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Swedish Veterinary Antimicrobial Resistance Monitoring – surveillance of resistance in bacteria from animals

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Abstract

To gather knowledge on resistance among bacteria from animals on a national level, the Swedish Antimicrobial Resistance Monitoring Programme (SVARM) was started in 2000. In the programme, antimicrobial susceptibility of zoonotic bacteria, specific animal pathogens and commensal enteric bacteria (*Escherichia coli* and *Enterococcus* spp.) is monitored on a regular basis. Results are presented yearly together with data on consumption of antimicrobial agents in veterinary medicine.

Introduction

Acquired resistance among bacteria is a consequence of the use of antimicrobials and follows the Darwinian laws on survival of the fittest. In this context, bacterial populations in man and in animals must both be considered, since resistant strains from animals can be transferred to man (e.g. *Salmonella* spp. *Campylobacter* spp.). Moreover, mobile genetic elements conferring resistance can be transferred from bacteria in animals to bacteria in man and vice versa (Summers, 2002).

In a longer perspective, resistance cannot be avoided but prudent use can increase the 'life-span' of a substance as stressed by influential organisations such as OIE and WHO. An essential basis for prudent use is knowledge of resistance in populations of bacteria coupled with data on use of antimicrobials. Such knowledge forms the basis for therapeutic decisions with the best chance of clinical success and minimal effect on antimicrobial resistance and for interventions on use of antimicrobials.

To this end, monitoring of antimicrobial susceptibility of bacteria, including commensal bacteria, and compiling consumption statistics has been advocated (WHO, 2001). The rationale of monitoring commensal bacteria is that these form a reservoir of mobile resistance genes that can be transferred to bacteria of clinical importance. Moreover, prevalence of acquired resistance among these 'indicator bacteria' is thought to reflect the magnitude of the selection pressure enforced by the use of antimicrobials in a population (van den Bogaard and Stobberingh, 2000). Guidelines for national monitoring programmes have been laid down by e.g. OIE (Franklin and others, 2001).

In Sweden, yearly statistics on antimicrobial use in animals have been compiled since the 80s and several studies on antimicrobial resistance in bacteria from animals have been performed since the early 70s. In the Swedish Antimicrobial Resistance Monitoring Programme (SVARM), started year 2000, efforts in these fields are combined. The programme is organised and run by the Dept. of Antibiotics at the National Veterinary Institute (SVA). This paper outlines the scope of the programme, the methodology used and presents examples of results obtained.

Objective

The remit of SVARM is to regularly monitor antimicrobial resistance in bacteria from animals and present and analyse the results in relation to data on use of antimicrobials. The objectives are to detect trends in resistance that might be cause for interventions and to provide a basis for

recommendations on appropriate choice of therapy in the clinical setting. In a wider perspective, data could be used for risk analysis in the field of antimicrobial resistance.

Methods

In SVARM, three types of bacteria are included: zoonotic bacteria, indicator bacteria (*Escherichia coli* and *Enterococcus* spp.) and specific animal pathogens.

Zoonotic bacteria

Salmonella from warm-blooded animals (wild and domesticated) are included in the monitoring. In Sweden, salmonellosis in animals is a notifiable disease and at least one isolate from each incident is confirmed at SVA. Isolates for susceptibility testing are therefore easily accessible and yearly one isolate from each notified incident is tested and included in the report.

Campylobacter spp. from slaughter pigs, broiler chickens and cattle are obtained from the Swedish *Campylobacter* programme (broilers) or isolated from samples of intestinal content collected from healthy animals at abattoirs. Yearly about 100 isolates from one of the animal species above are included.

Indicator bacteria

The indicator bacteria, *E. coli* and *Enterococcus* spp., are isolated from intestinal content of healthy animals sampled at slaughter. The number of samples collected at each abattoir is proportional to the respective annual slaughter volume and each isolate represents a unique herd or flock. Thereby, the included bacterial isolates represent randomly selected healthy individuals. Each year about 300 isolates of *E. coli* and *Enterococcus* spp. from either slaughter pigs, cattle or broiler chickens are included.

Pathogenic bacteria

Isolates of animal pathogens in SVARM mostly emanate from routine bacteriological examinations of clinical submissions or post-mortem examinations at SVA but also from specific field studies. Examples of bacteria included are: *E. coli* from pigs, cattle, horses, cats and dogs; *Brachyspira* spp. and *Actinobacillus* spp. from pigs; *Pasteurella/Mannheimia* and udder pathogens from cattle; *Streptococcus zooepidemicus* and *Rhodococcus equi* from horses; *Staphylococcus intermedius* from dogs. Extracting historic data from the database at SVA made it possible to present materials from the period back to 1992 already when SVARM was initiated in 2000.

Resistance monitoring based on diagnostic submissions is difficult since data are likely to be biased towards herds with disease problems and towards recurrent cases. Moreover, for some bacteria only a few isolates are available each year and anamnestic information is often scarce. Results should be interpreted bearing these inadequacies in mind.

