for which abundant monitoring data exists for Swedish rivers (Adielsson et al, 2006). The estimation will rely on conservative assumptions. We assume that all medicines given to animals end up in the parent form (unmetabolised) in the manure.

The annual amount of substance given per animal treated, M_F [milligram/yr], is calculated as:

$$M_F = Dose \cdot bw \cdot t \cdot n,$$

where *Dose* is the individual daily dose [mg substance per kg bw and day], *bw* is the body weight [kg], *t* is the number of days for which treatment proceeds, and *n* is the number of treatments per year. The concentration in manure at a farm, C_M [mg/kg wet manure] is calculated as:

$$C_M = M_F / G * x/y$$

where *G* is the annual production of manure per animal, *x* is the number of animals treated and y is the total number of animals at the farm. The area specific release of a veterinary substance *i* , R_i [g/hectare and year], is calculated as:

$$R_i = C_M \cdot A/1000$$

where *A* is the application rate of manure [kg wet manure per hectare]. General data on G and A were taken from SBA (2006). Dose is taken from FASS for veterinary purposes. Based on discussion with veterinaries, we assume that t=1 and n=5 for parasiticides. For antibiotics, t≅4 according to veterinaries but it is not possible to generally state the number of treatments per year and animal. We have assumed that n=1. For parasiticides, we assumed that x/y was equal to 0.33, i.e. every third animal is ca one year old. For antibiotics, we assumed that x/y equaled 0.25, i.e. every fourth animal is treated with antibiotics.

The results are shown in Table 7 and must be regarded as upper estimates and with a substantial uncertainty. Major contributions to uncertainty are that metabolism in the animals and degradation in the manure is disregarded. The release rates ranged from 0.2 to 30 g active substance per hectare and year and they are higher for antibiotics than for parasiticides, mainly due to higher individual doses of antibiotics.

We can compare these figures to those of pesticides. Applied dosage of pesticides is quite variable, ranging from a few grams per hectare (e.g. pyrethroids and sulfonyl urea herbicides) up to several kilograms per hectare (e.g. glyphosate, certain potato fungicides and sugar beet herbicides). However, there is a certain correlation between dosage and how toxic the compounds are. For example pyrethroids are toxic at very low concentrations to aquatic species, with Swedish guideline values close to or even below 1 ng/l.

In summary, the calculated upper release rates of antibiotics is in the same range as some of the more toxic pesticides, whereas many other pesticides are applied in larger doses. Sulfonyl ureas and pyrethroids are two groups of pesticides that are applied at a few grams per hectare. These substances are occassionally found in Swedish rivers, in levels of 10-50 ng/l (sulfonyl ureas) and 2-10 ng/l (pyrethroids) (Adielsson et al, 2006). The detection frequencies are fairly low. When comparing the use of antibiotics to that of pestidices, it is not surprising that levels of antibiotics in streams were lower than 10 ng/l.

Table 7. Upper estimates of area specific release of a veterinary substances to agricultural fields through application of manure.

Substance	Animal	R (g substance (ha * år)$^{-1}$
ANTIBIOTICS		
Enrofloxacin	dairy cow	1.5
Enrofloxacin	fattening pigs	1.7
Dihydrostreptomycin	dairy cow	15.0
Dihydrostreptomycin	fattening pigs	17.3
Sulfadoxin	dairy cow	9.0
Sulfadoxin	fattening pigs	10.4
Trimetoprim	dairy cow	2.4
Trimetoprim	fattening pigs	2.8
Oxytetracycline	”young cattle”	30
Tylosin	fattening pigs	24
PARASITICIDES		
Fenbendazol	”young cattle”	7.4
Ivermectin	”young cattle”	0.2

6.5. Limitations of the study

With regard to the overall question – whether the Swedish use of veterinary medicines causes an environmental impact – several aspects remains to be investigated. This is a first screening study on the subject and all aspects cannot be studied.

The study is restricted to 65 samples, and most study sites were only sampled once. International studies have frequently demonstrated that the levels of veterinary pharmaceuticals is highly varying over time (e.g. chapter 6.1). Therefore, it is likely that some of these substances occur at levels above our reporting limits at certain times and sites. Our reporting limits were in the range 10-100 ng/l. Although these reporting limits appears sufficient with regard to ecotoxicity of most substances (chapter 6.3), they cannot be used to infer that releases do not occur.

Veterinary substances have been detected in groundwaters (e.g., Batt et al., 2006). Very few groundwaters were analysed in the present study, why the question of long-term influence of manure application on the presence of pharmaceuticals in groundwater remains open.

