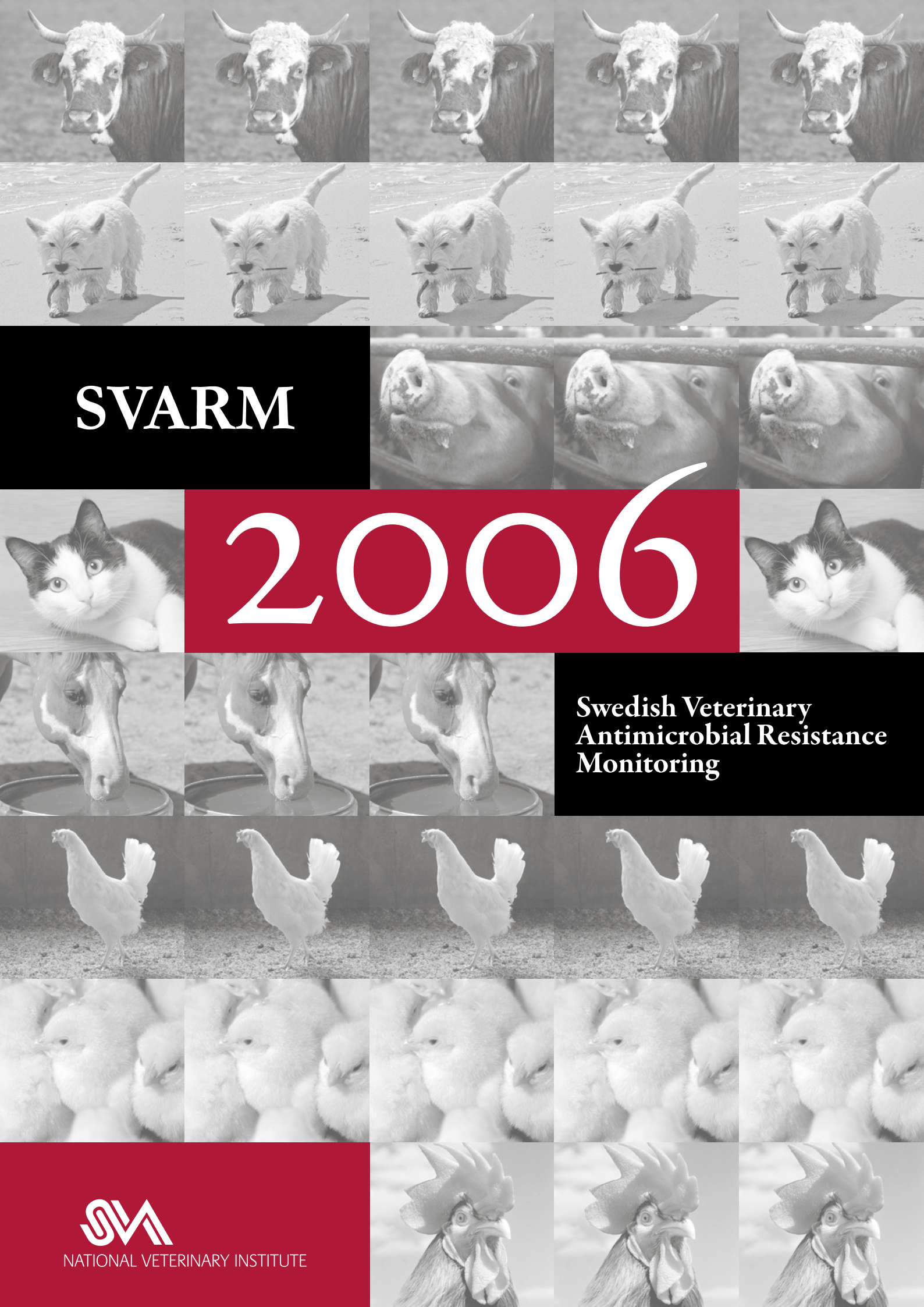

SVARM

2006

Swedish Veterinary Antimicrobial Resistance Monitoring

NATIONAL VETERINARY INSTITUTE

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Preface

Welcome to the seventh Swedish report combining results from the monitoring of antimicrobial resistance and antimicrobial usage in both veterinary and human medicine: SVARM and SWEDRES. It is today generally accepted that all use of antimicrobials in different sectors contributes to the development of resistance. This joint report will facilitate comparisons of resistance levels and incidence of use in the two areas.

According to the zoonosis-monitoring directive adopted in the EU in 2003, surveillance of antimicrobial resistance shall comprise zoonotic organisms such as *Salmonella* and *Campylobacter* isolated from food producing animals and from food. SVARM will therefore in the near future be extended to bacteria isolated from food of animal origin. This work will be initiated in collaboration between SVA and the National Food Administration. The zoonosis directive also indicates that resistance in bacteria such as *E. coli* and enterococci isolated from healthy animals and food is of public health interest. Such indicator bacteria constitute a reservoir of resistance genes that may be transferred to pathogenic bacteria in animals and man and are therefore routinely monitored in SVARM.

In 2005, SVARM was extended by SVARMpat, a monitoring programme run in collaboration between SVA and the Swedish Animal Health Service and financed by the Swedish Board of Agriculture. SVARMpat focuses on bacteria causing disease in pigs, cattle, sheep or poultry.

The year 2006 was in many respects dramatic in the area of antimicrobial resistance in veterinary medicine both on a national and on an international level. Methicillin-resistant *Staphylococcus aureus*, MRSA, were reported in pig herds and among people in contact with pigs in, e.g. The Netherlands. A screening for MRSA in Swedish pigs was therefore initiated through the SVARMpat programme. MRSA were not found among Swedish slaughter pigs.

However, in 2006 MRSA was isolated from dogs for the first time in Sweden. Furthermore, also for the first time in Sweden, methicillin-resistant *Staphylococcus intermedius*, MRSI, were found in several dogs, mainly in animal hospitals. Spread of MRSA in animal hospitals, between dogs and between personnel and dogs, is a public health problem, in particular since Sweden is still a country with comparatively low prevalence of MRSA in human health care settings. The emergence and spread of multiresistant MRSI in animal hospitals is a serious animal health problem, as the options left for treatment are scant. The spread of MRSA and MRSI among pets is most probably promoted by the heavy and increasing use of cephalosporins, fluoroquinolones and other broad-spectrum antimicrobials in small animal medicine.

A positive finding is that the increasing trend of vancomycin-resistant enterococci in Swedish broilers seems to have been broken in 2006. Further, bacteria producing extended spectrum beta-lactamases are increasingly isolated from both humans and farm animals in some European countries, but have to date not been isolated from Swedish animals.

Data in this report indicate, as also the data presented previously, that the Swedish strategies in human and veterinary medicine have been comparatively successful in containing resistance. The general concept is to use antimicrobials only when needed, on prescription by a professional only, and that the choice of treatment is based on relevant information. However, some of the presented results in both veterinary and human fields are cause for concern indicating that further efforts must be made to implement policies for prudent use of antimicrobials, increase the use of diagnostic services and to prevent infectious diseases both in human and in veterinary medicine by other means. Our hope is that this report will contribute to that work by providing updated information on the situation. The ultimate goal is to preserve the effectiveness of available antimicrobials for man and animals.

Acknowledgements

Several people have in various ways been involved in the work with SVARM. We would like to express our gratitude to all who have contributed to this report and in particular to:

Johanna Zwenson, who as her Final year project in the Veterinary programme, undertook a study on resistance in bacteria from healthy dogs. The data on indicator bacteria from dogs presented in SVARM 2006 derive from that work.

The Community Reference laboratory for antimicrobial resistance in food-borne pathogens (CRL) in Copenhagen, for confirming the resistance genotype of a cephalosporin resistant *Escherichia coli*.

Drs Luca Guardabassi and Arshene Moodley, University of Copenhagen, for PFGE analysis of methicillin resistant *Staphylococcus intermedius*.

Tack

Många personer har varit på olika sätt varit involverade i arbetet med SVARM. Vi vill tacka alla de som bidragit och särskilt:

Johanna Zwenson, som i sitt examensarbete i veterinärprogrammet genomförde en undersökning av antibiotikaresistens hos bakterier från hundar. De uppgifter om resistens hos indikatorbakterier från friska hundar som presenteras i SVARM 2006 är hämtade från detta arbete.

EU's referenslaboratorium för antimikrobiell resistens hos livsmedelsburna patogener (CRL) i Köpenhamn, för fastställande av resistensgenotyp hos ett cefalosporinresistent isolat av *Escherichia coli*.

Dr Luca Guardabassi och Dr Arshene Moodely vid Köpenhamns Universitet för PFGE-typning av meticillinresistenta *Staphylococcus intermedius*.

Summary

THE SEVENTH REPORT from SVARM shows that the situation regarding antimicrobial resistance in bacteria of animal origin is favourable from an international perspective.

The total amount of antimicrobials used for animals has declined since the mid 90s but the figures are roughly unchanged from year 2000. The amounts of antimicrobials for in-feed or in-water medication has decreased by 94 % since 1984 and is today but 13 % of the total sales. Most (68%) of the trimethoprim-sulphonamides were products for oral use in horses. The incidence of use of such products was estimated to 360–410 doses per 1000 horses. The amounts of antimicrobials for dogs and cats were substantial; 13 % of the total sales in 2006. The use of aminopenicillins, cephalosporins and fluoroquinolones is increasing which is explained by increased sales for dogs and cats.

Methicillin-resistant *Staphylococcus aureus* (MRSA) has been confirmed in dogs for the first time in Sweden. The isolates belonged to spa-type t032 and were highly resistant to fluoroquinolones. Spread of MRSA in animal hospitals is a public health problem, in particular since Sweden has a comparatively low prevalence in human health care settings. Prompted by the findings of MRSA in some other countries, a survey covering 100 Swedish slaughter pig-producing units was conducted but no MRSA were detected.

Salmonella is rare in Swedish farm animals and few incidents involve strains resistant to antimicrobials. The situation has been stable since the late 70s when monitoring began, most probably a result of the strategies in the Swedish *Salmonella* control programme. This year, only five incidents in major food producing animals involved resistant strains. Three of these strains were multiresistant *Salmonella* Typhimurium (DT104 or NT). No isolate from companion animals or wildlife was multiresistant. Resistance to fluoroquinolones or third generation cephalosporins was not observed, neither in isolates from food producing animals, nor from companion animals.

Campylobacter jejuni from dairy cows were susceptible to erythromycin, tetracycline and gentamicin but almost ten percent of the isolates were resistant to fluoroquinolones. The results tally with a previous study in calves/yearling cattle 2000. In *Campylobacter coli* from pigs, quinolone resistance is more common and found already in isolates from piglets. This suggests that selection of resist-

ant strains occurs in piglet producing herds where quinolones are used therapeutically. The situation could be similar in cattle with selection of resistant strains by use of quinolones in calves.