Use of antibacterials

Statistics on use of antimicrobials is based on electronic records of amount of drugs dispensed at or from pharmacies, i.e. sales statistics supplied by Apoteket AB (The National Corporation of Swedish Pharmacies). Antimicrobial drugs used in veterinary medicine in Sweden are only available on veterinary prescription and have to be dispensed through pharmacies. Data include all use of veterinary antimicrobials, i.e. from food-producing animals as well as for pets and horses. Unfortunately, statistics are not yet available per animal species but will be in the near future.

Laboratory methodology

All isolates included in SVARM are isolated or confirmed at SVA. Susceptibility tests are performed by microdilution, in essence following the standards of the Clinical and Laboratory Standards Institute (CLSI, 2006). The microdilution panels used, VetMIC™, are produced by the SVA. Different panels are used depending on bacterial species and the original purpose of the

investigation (monitoring or clinical diagnostics). Participating departments are accredited to perform susceptibility tests and isolation and identification of animal pathogens and *Salmonella* according to SS-EN ISO/IEC 17025 by the Swedish Board for Accreditation and Conformity Assessment (SWEDAC).

Isolates are classified as sensitive or resistant according to microbiological cut-off values. For classification of zoonotic bacteria (*Salmonella* and *Campylobacter*) and indicator bacteria (*E. coli* and enterococci) cut-off values recommended by the European Committee on Antimicrobial Susceptibility Testing (EUCAST) are used (http://www.escmid.org). For animal pathogens, breakpoints recommended by CLSI (formerly NCCLS, 2002) are also taken into consideration.

Report

Results from the monitoring programme are presented in a yearly report available at www.sva.se. Antibacterial susceptibility is presented as quantitative data, i.e. percentage of resistant isolates, but also a qualitative data in the form of distributions of MICs.

Results and Discussion

As examples of data in the SVARM report, statistics on use of antimicrobials, resistance in *S.* Typhimurium from food producing animals and resistance in *E. coli* from healthy pigs and from diagnostic submissions from pigs are presented.

Antimicrobials for use in animals in Sweden are only available on veterinary prescription and guidelines emphasising judicious use have been issued. Use for growth promotion was banned in year 1986. The overall sale of antimicrobials for veterinary use in Sweden has declined in the last 25 years (Table I). Most of the antimicrobials sold for use in animals year 2005 are products formulated for treatment of individual animals (86%). The use of most groups in this subset has decreased or been relatively unchanged over the last five years. However, the use of fluoroquinolones for treatment of individual animals has increased. Notably, the entire increase derives from increases sales of tablets for treatment of companion animals since 1999.

Table I. Yearly sales of antimicrobial drugs for veterinary use expressed as kg active substance (sales statistics from Apoteket AB).

Antimicrobial class	**1980**	**1984**	**1988**	**1992**	**1996**	**2000**	**2002**	**2003**	**2004**	**2005**
Tetracyclines[a]	9 819	12 955	4 691	8 023	2 698	1 754	1 415	1 307	1 329	1 562
Amfenicols	47	49	35	-	-	-	-	-	-	-
Penicillin G-and V[b]	3 222	4 786	7 143	7 446	8 818	8 254	8 179	7 579	7 814	7 571
Aminopenicillins	60	714	655	837	835	852	767	870	875	911
Other betalactams incl. cephalosporins	9	2	-	-	-	315	676	832	928	1 009
Aminoglycosides and polymixins[a]	5 274	5 608	3 194	2 139	1 164	797	753	645	606	762
Sulphonamides	6 600	4 325	3 072	2 362	2 198	2 338	2 477	2 326	2 462	2 535
Trimethoprim & derivatives	134	186	250	284	339	390	414	381	406	437
Macrolides & lincosamides	603	887	1 205	1 710	1 649	1 352	1 412	1 124	1 095	1 080
Fluoroquinolones	-	-	-	147	173	156	185	184	187	184
Pleuromutilins	-	-	124	268	1 142	871	988	744	387	338
Quinoxalines[c]	6 250	9 900	7 164	4 917	1 098	-	-	-	-	-
Streptogramins	-	8 800	1 088	1 275	525	-	-	-	-	-
Other substances[d]	861	1 637	1 567	1 634	-	-	-	-	-	-
Feed additives[e]	8 380	700	-	-	-	-	-	-	-	-
	41 259	**50 549**	**30 189**	**31 043**	**20 639**	**17 079**	**17 266**	**15 992**	**16 089**	**16 389**

[a] Includes drugs marketed with special marketing authorisation for years 2000-2005; [b] Calculated as benzyl-penicillin; [c] years 1980-1984 sold as feed additives, thereafter on veterinary prescription at therapeutic dosages; [d] Mainly nitroimidazoles; [e] Feed additives other than quinoxalines and streptogramins: avoparcin, bacitracin, nitrovin, oleandomycin and spiramycin.