Certain release pathways of veterinary substances was not included due to project administrative reasons, such as the impact from aquaculture (fish farms) and treatment of horses. In north-american fish farms, tetracyclines and sulfadimethoxine were occasionally detected in surface waters at levels below ca 1 µg/l (Thurman et al., 2002). The Swedish use of antibiotics in fish farms has decreased substantially over the last years, and it does certainly not represent the major veterinary use of these substances.

7. Further studies

If veterinary compounds should be studied further in the Swedish environment we suggest that sampling is focused onto one or two farms with large livestock. Sampling should be performed more frequently and also in the spring when the use of antiparasitic compounds is large. This should improve the chance of detecting the release of the compounds into the environment, by establishing close contact and information from the farmer when the substances are used. Furthermore, metabolites should be included if analytical methods are developed.

8. Conclusions

No single sample contained antibiotics or parasiticides in detectable levels. With the exception of ivermectin, all EC50 and NOEC values were orders of magnitude higher than the analytical reporting levels. Therefore, the data in this study suggest that the use of antibiotics and parasiticides in livestocks of dairy cows and pigs does not pose a general risk to the aquatic ecosystems or to agricultural soils.

The use and environmental occurrence of veterinary compounds were also compared to that of pesticides, a group of chemicals that are applied in similar regions and for which ample monitoring data exists. During 2005, the total Swedish sale of pesticides was ca 100 times higher than that of veterinary antibiotics and parasiticides. Annual area doses to agricultural soils were estimated, by conservatively assuming that the applied doses all end up in soil through the use of manure as fertilizer. The area doses ranged from 0.2 to 30 g active substance per hectare and year for individual substances. This corresponds to the lower range of application rates for individual pesticides.

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WSP Environmental

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Appendix 1. Analytical methods

Tetracyclines	Sulfonamides	Aminoglycosides	Benzimidazoles
Tetracycline	sulfabenzamide	Dihydrostreptomycin	Fenbendazole
Oxytetracycline	sulfacetamide	Gentamycin	Albendazole
Doxycycline	sulfachloropyridazine		Flubendazole
	sulfadiazine		Oxfendazole
	sulfadimethoxine		Febantel
	sulfadimidine		
	sulfadoxine		
	sulfafurazol		
	sulfaguanidine		
	sulfaclozin		
	sulfamerazine		
	sulfameter		
	sulfametoxazol		
	sulfamethoxypyridazine		
	sulfamoxole		
	sulfanilamide		
	sulfapyridine		
	sulfaquinoxaline		
	sulfatiazol		
	sulfamethizole		
	sulfamonomethoxine		
Methods for aquatic samples			
Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration
Adjustment of pH value	Adjustment of pH value	Adjustment of pH value	Adjustment of pH value
Adding internal standards	Adding internal standards	Adding internal standards	Adding internal standards
SPE	SPE	SPE	SPE
LC-MS-MS	Cleanup	Cleanup	Cleanup
	LC-MS-MS	LC-MS-MS	LC-MS-MS
Methods for solid samples			
Adding internal standards	Adding internal standards	Adding internal standards	Adding internal standards
Adjustment of pH value	Adjustment of pH value	Solid-liquid-extraction	Adjustment of pH value
Solid-liquid-extraction	Solid-liquid-extraction	Adjustment of pH value	Solid-liquid-extraction
Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration
SPE	SPE	SPE	SPE
LC-MS-MS	LC-MS-MS	LC-MS-MS	LC-MS-MS

Analytical methods, continued.

Fluoroquinolones	Macrocyclic lactones	Macrolides	Tetrahydro-pyrimidines	Trimethoprim
Ciprofloxacin	Ivermectin	Tylosin	Morantel	Trimethoprim
Danofloxacin	Doramectin	Spiramycin	Pyrantel	
Difloxacin	Eprinomectin			
Enrofloxacin	Moxidectin			
Flumequin				
Marbofloxacin				
Norfloxacin				
Sarafloxacin				
Oxolinsäure				
Ofloxacin				
Methods for aquatic samples				
Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration
Adjustment of pH value	Adjustment of pH value	Adjustment of pH value	Adjustment of pH value	Adjustment of pH value
Adding internal standards	Adding internal standards	Adding internal standards	Adding internal stan-dards	Adding internal standards
SPE	SPE	SPE	SPE	SPE
Cleanup	Cleanup	Cleanup	Cleanup	Cleanup
LC-MS-MS	LC-MS-MS	LC-MS-MS	LC-MS-MS	LC-MS-MS
Methods for solid samples				
Adding internal standards	Solid-liquid-extraction	Adding internal standards	Adding internal stan-dards	Adding internal standards
Adjustment of pH value	Adjustment of pH value	Adjustment of pH value	Adjustment of pH value	Adjustment of pH value
Solid-liquid-extraction	Centrifugation + filtration	Solid-liquid-extraction	Solid-liquid-extraction	Solid-liquid-extraction
Centrifugation + filtration	SPE	Centrifugation + filtration	Centrifugation + filtration	Centrifugation + filtration
SPE	LC-MS-MS	SPE	SPE	SPE
LC-MS-MS		LC-MS-MS	LC-MS-MS	LC-MS-MS