Indicator bacteria, i.e. *Escherichia coli* and *Enterococcus* spp. from healthy animals, are monitored since resistance in the normal gut flora reflects the antimicrobial selective pressure in an animal population. If harmonised methodology is used, data can be compared over time and on an international level. This year, indicator bacteria from healthy dairy cows and dogs were monitored. In indicator bacteria from dairy cows, resistance was rare and few isolates were multiresistant. This indicates that antimicrobial treatments of adult cattle, as applied in Sweden, exert a low selection pressure on the normal gut flora. Resistance was more common in isolates from dogs. This is in agreement with a common use of antimicrobials in this animal species, which is evident from data on prescriptions. In *E. coli*, transmissible resistance to third generation cephalosporins was observed in isolates neither from dairy cows nor from dogs.

Vancomycinresistant enterococci (VRE) were isolated from 29 of 102 samples of intestinal content from broilers. Samples were cultured on media supplemented with vancomycin. The proportion of samples positive for VRE is lower than in previous years, which indicates that the gradual increase observed since 2000 have abated.

Escherichia coli from diagnostic submissions were often resistant to ampicillin, streptomycin, tetracycline, or trimethoprim-sulphonamides, irrespective of source (pig, cattle, horse, dog, and cat). Multiresistance involving these substances was common, with a prevalence between 8 (horse) and 26% (cattle).

In *Brachyspira* spp. from pigs, resistance to tiamulin occurs among *B. pilosicoli* but was not observed in *B. hyodysenteriae*. The majority of both *B. pilosicoli* and *B. hyodysenteriae* were resistant to tylosin.

Streptococcus zooepidemicus from the respiratory tract of horses were uniformly susceptible to penicillin, but resistance to trimethoprim-sulphonamides was common.

Klebsiella spp. isolated from cows with mastitis were susceptible to most antimicrobials, and no isolate was resistant to third generation cephalosporins.

Most *Staphylococcus intermedius* from dogs were resistant to penicillins. Resistance to clindamycin, erythromycin, fusidic acid, or streptomycin was also common (between 16 to 29%) and of similar magnitude as in the last six years. In contrast, tetracycline resistance was more common than in previous years, 37%. More than one third of *S. intermedius* were multiresistant and 11% were resistant to at least five antimicrobials.

Methicillin-resistant *S. intermedius* (MRSI) in Swedish dogs were confirmed for the first time in 2006. The isolates were from post-operative wounds from two different animal hospitals suggesting nosocomial transmission. The MRSI-isolates appear to be related, indicating a clonal spread.

Sammanfattning

DEN SJUNDE SVARM-RAPPORTEN visar att läget när det gäller antibiotikaresistens hos bakterier från djur är gynnsamt ur ett internationellt perspektiv.

Den totala förbrukningen av antibiotika till djur har minskat sedan mitten av 90-talet men är i stort oförändrad från år 2000. Volymen antibiotika för inblandning i foder eller vatten har minskat med 94 % sedan 1984 och utgör idag endast 13 % av den totala försäljningen. Huvuddelen (68 %) av användningen av trimetoprim-sulfa var i form av produkter för oralt bruk till häst. Behandlingsincidensen med sådana produkter uppskattas till 360–410 doser per 1000 hästar. Den totala volymen antibiotika till hund och katt är ansenlig, och var 2006 13 % av den totala försäljningen. Den ökande användningen av aminopenicilliner, cefalosporiner och fluorokinoloner förklaras av ökad försäljning till hundar och katter.

Meticillinresistenta *Staphylococcus aureus* (MRSA) hos hund konfirmerades 2006 för första gången i Sverige. Isolaten var av spa-typ t032 och var höggradigt resistenta mot fluorokinoloner. Spridning av MRSA inom djursjukhus är ett folkhälsoproblem, särskilt som Sverige har en jämförelsevis låg prevalens inom humansjukvården. Föranlett av rapporter från andra länder om MRSA-förekomst hos gris genomfördes en studie i 100 svenska slaktsvinsbesättningar. MRSA påvisades inte i någon svinbesättning.

Salmonella är ovanligt hos svenska djur och antibiotikaresistenta stammar förekommer sällan. Sedan sent 70-tal, då resistensövervakningen påbörjades, har situationen varit stabil vilket troligen är en effekt av det svenska salmonellakontrollprogrammet. Under 2006 var endast fem av utbrotten hos de vanligare livsmedelsproducerande djurslagen orsakade av antibiotikaresistenta salmonellabakterier. Tre av dessa var orsakade av multiresistenta *Salmonella* Typhimurium (DT104 eller NT). Inget isolat från sällskapsdjur eller vildfågel var multiresistent. Resistens mot kinoloner eller cefalosporiner påvisades varken från livsmedelsproducerande djur eller från sällskapsdjur.

Campylobacter jejuni från mjölkkor var känsliga för erytromycin, tetracyklin och gentamicin men nästan 10 % av isolaten var resistenta mot fluorokinoloner. Resultaten stämmer väl med en tidigare studie av kalvar/ungnöt från år 2000. Hos *Campylobacter coli* från gris var kinolonresistens vanligare och påvisades redan hos spädgrisar. Detta talar för att selektionen av resistenta stammar sker i smågrisproducerande besättningar där kinoloner används för sjukdomsbehandling. Situationen kan vara liknande för

nötkreatur, med selektion av resistenta stammar genom användning av kinoloner till kalvar.

Resistensläget hos indikatorbakterier (*Escherichia coli* och *Enterococcus* spp.) från tarmfloran hos friska djur anses återspegla det selektionstryck som antibiotikaanvändningen i en djurpopulation innebär. Undersökningar av indikatorbakterier kan utvärderas över tid och resistensläget i olika länder jämföras om harmoniserad metodik används. I år undersöktes indikatorbakterier från mjölkkor och hundar. Bland indikatorbakterier från mjölkkor var resistens ovanlig och få isolat var multiresistenta. Detta tyder på att antibiotikabehandling av mjölkkor, som den tillämpas i Sverige, inte i någon större utsträckning selekterar för resistens hos tarmbakterier. Resistens var vanligare hos isolat från hundar vilket är i överensstämmelse med en hög förskrivning av antibiotika till detta djurslag. *Escherichia coli* med överförbar resistens mot tredje generationens cefalosporiner påvisades varken hos kor eller hos hundar.

Vankomycinresistenta enterokocker (VRE) isolerades i 29 av 102 prov av tarminnehåll från slaktkyckling. Proven odlades på odlingsmedier med tillsats av vankomycin. Andelen positiva prov är lägre än tidigare år och trenden av ökande VRE-förekomst hos slaktkyckling sedan 2000 har därmed brutits.

Escherichia coli från kliniska prov från svin, nötkreatur, hästar, hundar och katter var ofta resistenta mot ampicillin, streptomycin, tetracyklin eller trimetoprim-sulfa. Många isolat var resistenta mot flera av dessa antibiotika och därmed multiresistenta. Frekvensen multiresistens varierade beroende på djurslag och var lägst (8 %) hos isolat från hästar och högst (26 %) hos isolat från nötkreatur.

Hos *Brachyspira pilosicoli* i kliniska prov från grisar förekom resistens mot tiamulin men däremot inte bland *B. hyodysenteriae*. Majoriteten av såväl *B. pilosicoli* som *B. hyodysenteriae* var resistenta mot tylosin.

Streptococcus zooepidemicus från hästars luftvägar var genomgående känsliga för penicillin men resistens mot trimetoprim-sulfa var vanlig.

Klebsiella spp. isolerade från kor med juverinflammation var i huvudsak känsliga för antibiotika som används vid behandling av juverinflammation. Inget isolat var resistent mot tredje generationens cefalosporiner.

Staphylococcus intermedius från hundar var i stor utsträckning resistenta mot penicillin. Resistens mot klindamycin, erytromycin, fusidinsyra eller streptomycin var också vanlig (mellan 16 och 29 %) och av samma storleksordning som de senaste sex åren. Däremot var tetracyklinresistens (37 %) vanligare än tidigare år. Mer än tredjedel av *S. intermedius* var multiresistenta och 11 % var resistenta mot minst fem antibiotika.

Meticillinresistenta *S. intermedius* (MRSI) från svenska hundar konfirmerades för första gången under 2006. Isolaten var från sårinfektioner hos hundar opererade vid två olika djursjukhus. Typning med molekylärbiologiska metoder tyder på att MRSI-isolaten är besläktade, vilket talar för en klonal spridning.

Use of antimicrobials

THROUGH AN INITIATIVE of SVA and Apoteket AB (the National Corporation of Swedish Pharmacies), statistics on total sales of antimicrobials for use in animals in Sweden are available since 1980. For a review of the figures from 1980–2000 as well as references to publications on which that review is based, see SVARM 2000.

Material included

In Sweden, antimicrobials for use in animals are only available on veterinary prescription and all pharmaceuticals are dispensed by pharmacies. In 1986, the Feedstuffs Act restricted the use of antibiotics for veterinary medicinal purposes, i.e. their use as growth promoters was no longer authorised.

Drug statistics are based on sales figures provided by Apoteket AB and represent the total amount of antimicrobials authorised for veterinary use sold, calculated to kg active substance. These figures include antimicrobial formulations for all animal species (food producing animals, pets and horses etc) for systemic, intramammary and obstetric use, and intestinal anti-infectives. Drugs authorised for human use but prescribed for animals are not included. Such antimicrobials are almost exclusively prescribed in small animal medicine. In 2005, 8% of the total number of the antimicrobials prescribed for dogs was of products for human use (ATC group J01; SVARM 2005).

Up to and including year 2002, the source for the statistics has been sales of drugs from wholesalers to pharmacies. From year 2003, the statistics are based on the amounts of drugs dispensed by pharmacies and a new system for retrieval of data was introduced. In both systems, data represent an approximation on the real usage of antimicrobials, assuming that the amount sold is also used during the observation period.

Details on animal numbers are found in Appendix 1, on methodology in Appendix 2 and on antimicrobial agents with general marketing authorisation in Sweden in Appendix 4.

Overall use of antimicrobials

The total yearly sales of antimicrobials over the last decade are presented in Table AC I and in Figure AC I a & b, the long-term trends from year 1980 for classes currently used are illustrated. Figures on antimicrobials formerly used as feed additives are given for 1984 in Table AC III and for other years in SVARM 2000. In chickens, ionophoric antimicrobials are given to control coccidiosis. These substances are currently classified as feed additives, and are not included in the overall statistics based on sales from pharmacies. However, figures on the sales of these products, based on data from feed mills, are given under the section on group treatment (see Table AC III).

The potency of different antimicrobials is not equal and therefore each class should be evaluated separately. Nonetheless, the total figures may indicate trends in the material. The total amount used decreased by 13% since 1997. Most of that decrease was in the late 90s. Changes in the number of animals may affect trends in statistics on use of antimicrobials. In year 2006, the numbers of dairy cows and slaughtered pigs were 7 and 7.5% lower than in year 2002, respectively, while the number of slaughtered broilers was roughly unchanged.