Infections with Salmonella in food animals are rare in Sweden and resistance among *Salmonella enterica*, as well as in the subset *S.* Typhimurium, is uncommon (Table II). Occurrence of multiresistance greatly influences the prevalence of resistance but such isolates are rare among Swedish animals. Since 1997, when testing of one isolate of each serovar from each notified incident commenced, only 26 of 687 isolates tested have been multiresistant. All multiresistant isolates have been *S.* Typhimurium; 13 were from a total of nine incidents in food animals, 12 isolates from an equal number of incidents in companion animals and one isolate from a wild boar. Thus, multiresistant *Salmonella* was involved in only nine of about 330 notified incidents in food producing animals since 1997. Therefore, the overall situation of antimicrobial resistance in *Salmonella* among food-producing animals is favorable and spread of multiresistant clones is contained, most likely a result of the strategies in the Swedish *Salmonella* control programme. Moreover, there is no indication of spread of such clones among wild animals as only one of 102 *Salmonella enterica* isolates from wild animals tested since 1997 was multiresistant.

Table II. Distribution of MICs for *Salmonella* Typhimurium (n=86) from food animals years 2000-2005.

Substance	**Resistance**	**Distribution** (%) **of MICs**[a] (mg/L)															
	(%)	**≤0.03**	**0.06**	**0.12**	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**>512**
Ampicillin	9						58.1	31.4	1.2			9.3					
Cefotaxime	0[b]			75.0	25.0												
Ceftiofur	0					29.1	68.6	2.3									
Chloramph.	9							9.3	79.1	2.3		9.3					
Enrofloxacin	3		53.5	43.0	3.5												
Florfenicol	8								88.4	2.3	1.2	8.1					
Gentamicin	0					12.8	55.8	26.7	4.7								
Nalidixic acid	1								57.0	27.9	14.0	1.2					
Neomycin	0							84.9	15.1								
Streptomycin	10								1.2	18.6	47.7	22.1	5.8	3.5		1.2	
Sulphonamide	10												38.4	41.9	9.3		10.5
Tetracycline	9						3.5	70.9	16.3				4.7	4.7			
Trimethoprim	0				19.8	67.4	12.8										

[a] The white fields denote range of dilutions tested for each substance. MICs above the range are given as the concentration closest to the range. MICs equal to or lower than the lowest concentration tested are given as the lowest tested concentration. Bold vertical lines indicate microbiological cut-off values defining resistance; [b] 16 isolates tested.

Occurrence of resistance in *E. coli* from healthy pigs sampled at slaughter, i.e. indicator bacteria, is low and has been stable over the period studied in SVARM (Table III). Resistance mostly occurs to substances that are, or have been, used in pig production. In contrast, resistance is more common in *E. coli* isolated from diagnostic submissions, although the same substances are involved (Table III). Most likely diagnostic submissions are from herds with disease problems where antimicrobial use is common. Moreover, samples taken for cultures are also likely to be from animals not responding to treatment.

This illustrates the importance of selection of isolates for testing. Data on resistance in pathogenic bacteria from diseased animals is biased by selection criteria and probably represents a worst case scenario not relevant for the situation on most farms. However such data can give early warning on emerging resistance but should be used carefully for conclusions on trends or regional differences (Aarestrup, 2004). Data on resistance in indicator bacteria, collected by defined methodology, are more suited for such purposes.

For comparison of results in a wider context, e.g. between countries, harmonized methodology, with respect to principles for collection of samples for culture, culture and identification of isolates and susceptibility testing is imperative. This applies also to interpretation criteria, i.e. the cut-off values used for classifying isolates as susceptible or resistant. Establishment of microbiological cut-off values has been initiated by the European Society for Clinical Microbiology and Infectious Diseases through the EUCAST committee (http://www.escmid.org), which facilitates harmonisation. Further, comparability is increased if antimicrobial susceptibility is reported not only as quantitative data but

also in a qualitative format i.e. distributions of MICs (see Table II) or distribution of inhibition zones.

Table III. Occurrence of resistance (%)among isolates of *Escherichia coli* from healthy pigs, years 2000, 2001, 2003 and 2005 and among isolates from diagnostic submissions year 2005.

Substance	Cut-off value (mg/L)	Resistance (%)				
		Healthy pigs				Diagnostic submissions
		2005 n=390	2003 n=303	2001 n=308	2000 n=260	2005 n=325
Ampicillin	>8	6	3	3	3	22
Ceftiofur	>1	0	0	0	0	<1
Chloramphenicol	>16	3	<1	2	<1	-
Enrofloxacin	>0.12	<1	<1	<1	0	9
Florfenicol	>16	0	0	0	0	0
Gentamicin	>4	0	0	2	2	<1
Nalidixic acid	>16	<1	1	<1	0	-
Neomycin	>8	1	1	<1	1	3
Streptomycin	>32	11	10	9	13	30
Sulphamethoxazole	>256	11	9	10	7	-
Tetracycline	>8	9	12	8	7	24
Trimethoprim	>2	6	4	3	5	-

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