Appendix 2. Analytical reporting limits. Detection limits were 3-5 times lower.

ELEMENT	Surface water (ng/l)*	Sediment (µg/kg dw)*	Manure (µg/kg dw)*	Soil (µg/kg dw)*
ciprofloxacin	<10	<10	<10	<10
danofloxacin	<10	<10	<10	<10
difloxacin	<10	<10	<10	<10
enrofloxacin	<10	<10	<10	<10
flumequine	<10	<10	<10	<10
marbofloxacin	<10	<10	<10	<10
norfloxacin	<10	<10	<10	<10
ofloxacin	<10	<10	<10	<10
sarafloxacin	<10	<10	<10	<10
oxolinic acid	<10	<10	<10	<10
sulfabenzamide	<10	<10	<10	<10
sulfacetamide	<10	<10	<10	<10
sulfachloropyridazine	<10	<10	<10	<10
sulfadiazine	<10	<10	<10	<10
sulfadimethoxine	<10	<10	<10	<10
sulfadimidine	<10	<10	<10	<10
sulfadoxine	<10	<10	<10	<10
sulfafurazol	<10	<10	<10	<10
sulfaguanidine	<10	<10	<10	<10
sulfaclozin	<10	<10	<10	<10
sulfamerazine	<10	<10	<10	<10
sulfameter	<10	<10	<10	<10
sulfametoxazol	<10	<10	<10	<10
sulfamethoxypyridazine	<10	<10	<10	<10
sulfamoxole	<10	<10	<10	<10
sulfanilamide	<10	<10	<10	<10
sulfapyridine	<10	<10	<10	<10
sulfaquinoxaline	<10	<10	<10	<10
sulfatiazol	<10	<10	<10	<10
sulfamethizole	<10	<10	<10	<10
sulfamonomethoxine	<10 - 50	<10	<10	<10
oxytetracycline	<10 - 50	<10 - 50	<10 - 50	<10 - 50
chlorotetracycline	<10 - 50	<10 - 50	<10 - 50	<10 - 50
doxycycline	<10 - 50	<10 - 50	<10 - 50	<10 - 50
tylosin	<10 - 30	<10	<10	<10
spiramycin	<10 - 100	<10 - 30	<10 - 30	<10 - 30
ivermectin	<100	<100	<100	<100
doramectin	<100	<100	<100	<100
eprinomectin	<100	<100	<100	<100
moxidectin	<100	<100	<100	<100
dihydrostreptomycin	<100	<100	<100	<100
gentamycin	<100	<100	<100	<100
fenbendazole	<10	<10	<10	<10
oxfendazole	<10	<10	<10	<10
albendazole	<10	<10	<10	<10

febantel	<10	<10	<10	<10
flubendazole	<10	<10	<10	<10
pyrantel	<10 - 50	<10 - 50	<10 - 50	<10 - 50
morantel	<10 - 50	<10 - 50	<10 - 50	<10 - 50
trimethoprim	<10 - 20	<10 - 20	<10 - 20	<10 - 20
PCB 28	<0,001 - 0,01	<0,003 - 0,02	<0,01 - 0,03	<0,003
PCB 52	<0,001 - 0,01	<0,003 - 0,02	<0,01 - 0,03	<0,003
PCB 101	<0,001 - 0,01	<0,003 - 0,02	<0,01 - 0,03	<0,003
PCB 118	<0,001 - 0,01	<0,003 - 0,02	<0,01 - 0,03	<0,003
PCB 138	<0,001 - 0,01	<0,003 - 0,02	<0,01 - 0,03	<0,003
PCB 153	<0,001 - 0,01	<0,003 - 0,02	<0,01 - 0,03	<0,003
PCB 180	<0,001 - 0,01	<0,003 - 0,02	<0,01 - 0,03	<0,003
sum PCB	<0,004 - 0,035	<0,01 - 0,07	<0,035 - 0,1	<0,01

*PCB anges i µg/l alternativt mg/kg TS