The lower total figures shown for years 2003–2005 are uncertain, as there was a change in the system for data retrieval in year 2003. It is possible that initially, part of the sales of antimicrobials sold with special licence prescription was not captured by searches in the new system. Data collection for these products was made through specified searches for individual products, which in turn

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

ATCvet code	Antimicrobial class	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006
QJ01AA, QG01A	Tetracyclines[a]	2 558	2 897	2 251	1 754	1 453	1 415	1 307	1 329	1 562	1 516
QJ01CE, QJ01R, QJ51	Penicillin G-and V[b]	8 781	8 547	8 692	8 254	8 414	8 179	7 579	7 814	7 571	7 860
QJ01CA, QJ01CR	Aminopenicillins	841	824	809	852	752	767	870	875	911	920
QJ01D, QJ51CA	Other betalactams	53	133	245	315	474	676	832	928	1 009	1 217
QA07AA, QJ01G, QJ01R, QJ51R	Aminoglycosides and polymixins[a]	1 077	930	846	797	770	753	645	606	762	750
QA07AB, QJ01E	Sulphonamides	2 151	2 345	2 403	2 338	2 485	2 477	2 326	2 462	2 535	2 543
QJ01E	Trimethoprim & derivatives	352	390	397	390	414	414	381	406	437	450
QJ01F	Macrolides & lincosamides	1 747	1 846	1 467	1 352	1 510	1 412	1 124	1 095	1 080	1 254
QJ01MA	Fluoroquinolones	179	175	155	156	182	185	184	187	184	195
QJ01XX92, QJ01XX94	Pleuromutilins	1 094	1 032	847	871	841	988	744	387	338	459
QJ01MB	Quinoxalines[c]	534	-	-	-	-	-	-	-	-	-
QJ01XX91	Streptogramins	288	150	125	-	-	-	-	-	-	-
Total		**19 655**	**19 269**	**18 237**	**17 079**	**17 295**	**17 266**	**15 992**	**16 089**	**16 389**	**17 164**

[a] Includes drugs marketed with special licence prescription for years 2000–2006; [b] Calculated as benzyl-penicillin; [c] From 1986 sold only on veterinary prescription at therapeutic dosages.

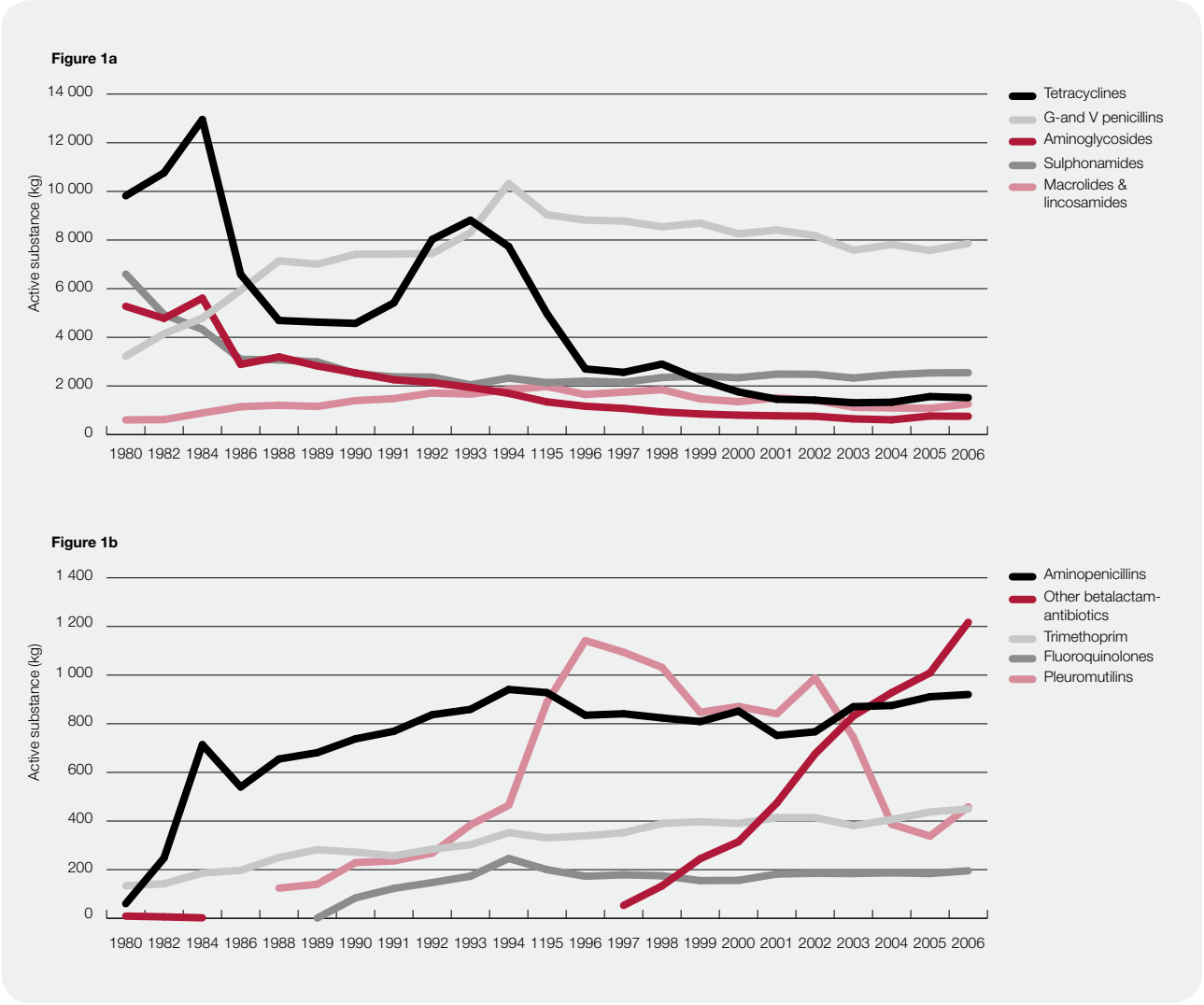

Figure AC I a & b. Sales of antimicrobials for animals from 1980–2006. Amfenicols , nitromimidazoles, streptogramins, quinoxalines and other feed additives were withdrawn from the market during the time period and are not shown. Note that the scales on the Y-axis are different in figure a and b.

depended on knowledge of what specific products to search for. This problem has been addressed, and from year 2006 all products dispensed should be captured in the searches. In year 2006, sales of products with special licence prescription amounted to 8% of the total sales, 55% of which was tetracyclines.

The total use of 'macrolides and lincosamides' and of pleuromutilins has decreased from year 1997 to year 2006 (by 28 and 41%, respectively). These antimicrobials are mainly used in pigs, and as the pig population has not changed to that extent, at least part of the observed decrease in total consumption is a true decrease in incidence of use. By contrast, the decrease noted for penicillin (10%) and aminoglycosides (30%) may be attributed to a decrease in the number of dairy cows. Penicillin is widely used for treatment of mastitis, and this was formerly also the case for penicillin in combination with aminoglycosides (dihydrostreptomycin).

In year 2006 about 13% of the total sales of veterinary products (2278 kg) were prescribed for use in dogs and cats in 'out-patient care'. Trends in the use of certain classes are highly influenced by this use. This is the case for the marked increase in use of 'other betalactams', 97% of which consisted of first generation cephalosporins in 2006. Likewise, sales of products for use in dogs and cats explain the increases of fluoroquinolones and aminopenicillins (9% increase for both classes). The use of specific antimicrobial classes is further commented in the following sections.

Route of administration

The sales of antimicrobials in QJ01 and QA07 (presented in Tables ACII and ACIII) are presented in Figure AC II as relative amounts of each class of products for injection, for oral use in individual animals (tablets, gels etc.) and for oral administration to groups of animals by mixing into feed or water. The tetracyclines, pleuromutilins and macrolides are mainly used for treatment of groups of animals, mostly pigs. Penicillins for systemic use are exclusively sold as injectables, and that type of formulation also dominates for the aminoglycosides and fluoroquinolones. These products are probably mainly used for treatment of cattle, in particular dairy cows but also for pigs and horses. Sales of broad-spectrum betalactam antimicrobials (aminopenicillins with or without clavu-

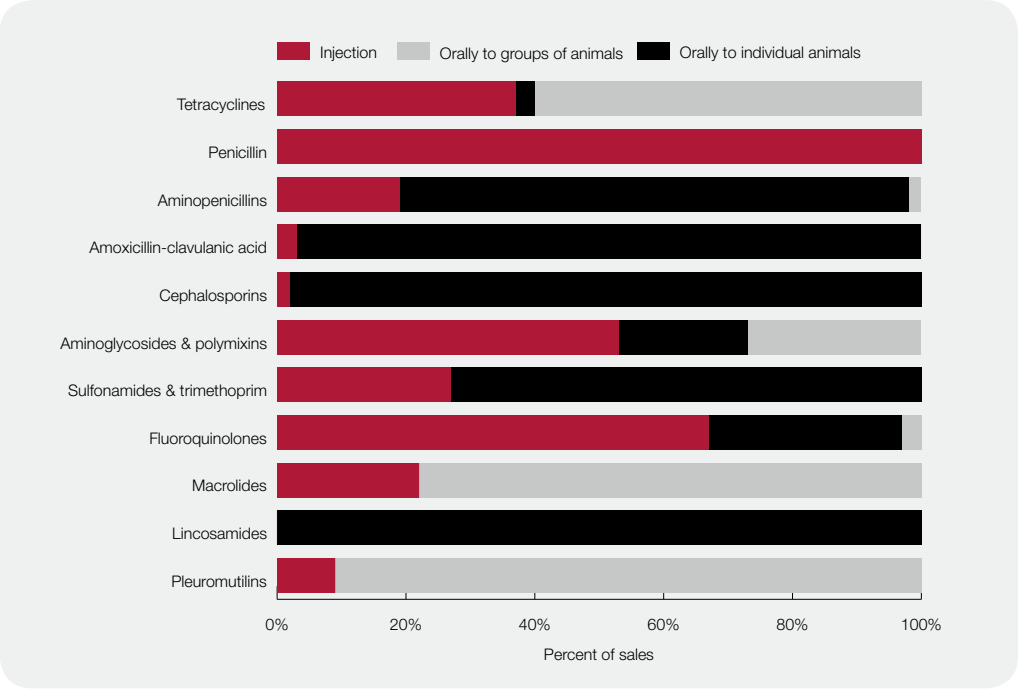

Figure AC II. Proportions of the total sales of intestinal anti-infectives and for systemic use (QA07 and QJ01) of drugs that are formulated for injection, for oral individual use or for oral use for groups of animals (amounts given in Tables AC II and AC III).

lanic acid and cephalosporins) and lincosamides are mainly or exclusively products for oral use in pets. Finally, sales of sulphonamides and trimethoprim are also largely products for oral use in individual animals, mainly for horses.

Treatment of individual animals

In table AC II, the sales of products for use in individual animals, excluding topical, intrauterine and intramammary use are presented. In year 2006, this subset was 85% of the overall use. The total sales in this subset have been unchanged over the last decade. The use of most classes of antimicrobials has decreased or been relatively unchanged over the last five years. A large part of the injectable antimicrobials is probably used for treatment of bovine mastitis. Therefore, part of the decrease in for example penicillins may be explained by a decreasing number of dairy cows. However, these formulations are also used in horses, for which reliable statistics on changes in population-size over time is not available. Therefore, trends in sales of drugs of this category should be assessed with caution.

The use of products for individual use from the classes aminoglycosides and intestinal anti-infectives (mainly aminoglycosides) has declined by 25 and 27%, respectively, since year 2002. For aminoglycosides, this trend is mainly explained by a decreased use of combinations of procaine-penicillin and dihydrostreptomycin (ATCvet code QJ01R). This is in line with current policy recommendations. For the group of 'macrolides and lincosamides', the decrease by 13% mainly derives from a declining use of injectable macrolides for use in cattle and pigs, while the lincosamides, exclusively used for pets, show a less prominent decrease.

The use of cephalosporins and fluoroquinolones for individual use has increased by 79 and 7%, respectively, from year 2002 to 2006. In both cases, the increase is derived from the amounts prescribed for dogs (see SVARM 2005 for statistics on use for dogs). Cephalosporins were introduced for veterinary use in 1997. Part of the initial increase in use of veterinary products can be ascribed to a shift from off-label of cephalosporins authorised for human use (not routinely included in the statistics presented in SVARM). In 1998 and 2005, 26 and 10% of the total number

Table AC II. Yearly sales of antimicrobial drugs authorised for individual treatment expressed in kg active substance. Only products for systemic use (QJ01) or for use as intenstinal anti-infective (QA07) are included. Based on sales statistics from Apoteket AB.

ATCvet code	Antimicrobial class	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006
QA07A	Intestinal anti-infectives[a]	706	649	607	587	614	594	594	586	496	434
QJ01A	Tetracyclines	663	656	695	634	623	628	606	611	623	609
QJ01C	Penicillins[b, c]	9 530	9 287	9 424	9 037	9 095	8 894	8 406	8 644	8 404	8 686
QJ01D	Cephalosporins	53	133	245	315	474	676	832	928	1 009	1 212
QJ01E	Sulfonamides & trimethoprim	2 107	2 335	2 376	2 336	2 478	2 483	2 280	2 427	2 610	2 689
QJ01F	Macrolides & lincosamides	652	645	559	531	522	477	430	382	400	417
QJ01G	Aminoglycosides[c, d]	617	535	528	474	454	460	367	344	362	345
QJ01M	Fluoroquinolones	147	150	144	150	169	178	177	180	179	190
QJ01X	Pleuromutilins	65	64	52	56	48	49	77	32	29	39

[a] Drugs marketed with special licence prescription are included from year 2000; [b] Procaine-penicillin calculated to benzyl-penicillin; [c] The amount includes QJ01R; [d] Does not include QA07A, intestinal anti-infectives.

Table AC III. Yearly sales of antimicrobial drugs authorised for group treatment and ionophoric anticoccidials sold expressed as kg active substance. Based on sales statistics from Apoteket AB and from the Board of Agriculture

ATCvet code	Antimicrobial class	1984	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006
QA07A	Intestinal anti-infectives[a]					-	-	-	-	-	163	170
QJ01A	Tetracyclines[b]	12 300	1 881	2 230	1 545	1 111	822	777	695	712	934	903
QJ01C	Penicillins	-				-	-	-	-	-	-	11
QJ01F	Macrolides & lincosamides	607	1 096	1 201	908	821	988	935	694	713	680	837
QJ01M	Fluoroquinolones	-	32	25	11	7	13	7	8	7	5	5
QJ01M	Quinoxalines[c]	9 900	534			-	-	-	-		-	-
QJ01XX91	Streptogramins[c]	8 800	288	150	125	-	-	-	-		-	-
QJ01XX92, QJ01XX94	Pleuromutilins	-	1 029	969	795	815	793	939	667	355	309	420
QP51AA	Nitroimidazoles	1 440	-	-	-	-	-	-	-	-	-	-
	Feed additives	700	-	-	-	-	-	-	-	-	-	-
QP51AH	Ionophoric antibiotics (coccidiostats)[e]	7 900	6 991	8 267	11 643	9 368	10 019	8 439	10 920	10 486	11 095	12 335

[a] Drugs with special licence prescription are included from year 2005; [b] Drugs marketed with special licence prescription are included from year 2000; [c] Years 1980–1984 sold as feed additives, thereafter on veterinary prescription at therapeutic dosages; [d] Feed additives other than quinoxalines and streptogramins: avoparcin, bacitracin, nitrovin, oleandomycin and spiramycin; [e] From 1999 regulated and classified as feed additives (dir 70/524/EEC).
Figures from 1999 and onwards are from the Feed Control of the Board of Agriculture (www.sjv.se).

of prescriptions of cephalosporins for dogs were products authorised for human use (SVARM 2005). However, prescription data support the observation of prominent increase in use of this class, as the total number of prescriptions of cephalosporins for dogs was 24 400 in 1998 compared with 52 400 in 2005.

In year 2006, 68% of the sales of sulphonamides and trimethoprim were products for oral use in horses (paste or powder). This type of products was introduced on the market in the late 80s, and since, most of the increasing trend in use of trimethoprim and sulphonamides (Figure ACI a & b) is derived from that type of products. In year 2006, a total number of 107 000 doses (dose applicators or powders) of this type of products was sold. The equine population was estimated to around 265 000–300 000 horses in year 2004. Using that figure, the approximate incidence of use is 360–410 doses/1000 horses or 1–1.1 doses/1000 horses and day. This is more than in out patient care in human medicine, where sales of trimethoprim alone or in combination with sulphonamides amounted to 0.7 DDD/1000 inhabitants and day in 2006 (SWEDRES 2006). Trimethoprim-sulphonamides is the only class of antimicrobials available for oral use in adult horses, and the convenience of the route of administration probably influences the veterinarian's choice of treatment.

Treatment of groups or flocks

When considering the risk for development of resistance, the consumption of antimicrobials intended for group or flock medication, e.g. administration via feed or water, is of special interest. Figures on sales of that subset of drugs over the last decade are given in Table AC III. As a reference, figures for 1984, the last year before the ban of antimicrobials feed additives (growth promoting use), are given. More complete data sets for previous years are available in SVARM 2000. From year 2005, products of the class 'intestinal anti-infectives' that are sold with a special licence prescription are included. The active substances in products in that group are currently neomycin or colistin.

The proportion of products intended for medication of groups of animals of the total use of antimicrobials has decreased steadily over the years and is today but 13% of the total sales (total sum of Table AC III divided by total sum of Table I). The sales of this particular subset have decreased by 94% since 1984.

Products for group treatment are mainly used in pigs. The number of pigs slaughtered has decreased by 22% over the last 10 years and by 7% from year 2002 to 2006. Part of the observed decreases in the last decade can therefore probably be attributed to a decrease in the population of pigs.

The sales of pleuromutilins has decreased by 59% over the last 10 years. Pleuromutilins (tiamulin, valnemulin) are only authorised for use in pigs, with swine dysentery as the main indication. It is probable that efforts to control the disease have resulted in a decreased need to treat swine dysentery, leading to declining sales figures. Increases in use of pleuromutilins in occasional years, such as seen in year 2002 and 2006, may be related to a temporary, but extensive, use within programmes for eradication of swine dysentery. The sales of macrolides has also decreased, by 24% since 1997, a figure that comes close to the decrease in number of pigs slaughtered.

The observed decrease in use of tetracyclines over the last five years is confounded by an increased use of doxycycline. Doxycycline has a higher bioavbailability, and the dose is lower (250 ppm when mixed in feed) compared with that for, e.g. chlortetracycline (1000 ppm when mixed in feed). The use of doxycycline has increased steadily over the last six years. When the sales figures are corrected for the lower dose of doxycycline, the use of tetracyclines decreased by 48% from years 1996 to 2000 but then increased by 17% from year 2000 to 2006. Corrected both for dose and for population size, the increase from year 2000 to 2006 is 26%. This is largely explained by an increased use of doxycycline for treatment of respiratory diseases. However, it is probable that it partly also reflects increased off-label use for, e.g. intestinal spirochetosis.

Coccidiostats of the ionophore group are used as feed additives to control coccidiosis in the production of chickens for slaughter. Since the late 80s, narasin is by far the most widely applied substance.

Resistance in zoonotic bacteria

ZOONOSES are diseases and infections that can be naturally transmitted between animals and man. Antimicrobial resistance in zoonotic bacteria is therefore of public health concern. For that reason, susceptibility of all *Salmonella* from reported incidents in warm-blooded animals is monitored yearly in SVARM. In addition, selected *Campylobacter* from animals are tested for antimicrobial susceptibility. This year, *Campylobacter* spp. from dairy cows were tested. More information on infections with these bacteria in Sweden is presented in the yearly report, Zoonoses in Sweden available at www.sva.se.

Some microbiological cut-off values defining resistance (breakpoints) used in SVARM 2000–2005 have been changed. To facilitate comparisons when data from these reports are presented here, levels of resistance have been recalculated using current cut-off values.

Salmonella

Isolates included

Findings of *Salmonella* in animals are notifiable in Sweden and isolates from each incident are confirmed at SVA. From each notified incident 2006, one isolate from each involved warm-blooded animal species (wild and domesticated) was tested for antimicrobial susceptibility. If an incident involved more than one serovar or phage type, one isolate of each serovar and phage type was tested. For details on methodology, see Appendix 3.

Antimicrobial susceptibility of *Salmonella* from Swedish animals has been monitored regularly since 1978. Tested antimicrobials have varied, but microdilution methods have been used in all surveys. Data from previous years are therefore presented for comparison to data for 2006.

Results and comments

This year, 82 isolates from 79 notified incidents were tested. About half (56%) of the isolates were from major food-producing animals (cattle, pigs and poultry) and the majority (64%) were *S.* Typhimurium (Table Salm I). Occurrence of resistance and distributions of MICs and are given in Table Salm II-III.

The majority of isolates (89%) were susceptible to all antimicrobials tested but nine isolates, all *S.* Typhimurium, were resistant to at least one substance. One isolate, not phage typed, was from a dog and resistant to ampicillin only. One isolate of phage type DT 104 from an incident in cattle had the typical penta-resistance; ampicillin, sulphonamides, streptomycin, tetracycline and chloramphenicol/florfenicol. Two isolates from epidemiologically linked incidents in cattle and pigs were of phage type DT 104 and resistant to ampicillin and sulphonamides. Finally, five isolates, not possible to phage type (NT), were from four incidents involv-

Table Salm I. Number of *Salmonella enterica* tested year 2006 presented by serovar and source.

Serovar	Cattle[a]	Pig	Poultry	Horse	Dog	Cat	Wildlife	Total
Agona	1	3	1		2			7
Braenderup		1						1
Dublin	10							10
Livingstone	1							1
Senftenberg			1					1
Duesseldorf	1							1
Worthington			2					2
Thompson	1							1
Typhimurium DT 104	2	1						3
Typhimurium DT 120		1		1				2
Typhimurium DT 40		2				1		3
Typhimurium DT NST	1		3	1				5
Typhimurium DT NT	2	1	3					6
Typhimurium DT 10	2							2
Typhimurium, not phage typed		3		1	1	14	13	32
Bredeney					1			1
Liverpool					1			1
Oranienburg		1						1
Paratyphi var Java			1					1
Rubislaw			1					1
Total	**21**	**13**	**12**	**3**	**5**	**15**	**13**	**82**
Percent of total	26	16	15	4	6	18	16	

[a] One isolate of *S.* Duesseldorf and one isolate of *S.* Dublin not available for testing.

ing; cattle, cattle and horses, ducks for food production, and ducks in a hobby flock. These isolates had the same resistance phenotype including resistance to ampicillin, sulphonamides, streptomycin, and tetracycline. Possible epidemiological links between these incidents are being investigated.

From a public health perspective, the prevalence of resistance in *Salmonella* from food-producing animals is more important than resistance in isolates from wild animals or pets. In SVARM, 240 isolates from incidents in food-producing animals have been tested the years 2000 to 2006. In certain years, occasional isolates were unavailable for testing but the material includes the vast majority of isolates involved in notified incidents in food-producing animals in the period. Of the isolates tested, 101 (42%) were *S.* Typhimurium. About half of these were from pigs (52%), and about one fourth from cattle (24%) and poultry (22%), respectively. Two isolates (2%) were from sheep. Eighteen isolates of *S.* Typhimurium (8% of total) and 14 of other serovars (6% of total) were resistant to at one least antimicrobial. Data on *S.* Typhimurium are given in Table Salm V & IV.

Nine of the 100 incidents of *S.* Typhimurium in food-produc-

Table Salm II. Distribution of MICs for all serovars of *Salmonella enterica* (n=82) from animals in 2006.

	Resis-tance	Distribution (%) of MICs[a] (mg/L)																			
Antimicrobial	(%)	≤0.008	0.016	0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	2048	>2048
Ampicillin	11							6.1	26.8	43.9	12.2				11.0						
Cefotaxime	0				14.6	47.6	35.4	2.4													
Ceftiofur	0					1.2	7.3	23.2	65.9	2.4											
Chloramphenicol	1									15.9	63.4	19.5					1.2				
Ciprofloxacin	0			13.4	86.6																
Florfenicol	1										80.5	18.3		1.2							
Gentamicin	0							23.2	68.3	8.5											
Kanamycin	0									14.6	72.0	13.4									
Nalidixic	0										74.4	25.6									
Streptomycin	7											14.6	63.4	14.6		1.2	3.7	2.4			
Sulphonamide	10													9.8	26.8	39.0	14.6				9.8
Tetracycline	7								28.0	64.6			1.2			6.1					
Trimethoprim	0						20.7	56.1	18.3	4.9											

[a] White fields denote range of dilutions tested. Values above the range denote MICs greater than the highest concentration tested. MICs equal to or lower than the lowest concentration tested are given as the lowest tested concentration. Vertical lines indicate cut-off values for resistance.

Table Salm III. Distribution of MICs for *Salmonella* Typhimurium (n=53) from animals in 2006.

	Resis-tance	Distribution (%) of MICs[a] (mg/L)																			
Antimicrobial	(%)	≤0.008	0.016	0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	2048	>2048
Ampicillin	17								28.3	39.6	15.1				17.0						
Cefotaxime	0				9.4	50.9	35.8	3.8													
Ceftiofur	0							20.8	75.5	3.8											
Chloramphenicol	2									11.3	75.5	11.3					1.9				
Ciprofloxacin	0			7.5	92.5																
Florfenicol	2										90.6	7.5		1.9							
Gentamicin	0							22.6	71.7	5.7											
Kanamycin	0									17.0	75.5	7.5									
Nalidixic	0										79.2	20.8									
Streptomycin	11											13.2	71.7	3.8		1.9	5.7	3.8			
Sulphonamide	15														18.9	43.4	22.6				15.1
Tetracycline	11								22.6	66.0			1.9			9.4					
Trimethoprim	0						20.8	50.9	24.5	3.8											

[a] White fields denote range of dilutions tested. Values above the range denote MICs greater than the highest concentration tested. MICs equal to or lower than the lowest concentration tested are given as the lowest tested concentration. Vertical lines indicate cut-off values for resistance.

Table Salm IV. Resistance (%) and source of isolates in *Salmonella* Typhimurium from animals 1978 to 2006.

		Resistance (%)						
Antimicrobial	**Cut-off value** (mg/L)	**1978–88**[a] (n=125)	**1989–99** (n=317)	**2000–02** (n=108)	**2003** (n=49)	**2004** (n=49)	**2005** (n=85)	**2006** (n=53)
Ampicillin	>4	2[b]	6[b]	3	0	8	9	17
Cefotaxime	>0.5	-	-	-	-	-	0	0
Ceftiofur	>2	-	-	0	0	0	0	0
Chloramphenicol	>16	4[b]	5[b]	3	0	8	9	2
Ciprofloxacin	>0.06	-	-	-	-	-	-	0
Enrofloxacin	0.25	-	1	0	0	0	1	-
Florfenicol	>16	-	-	3	0	6	8	2
Gentamicin	>2	-	0[b]	0[c]	2	0	0	0
Kanamycin	>16	-	-	-	-	-	-	0
Nalidixic acid	>16	-	-	4	0	0	1	0
Neomycin	>4	0[b]	1[b]	4	0	0	0	-
Streptomycin	>32	74	15	4	2	8	10	11
Sulphonamide	>256	-	-	3	2	8	10	15
Tetracycline	>8	13	6	3	0	8	9	11
Trimethoprim	>2	-	-	0	0	0	0	0
Trim/sulph.	>0.5/9.5	0	3	-	-	-	-	-
Percent of isolates from:								
Cattle, sheep, pigs, poultry		100	46	45	12	33	19	40
Horses, cats, dogs			29	36	82	61	58	36
Wildlife			25	19	6	6	23	24

[a] 1988 includes isolates to September, isolates from October-December 1988 given under 1989; [b] Cut-off value for resistance >8 mg/L; [c] Cut-off value for resistance >4 mg/L.

Table Salm V. Distribution of MICs for *Salmonella* Typhimurium (n=101) from food-producing animals years 2000–2006.

		Distribution (%) of MICs[a] (mg/L)																			
Antimicrobial	**Resistance** (%)	**≤0.008**	**0.016**	**0.03**	**0.06**	**0.12**	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**1024**	**2048**	**>2048**
Ampicillin	12								51.5	33.7	3.0				11.9						
Cefotaxime[b]	0				6.1	63.6	30.3														
Ceftiofur	0							28.7	68.3	3.0											
Chloramphenicol	6									8.9	79.2	5.9				2.0	4.0				
Ciprofloxacin[c]	0			15.0	85.0																
Enrofloxacin[d]	0				54.3	42.0	3.7														
Florfenicol	5										89.2	5.0	1.0	5.0							
Gentamicin	4							11.9	63.4	20.8	4.0										
Kanamycin[c]	0										85.0	15.0									
Nalidixic acid	1										62.4	24.8	11.9	1.0							
Neomycin[d]	0									84.0	16.0										
Streptomycin	11										1.0	16.8	53.5	17.8	4.0	2.0	3.0	2.0			
Sulphonamide	13														34.7	41.6	10.9				12.9
Tetracycline	10								5.0	72.3	12.9		1.0		4.0	5.0					
Trimethoprim	0						23.8	62.4	13.9												

[a] White fields denote range of dilutions tested. Values above the range denote MICs greater than the highest concentration tested. MICs equal to or lower than the lowest concentration tested are given as the lowest tested concentration. Vertical lines indicate cut-off values for resistance; [b] 33 isolates tested; [c] 20 isolates tested; [d] 81 isolates tested.

ing animals years 2000–06, involved multiresistant strains, i.e. resistance to at least three antimicrobials. All nine incidents occurred among the 48 incidents reported in years 2004–06, whereas none of 52 incidents reported in 2000 to 2003 involved multiresistant strains. Of the incidents with multiresistant strains, six occurred in cattle, one in ducks for food production, and one in ducks in a hobby flock. Finally, one isolate was detected on routine sampling of pig carcasses at slaughter but *Salmonella* was not confirmed on the farm of origin. Three of the incidents in cattle were epidemiologically linked through trade of calves.

Multiresistant strains in food-producing animals were reported also before year 2000. In 1997 to 1999, five of 51 incidents of *S.* Typhimurium involved multiresistant DT 104 or DT 193. Considering the small number of incidents of *S.* Typhimurium in food-producing animals each year, the cluster of incidents with multiresistant strains 2004 to 2006 is probably coincidental and not an indication of an overall increased occurrence. Nevertheless, in view of the public health consequences of multiresistant *Salmonella* vigilance towards such strains in food-producing animals is warranted.

From an international perspective, the overall situation of *Salmonella* among Swedish animals is favourable and resistance rare. Swedish food-producing animals are virtually free from *Salmonella*, most likely a result of the strategies in the Swedish *Salmonella* control programme, and few incidents involve multiresistant strains. In isolates from cats, dogs, horses, and wildlife, only eight of 239 *S.* Typhimurium tested years 2000 to 2006 were multiresistant. None of these isolates were from wildlife.

Table Salm VI. Resistance phenotypes and multiresistance (%) of *Salmonella* Typhimurium (n=101) from food-producing animals years 2000–2006. All isolates tested for susceptibility to ampicillin, ceftiofur, enrofloxacin/ciprofloxacin, florfenicol, gentamicin, chloramphenicol, nalidixic acid, streptomycin, sulphamethoxazole, tetracycline, and trimethoprim.

		Phage type														
Resistance pattern[a]	Animal species	104	120	195	193	41	40	15a	10	12	9	1	NT	NST	Not typed	Total
AmFfCmSmSuTc	Cattle	3	1													4
AmFfCmSmSuTc	Pigs	1														1
AmCmSmSuTc	Cattle	1														1
AmSmSuTc	Cattle												2			2
AmSmSuTc	Poultry												2			2
AmSu	Cattle	1														1
AmSu	Pigs	1														1
SmSu	Poultry							1								1
Nal	Pigs									1						1
Gm	Cattle					1										1
Gm	Pigs						1									1
Gm	Poultry					1								1		2
Susceptible	Cattle	1	2			2		1	2				1	5	1	15
Susceptible	Pigs	1	3			7	23			2		1	2	5	5	49
Susceptible	Poultry			1	1	2	1			1	1	1	1	8		17
Susceptible	Sheep														2	2
Number of isolates		**9**	**6**	**1**	**1**	**13**	**25**	**2**	**2**	**4**	**1**	**2**	**8**	**19**	**8**	**101**
Percent of total		8.9	5.9	1.0	1.0	12.9	24.8	2.0	2.0	4.0	1.0	2.0	7.9	18.8	7.9	
Multiresistance (%)																
Susceptible to all antimicrobials		22.2	83.3	100.0	100.0	84.6	96.0	50.0	100.0	75.0	100.0	100.0	50.0	94.7	100.0	82.2
Resistant to one antimicrobial						15.4	4.0			25.0				5.3		5.0
Resistant to two antimicrobials		22.2						50.0								3.0
Resistant to three antimicrobials																
Resistant to >three antimicrobials		55.6	16.7										50.0			9.9

[a] Am: ampicillin; Ff : florfenicol; Cm: chloramphenicol; Sm: streptomycin; Su: sulphonamides; Tc: tetracycline; Nal: nalidixic acid; Gm: gentamicin.

Campylobacter

Isolates included

Samples for culture of *Campylobacter* spp. were selected from the total number of samples of colon content from dairy cows collected at abattoirs for isolation of indicator bacteria. Isolates were identified as *Campylobacter jejuni* or as hippurate-negative thermophilic *Campylobacter* spp. Antimicrobials included in the test panel and concentration ranges are given in Table Camp I. For details on methodology and sampling strategy, see Appendix 3.

Results and comments

Campylobacter were isolated from 15% of the 460 samples cultured. All isolates (n=68) were *C. jejuni*. Most isolates (90%) were susceptible to all antimicrobials tested but seven isolates were resistant. Of these, one isolate was resistant to ampicillin and six to quinolones/fluoroquinolones (enrofloxacin/nalidixic acid).

Interestingly, one isolate was resistant to nalidixic acid (MIC 64 mg/L) but susceptible to enrofloxacin (MIC 0.5 mg/L). Usually quinolone resistance in *Campylobacter* is due to single base mutations of the *gyrA* gene conferring resistance both quinolones (e.g. nalidixic acid) and fluoroquinolones (e.g. enrofloxacin, ciprofloxacin) but single mutations conferring resistance to only nalidixic acid were recently described (Jesse *et al.*, 2006).

The results from this year's survey tally with data on isolates from yearling cattle sampled year 2000 (Table Camp I). Resistance to erythromycin or tetracycline was not observed in this year's material, or among isolates from year 2000. In both surveys, only one isolate was resistant to more than one antimicrobial. The isolate, from 2000, was resistant to ampicillin and gentamicin.

Table Camp I. Distribution of MICs for *Campylobacter jejuni* from dairy cows (n=68) 2006. Data for yearling cattle (n=67) from SVARM year 2001 are given for comparison.

			Distribution (%) of MICs[a] (mg/L)													
Antimicrobial (%)	Year	Resistance	≤0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	>128
Ampicillin	-06	1					10.3	4.4	45.6	32.4	5.9			1.5		
	-00	6					7.5	4.5	38.8	40.3	3.0		1.5	4.5		
Enrofloxacin	-06	7		4.4	57.4	20.6	10.3	1.5		2.9	2.9					
	-00	1	3.0	22.4	61.2	11.9				1.5						
Erythromycin	-06	0			1.5	5.9	41.2	42.6	5.9	2.9						
	-00	0			3.0	7.5	34.3	34.3	19.4	1.5						
Gentamicin	-06	0				2.9	54.4	42.6								
	-00	3				1.5	25.4	70.1	3.0							
Nalidixic acid	-06	9								35.3	45.6	10.3		1.5	2.9	4.4
	-00	1						1.5	22.4	52.2	19.4	3.0			1.5	
Tetracycline	-06	0				94.0	6.0									
	-00	0				97.1		2.9								

[a] White fields denote range of dilutions tested. Values above the range denote MICs greater than the highest concentration tested. MICs equal to or lower than the lowest concentration tested are given as the lowest tested concentration. Vertical lines indicate cut-off values for resistance.

Antimicrobial resistance in *Campylobacter* spp. from pigs of different ages

ANTIMICROBIAL RESISTANCE in *Campylobacter* spp. from healthy animals is studied yearly in SVARM. In three previous surveys, between 17 and 30% of isolates from healthy slaughter pigs (about 6 months old) were resistant to quinolones (enrofloxacin and nalidixic acid). Resistance to other antimicrobials was rare. The high prevalence of quinolone resistance is remarkable since neither quinolones nor fluoroquinolones are authorised or used for treatment of groups of pigs via feed or water in Sweden. Injectables, i.e. enrofloxacin and danofloxacin, are authorised, however.

Detailed consumption statistics are not available and therefore the extent of fluoroquinolone usage in pigs is unknown. Injectables are unlikely to be used in fattening enterprises to pigs older than 12 weeks, but probably to some extent in piglet producing herds in treatment of diarrhoea or respiratory disease in piglets or the mastitis-metritis-agalactia syndrome in sows. Thus, the selection for quinolone resistance in *Campylobacter* likely occurs in younger pigs and/or sows and resistant isolates should therefore be present already in piglets.

To evaluate this hypothesis, faecal samples were collected by convenience from 36 piglet-producing herds from all parts of Sweden in the period April to June 2006. In each herd, one suckling (<5 weeks old) and one weaned piglet (5–12 weeks old) were sampled. Methods for culture, identification, and susceptibility testing are described in Appendix 3. Twenty-two isolates were obtained from suckling piglets and 24 from weaned piglets. All isolates were hippurate negative *Campylobacter* spp., probably *Campylobacter coli*.

In *Campylobacter* from suckling or weaned piglets, resistance to ampicillin or gentamicin was not observed and erythromycin or tetracycline resistance was rare (Table Camp II). This is in agreement with data for isolates from slaughter pigs studied 2005. Resistance to fluoroquinolones was common, however, and the prevalence was higher than among isolates from the older slaughter pigs. This supports the hypothesis that selection of quinolone resistant *Campylobacter* occurs in piglet producing herds before pigs are moved to the finishing stage, most likely as a consequence of use of enrofloxacin or danofloxacin in piglets or sows.

Table Camp II. Distribution of MICs for *Campylobacter* spp. from suckling or weaned pig (n=46) 2006. Data for slaughter pigs (n=97) 2005 given for comparison (SVARM 2005).

		Resistance	Distribution (%) of MICs[a] (mg/L)													
Antimicrobial	Age	(%)	≤0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	>128
Ampicillin	Suckling/Weaned	0					4.3	6.5	10.9	45.7	30.4	2.2				
	Slaughter	5					1.0	16.5	20.6	46.4	10.3		4.1		1.0	
Enrofloxacin	Suckling/Weaned	39		8.7	45.7	6.5			2.2	21.7	15.2					
	Slaughter	24		17.5	43.3	13.4	2.1		1.0	14.4	8.2					
Erythromycin	Suckling/Weaned	7				2.2	4.3	19.6	28.3	30.4	8.7		6.5			
	Slaughter	0				1.0	5.2	24.7	41.2	24.7	2.1	1.0				
Gentamicin	Suckling/Weaned	0					2.2	80.4	17.4							
	Slaughter	0					4.1	52.6	43.3							
Nalidixic acid	Suckling/Weaned	37								26.1	28.3	6.5	2.2	4.3	26.1	6.5
	Slaughter	24								7.2	44.3	21.6	3.1	2.1	17.5	4.1
Tetracycline	Suckling/Weaned	2				50.0	32.6	10.9	4.3	2.2						
	Slaughter	4				60.8	20.6	9.3	5.2	2.1	1.0			1.0		

[a] 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 epidemiological cut-off values defining resistance

Methicillin-resistant *Staphylococcus aureus* in animals

IN OCTOBER 2006, the two first cases of infection with methicillin-resistant *Staphylococcus aureus* (MRSA) in dogs were confirmed at SVA. Both dogs had post-operative wound infections, were sampled at the same animal-hospital and the isolates were referred to SVA for confirmation by a private laboratory. Minimum inhibitory concentrations (MIC) of oxacillin with 2% NaCl and of cefoxitin were > 16 mg/L. In addition, the isolates were highly resistant to ciprofloxacin (MIC>16 mg/L). Both isolates were positive for the *mec*A and nuc genes by polymerase chain reaction. Analyses performed by the Swedish Institute for Disease Control showed that they were negative for genes encoding the PVL-toxin and of spa-type t032. People in contact with the dogs (owners and their families, personnel at the animal hospital) were contacted by the County Medical Officer and an investigation was initiated (see SWEDRES 2006).

In the first quarter of 2007, two new cases were confirmed. These cases were sampled at another animal hospital, in another county, but both dogs had wound infections and the antibiogram and spa-type of the isolates were identical to the previous cases. As for the first cases, the County Medical Officer initiated an investigation.

The finding of MRSA in dogs is in line with reports from other countries with high, but also with low prevalence of MRSA in people. Generally, the subtypes found in dogs are those that are most common in hospitals in the region concerned.

S. aureus from healthy and diseased dogs

In a special survey February to September 2006, 299 healthy dogs were sampled by swabbing the perineal region. *Staphylococcus aureus* was isolated from four dogs, but none of these was methicillin-resistant (Zwenson, 2007). During years 2004–2006, a total of 5293 samples taken from sites were staphylococci are likely to be isolated, such as skin, wounds and ear canal of dogs were processed for routine bacteriology at SVA. A total of 2679 staphylococci were reported from these samples, with *S. intermedius* being by far the most common species (79%). *S. schleiferi* subspecies coagulans was the second most common (14%), and *S. aureus* represented only 5% of the total number of identified staphylococci. This is in line with the general knowledge that *S. aureus* is not commonly isolated from infections in dogs.

Screenings performed in food-producing animals

In the fall of 2006 and early spring 2007, a screening of pigs for MRSA was performed in collaboration with the Swedish Animal Health Services. In each of 100 slaughter pig production units distributed across the country, samples were taken from the nostrils from five pigs in five different pens (see Appendix 3 for methods). In previous years, samples of milk from dairy cows have been screened for MRSA on several occasions. Further, in 2003 the Food Production Agency screened 200 *S. aureus* isolated from chicken carcasses. Hitherto, all samples from food-producing animals have been negative for MRSA.

An emerging zoonosis

Sweden is still a country with comparatively low prevalence of MRSA in people but the annual number of reported cases is increasing both in hospitals and in the community (SWEDRES 2006). The findings of MRSA in dogs highlighted above clearly show that the situation in veterinary medicine needs to be watched closely. More sampling for bacteriology is needed, susceptibility testing should be performed according to international standards, and should always include antimicrobials that are appropriate for detection of methicillin resistance in staphylococci. The current recommendation from SVA is that all suspected isolates should be sent for confirmatory testing to the reference laboratory, and both the County Veterinary Officer and the County Medical Officer should be informed of any confirmed cases. Further, reports from other countries on MRSA in food-producing animals, including horses, call for continued surveillance in Sweden as in other countries.

Resistance in indicator bacteria

THE PREVALENCE of acquired antimicrobial resistance in bacteria of the normal enteric microflora indicates the magnitude of the selective pressure exerted by use of antimicrobials in a population. Although these bacteria are unlikely to cause disease, they form a reservoir for transferable resistance determinants from which resistance genes can spread to bacteria that cause infections in animals or humans. Monitoring of resistance in indicator bacteria from the normal enteric microbiota from healthy animals is therefore of great value to detect trends and to follow effects of interventions. In SVARM, *Escherichia coli* and *Enterococcus* spp. from healthy animals serve as indicator bacteria.

Of special interest is the occurrence of specific patterns of resistance in indicator bacteria. Such patterns, or phenotypes, can indicate that resistance genes are located on the same genetic element. Thereby, a single transfer event can convey resistance to several antimicrobials to a recipient bacterium (co-transfer). Thus, use of one antimicrobial can select for resistance to other, unrelated antimicrobials (co-selection).

In 2006, antimicrobial susceptibility of isolates from dairy cows and dogs was monitored. In addition, occurrence of vancomycin resistant enterococci (VRE) in broilers was investigated using selective cultures.

Some cut-off values defining resistance (breakpoints) used in SVARM 2000–2005 have been changed. To facilitate comparisons, data on prevalence of resistance from these reports have been recalculated using current cut-off values.

Isolates included

Escherichia coli and *Enterococcus* spp. from dairy cows and dogs were isolated from caecal or colon content and from rectal swabs, respectively. Dairy cows were sampled at slaughter and dogs at dog shows. For detection of VRE, caecal content from broilers sampled at slaughter was cultured on vancomycin-supplemented media. Each sample from dairy cows and dogs were from unique herds or households, respectively. Samples from broilers were from unique flocks but not necessarily from unique production sites. Antimicrobials tested and concentration ranges used are given in Table EC IV and ENT VII. For details on methodology, including sampling strategy, see Appendix 3.

Escherichia coli

Dairy cows

Escherichia coli were isolated from 86% of 365 samples cultured. The majority (97%) of the 314 isolates were sensitive to all 13 antimicrobials tested. Only nine isolates were resistant to at least one substance. Resistance to streptomycin, sulphonamides or tetracycline were the most common traits (2%) (Table EC I). Two isolates (<1%) were quinolone resistant (nalidixic acid and ciprofloxacin) and one isolate was resistant to trimethoprim. No isolate was resistant to ampicillin, third generation cephalosporins (ceftiofur or cefotaxime), amphenicols (chloramphenicol or florfenicol), gentamicin, or kanamycin.

Multiresistance was rare, only five isolates (2%) were resistant to more than one antimicrobial, and four of these to more than two substances (Table I). The phenotypes of all four isolates included resistance to sulphonamides, streptomycin, and tetracycline, and one isolate was, in addition, resistant to trimethoprim.

Dogs

Escherichia coli were isolated from 86% of 299 samples cultured. The majority (87%) of the 257 isolates were sensitive to all 13 antimicrobials tested but 34 isolates (14%) were resistant to at least one substance. Streptomycin, sulphonamides, ampicillin or trimethoprim were the most common traits (4–7%) (Table EC I). Resistance to quinolones (ciprofloxacin and nalidixic acid), tetracycline, kanamycin, and chloramphenicol was less common (2%) and only one isolate was resistant to third generation cephalosporins (ceftiofur and cefotaxime). Resistance to gentamicin and florfenicol was not detected.

About 8% of the isolates were resistant to more than one antimicrobial and 4% to more than two substances (Table EC I). Ampicillin, trimethoprim, or sulphonamides were the most common traits among isolates resistant to three or more substances (Table EC II).

The isolate resistant to third generation cephalosporins had MICs for cefotaxime and ceftiofur of >2mg/L and 2mg/L, respectively. Resistance was caused by hyperproduction of AmpC due to up-regulation of the *AmpC* gene caused by a mutation in the promoter region of the gene. The genotype was confirmed by sequencing of the gene at the Community reference laboratory for antimicrobial resistance in food-borne pathogens, Copenhagen, Denmark.

Comments

Resistance in *E. coli* from dairy cows is rare in agreement with the results for calves/yearling cattle year 2000. This indicates that selection towards resistance in adult cattle is limited, probably because these animals usually are treated individually with injectables or intramammaries, mainly using penicillin (Landin, 2006). The higher occurrence of resistance in commensal *E. coli* from younger calves, <6 weeks, reported in SVARM 2004 is probably due to a higher treatment incidence and a more extensive use of other antimicrobials than penicillin, e.g. tetracycline, trimethoprim-sulpha or enrofloxacin, in cattle of this age group. Moreover, in calves, group treatment is sometimes applied and antimicrobials occasionally administered orally.

Resistance was more common in *E. coli* from dogs than in isolates from dairy cows (Fig EC I). This indicates the presence of a selection pressure, which is in agreement with the high use of antimicrobials in dogs reported in SVARM 2005. Aminopenicillins are the most frequently prescribed antimicrobials, and accordingly ampicillin resistance is among the most common resistance traits (5%). Since the resistance phenotypes of multiresistant strains often include ampicillin, use of aminopenicillins probably selects also for resistance to other substances. Fluoroquinolones are also

commonly used in dogs and resistance to these antimicrobials occurs to some extent (2%) in *E. coli* from healthy dogs. Resistance in *E. coli* from dogs with urinary tract infections involves the same antimicrobials as in commensals but the prevalence is much higher and sometimes limits the therapeutic alternatives (see Resistance in animal pathogens).

The prevalence of resistance in *E. coli* from healthy dogs, pigs, and broilers is of similar magnitude (Fig EC I). Most dogs are kept individually or in small groups as household pets and group medication is not applied. This is in contrast to the situation in production animals, were medication of large groups is common practice. In a population of animals, group medication is likely to have a larger impact on occurrence of resistance than treatment of individual animals. The rather high prevalence of resistant *E. coli* in healthy dogs is therefore remarkable and reflects the very high treatment incidence reported in SVARM 2005.

Table EC I. Occurrence of resistance (%) and multiresistance (%) among isolates of indicator *Escherichia coli* from dairy cows and dogs, 2006. Data for calves/yearling cattle, chickens and pigs from previous SVARM reports are given for comparison.

		Resistance (%) (95% confidence interval inside brackets)									
		Cattle									
		Dairy cows		Calves/Yearlings		Dogs		Broilers		Pigs	
Antimicrobial	Cut-off value (mg/L)	2006 n=314		2000 n=293		2006 n=257		2004 n=300		2005 n=390	
Ampicillin	>8	0	(0.0-1.2)	0	(0.0-1.3)	5	(3.0-9.0)	4	(2.1-6.9)	6	(4.2-9.3)
Cefotaxime	>0.25	0	(0.0-1.2)	-		<1	(0.0-2.1)	-		0	(0.0-0.9)
Ceftiofur	>1	0	(0.0-1.2)	0	(0.0-1.3)	<1	(0.0-2.1)	0	(0.0-1.2)	0	(0.0-0.9)
Chloramphenicol	>16	0	(0.0-1.2)	0	(0.0-1.3)	<1	(0.1-2.9)	0	(0.0-1.2)	3	(1.8-5.6)
Ciprofloxacin	>0.06	<1	(0.1-2.3)	<1[b]	(0.0-1.9)	2	(0.6-4.5)	5[b]	(2.8-8.1)	<1[b]	(0.1-1.4)
Florfenicol	>16	0	(0.0-1.2)	0	(0.0-1.3)	0	(0.0-1.4)	0	(0.0-1.2)	0	(0.0-0.9)
Gentamicin	>4	0	(0.0-1.2)	<1	(0.1-2.4)	0	(0.0-1.4)	<1	(0.0-1.8)	0	(0.0-0.9)
Kanamycin	>16	0	(0.0-1.2)	-		1	(0.2-3.4)	-		-	
Nalidixic acid	>16	<1	(0.1-2.3)	<1	(0.1-2.4)	2	(0.6-4.5)	5	(2.8-8.1)	<1	(0.1-1.4)
Streptomycin	>16	2	(0.3-3.3)	5	(2.9-8.3)	7	(4.2-10.8)	6	(3.3-8.8)	14	(10.8-18.0)
Neomycin	>8	-		0	(0.0-1.3)	-		3	(1.6-6.1)	1	(0.3-2.6)
Sulphamethoxazole	>256	2	(0.5-3.7)	1	(0.4-3.5)	7	(3.9-10.4)	9	(6.0-12.8)	11	(7.7-14.0)
Tetracycline	>8	2	(0.5-3.7)	1	(0.4-3.5)	2	(0.9-5.0)	6	(3.6-9.3)	9	(6.1-12.0)
Trimethoprim	>2	<1	(0.0-1.8)	2	(0.6-3.9)	4	(1.9-7.0)	<1	(0.1-2.4)	6	(4.2-9.3)
Multiresistance[a]											
Susceptible to all		97.1		91.8		86.8		83.7		77.4	
Resistant to 1		1.3		6.5		5.4		8.7		9.2	
Resistant to 2		0.3		<1		3.5		3.0		5.4	
Resistant to 3		1.0		1.0		1.2		1.3		3.3	
Resistant to >3		0.3		-		3.1		3.3		4.6	

[a] Enrofloxacin/ciprofloxacin and nalidixic acid regarded as one substance; [b] Enrofloxacin tested, cut-off value >0.12 mg/L.

Table EC II. Number of *Escherichia coli* resistant to three or more antimicrobials, presented by resistance phenotype. "R" in shaded fields indicates resistance.

Animal species		Resistance pattern[a]										
Dairy cows 2006 n=314	Dogs 2006 n=257	Su	Sm	Tc	Am	Tm	Cm	Nal	Ci/Ef	Ctx	Ce	Ka
	2	R	R	R	R	R						
1		R	R	R		R						
3		R	R	R								
	1	R	R	R			R					R
	1	R	R		R	R						
	2	R	R			R						R
	1	R		R	R		R					
	3		R		R	R						
	1			R	R			R	R	R	R	
4 (1.3%)	11 (4.3%)	Number of isolates (percent of all isolates)										

[a] Su: sulphonamides; Sm: streptomycin; Tc: tetracycline; Am: ampicillin; Tm: trimethoprim; Cm: chloramphenicol; Nal: nalidixic acid; Ci: ciprofloxacin; Ef: enrofloxacin; Ctx: cefotaxime; Ce: ceftiofur; Ka: kanamycin.

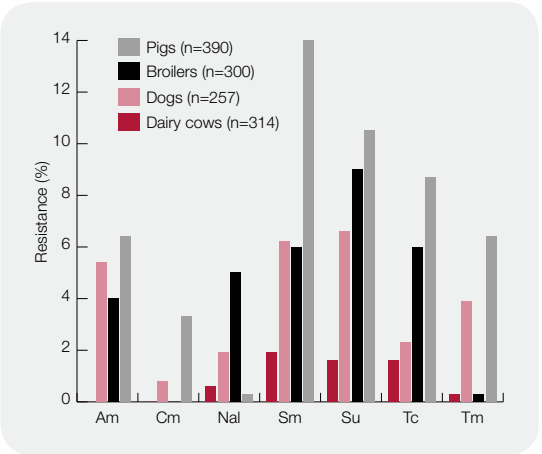

Figure EC I. Resistance (%) among indicator *Escherichia coli* from dairy cows, dogs, broilers and slaughter pigs. Data for broilers and pigs from SVARM 2004 and 2005. (Am: ampicillin; Cm: chloramphenicol; Nal: nalidixic acid; Sm: streptomycin; Su: sulphonamides; Tc: tetracycline; Tm: trimethoprim.

Table EC IV. Distribution of MICs for *Escherichia coli* from dairy cows (n=314) and dogs (n=257). Data for calves/yearling cattle (n=293) from SVARM year 2000 are given for comparison.

Anti-microbial (%)	Animal species	Resis-tance	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	1024	2048	>2048
Ampicillin	Dairy cows	0				0.3	3.8	17.2	51.3	20.7	6.7									
	Calves/ Yearlings	0						1.4	18.8	78.8	1.0									
	Dogs	5				0.4	1.2	21.4	69.6	1.6	0.4			5.4						
Cefotaxime	Dairy cows	0		66.9	31.8	1.3														
	Calves/ Yearlings	NT[b]																		
	Dogs	<1		48.6	49.8	1.2				0.4										
Ceftiofur	Dairy cows	0			6.7	38.9	51.9	2.5												
	Calves/ Yearlings	0				24.9	72.0	3.1												
	Dogs	<1			1.9	27.6	65.0	5.1	0.4											
Chloram-phenicol	Dairy cows	0							12.4	75.2	12.4									
	Calves/ Yearlings	0							1.0	37.9	60.8	0.3								
	Dogs	<1						0.8	1.6	59.1	37.0	0.8	0.4			0.4				
Cipro-floxacin	Dairy cows	<1	73.6	25.8			0.3		0.3											
	Calves/ Yearlings	<1[c]	29.4	69.3	1.0		0.3													
	Dogs	2	33.5	64.6		0.4	1.2		0.4											
Florfenicol	Dairy cows	0								51.6	48.4									
	Calves/ Yearlings	0							0.7	23.5	70.6	5.1								
	Dogs	0								25.3	71.6	3.1								
Gentamicin	Dairy cows	0					21.3	69.1	8.6	1.0										
	Calves/ Yearlings	<1					2.4	37.2	49.8	9.9	0.7									
	Dogs	0					26.1	67.7	5.8	0.4										
Kanamycin	Dairy cows	0							9.7	80.6	8.9	0.8								
	Calves/ Yearlings	NT																		
	Dogs	1							30.0	61.1	6.6	1.2	1.2							
Nalidixic acid	Dairy cows	<1						3.8	19.4	71.3	4.8			0.3		0.3				
	Calves/ Yearlings	<1							0.7	22.5	70.3	5.8		0.3		0.3				
	Dogs	2						0.8	30.0	65.0	2.3					1.9				
Strepto-mycin	Dairy cows	2								15.6	76.1	6.4	0.3	0.6	0.6		0.3			
	Calves/ Yearlings	5							0.7	13.0	67.2	14.0		1.4	1.0	1.7	1.0			
	Dogs	6							0.8	33.5	53.3	5.4	0.8	1.6	1.9	1.2	1.6			
Sulpha-methoxa-zole	Dairy cows	2										79.3	18.5	0.6						1.6
	Calves/ Yearlings	1												42.3	54.6	1.7	0.3	1.0		
	Dogs	7										19.5	32.7	34.6	6.6		0.4			6.3
Tetracy-cline	Dairy cows	2					0.3	65.6	31.8	0.6			0.3	0.3	1.0					
	Calves/ Yearlings	1						8.9	57.7	30.7	1.4			0.3	1.0					
	Dogs	2						10.1	86.8	0.8				0.8	1.6					
Trimetho-prim	Dairy cows	<1				46.2	51.0	2.2	0.3					0.3						
	Calves/ Yearlings	2			3.1	9.2	34.8	42.3	8.9	1.7										
	Dogs	4				7.0	45.5	38.1	5.4				0.4	3.5						
			≤0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	2048	>2048

[a] 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 epidemiological cut-off values defining resistance;
[b] Not tested; [c] Enrofloxacin tested, Cut-off value 0.12 mg/L.

Enterococcus

Dairy cows

Enterococci were isolated from 83% of the 461 samples cultured. *Enterococcus hirae* (38%) was the predominant species followed by *E. faecium* (26%), *E. durans* (14%) and *E. mundtii* (5%) (Table ENT I). Only 3 % of the isolates were *E. faecalis* and 13 % could not be typed to species level with the simplified scheme used.

Resistance was rare among all species of enterococci and most isolates were susceptible to all antimicrobials tested (Table ENT II & III). No isolate was resistant to ampicillin, chloramphenicol, gentamicin, kanamycin, linezolid, or streptomycin. Among the 13 isolates of *E. faecalis* available for testing, only resistance to narasin or tetracycline was observed. Among *E. faecium*, resistance to bacitracin, tetracycline, erythromycin, or virginiamycin occurred but only one isolate was resistant to more than one antimicrobial (tetracycline and erythromycin). Resistance was rare also among *E. hirae*, only erythromycin or narasin resistance occurred and no isolate was resistant to more than one antimicrobial.

Dogs

Enterococci were isolated from 87% of the 299 samples cultured. *E. faecalis* (52%) was the predominant species followed by *E. faecium* (11%), *E. hirae* (9%), *E. mundtii* (5%), *E. durans* (<1%), and *E. flavescens* (<1%) (Table ENT I). Twenty-three percent of the isolates could not be typed to species level with the simplified scheme used.

In all species of enterococci, tetracycline and erythromycin were the predominant resistance traits (Table ENT II & III). Resistance to streptomycin, chloramphenicol, kanamycin, or narasin was less common and ampicillin, bacitracin or gentamicin resistance occurred in a few isolates only. No isolate was resistant to linezolid or vancomycin.

Among the predominant species, *E. faecalis*, about one third of the isolates (32%) were resistant to tetracycline and 14% to erythromycin. Ten isolates (7% of all *E. faecalis*) were resistant to at least both of these antimicrobials.

Resistance to more than two antimicrobials occurred only among *E. faecalis* where eight isolates (6%) were resistant to at least three substances (Table EC IV). Erythromycin was included in the phenotype of all these isolates.

Table ENT I. Prevalence of enterococci in samples of caecal content and faecal swabs from dairy cows and dogs, respectively, 2006. Species not identified as *Enterococcus faecalis, E. faecium* or *E. hirae* are given as "other species". Previous data from SVARM are given for comparison.

Animal species	Year	No. of samples cultured	Positive cultures	Isolates tested for susceptibility	*Enterococcus* species isolated No. of isolates (percent of total No. of isolates inside brackets)			
					E. faecalis	*E. faecium*	*E. hirae*	Other species
Dairy cows	2006	461	83%	383	13 (3%)	98 (26%)	147 (38%)	125 (33%)
Calves/Yearlings	2000	415	67%	277	22 (8%)	71 (26%)	127 46%)	57 (21%)
Dogs	2006	299	87%	259	135 (52%)	29 (11%)	22 (8%)	73 (28%)

Table ENT II. Occurrence of resistance (%) among isolates of *Enterococcus* spp. from dairy cows and dogs, 2006. Previous data from SVARM are given for comparison.

		Resistance (%) 95% confidence interval inside brackets									
Antimicrobial	Cut-off value (mg/L)	Dairy cows 2006 n=383		Calves/Yearlings 2000 n=277		Dogs 2006 n=259		Pigs 2005 n=262		Broilers 2004 n=306	
Ampicillin	>4	0	(0.0-1.0)	1	(0.4-3.7)	1	(0.2-3.3)	1	(0.2-3.3)	1	(0.4-3.3)
Bacitracin[a]	>32	<1	(0.0-1.4)	<1	(0.1-2.6)	1	(0.2-3.3)	<1	(0.1-2.7)	25	(20.4-30.4)
Chloramphenicol	>32	0	(0.0-1.0)	-		3	(1.6-6.4)	2	(0.4-3.9)	0	(0.0-1.2)
Erythromycin	>4	3	(1.4-5.1)	3	(1.0-5.1)	17	(12.6-22.1)	13	(8.8-17.2)	18	(13.5-22.4)
Gentamicin	>32	0	(0.0-1.0)	0	(0.0-1.3)	<1	(0.0-2.1)	-		0	(0.0-1.2)
Kanamycin	>1024	0	(0.0-1.0)	-		3	(1.1-5.5)	-		-	
Linezolid	>4	0	(0.0-1.0)	-		0	(0.0-1.4)	-		-	
Narasin	>2	1	(0.3-2.7)	1	(0.4-3.7)	2	(0.9-5.0)	3	(1.3-5.9)	81	(75.9-85.0)
Streptomycin	>512/>128[b]	0	(0.0-1.0)	2[d]	(0.8-4.7)	7	(3.9-10.3)	-		1	(0.2-2.8)
Tetracycline	>2	4	(2.2-6.4)	8	(5.3-12.2)	25	(19.9-30.8)	27	(21.5-32.5)	22	(17.4-27.0)
Vancomycin	>4	<1[c]	(0.2-2.3)	1[c]	(0.2-3.1)	0	(0.0-1.4)	0	(0.0-1.4)	1	(0.2-2.8)

[a] MIC in U/mL; [b] Cut-off for E. faecalis >512 mg/L, for other species >128 mg/L; [c] Isolates with MIC 8 mg/L; [d] Cut-off >128 mg/L for all species;

Table ENT III. Occurrence of resistance (%) and multiresistance (%) among *Enterococcus faecalis*, *E. faecium* and *E. hirae* from dairy cows and dogs, presented by bacterial species and source of isolates, 2006. Previous data from SVARM are given for comparison. Cut-off values defining resistance are given in Table ENT II.

	E. faecalis					*E. faecium*					*E. hirae*				
Antimicrobial	Dairy cows 2006 n=13	Calves/ Year-lings 2000 n=22	Dogs 2006 n=135	Pigs 2005 n=55	Broilers 2004 n=48	Dairy cows 2006 n=98	Calves/ Year-lings 2000 n=71	Dogs 2006 n=29	Pigs 2005 n=47	Broilers 2004 n=163	Dairy cows 2006 n=147	Calves/ Year-lings 2000 n=127	Dogs 2006 n=22	Pigs 2005 n=112	Broilers 2004 n=34
Ampicillin	0	0	<1	0	0	0	1	0	0	2	0	2	0	0	0
Bacitracin	0	0	1	2	29	1	1	3	2	32	0	0	0	0	0
Chloramphenicol	0	-	7	5	0	0	-	0	2	0	0	-	0	0	0
Erythromycin	0	5	14	33	25	7	6	28	21	10	<1	0	14	0	26
Gentamicin	0	0	<1	5[b]	0	0	0	0	2[b]	0	0	0	0	0[b]	0
Kanamycin	0	-	4	-	-	0	-	0	-	-	0	-	5	-	-
Linezolid	0	-	0	-	-	0	-	0	-	-	0	-	0	-	-
Narasin	8	0	1	0	35	0	1	7	0	93	2	2	5	5	91
Streptomycin	0	14[a]	9	16	4	0	0	0	0[d]	<1	0	0	5	0[d]	0
Tetracycline	15	14	32	64	48	3	7	17	13	18	0	3	14	11	3
Vancomycin	0	0	0	0	0	0	1[c]	0	0	2	0	0	0	0	0
Virginiamycin	0	0	0	0	0	1	7	0	13	9	0	15	5	2	3
Multiresistance															
Susceptible to all	77	77	59	26	23	89	82	55	62	3	97	80	77	82	3
Resistant to 1	23	18	24	38	29	10	13	34	30	41	3	18	18	18	50
Resistant to 2		5	11	27	35	1	4	10	6	44		2	5		44
Resistant to 3			2	4	10		1			10					3
Resistant to >3			4	6	2				2	2					

[a] Cut-off value >128 mg/L; [b] Cut-off value >512 mg/L; [c] Isolates with MIC 8 mg/L; [d] Cut-off value >256 mg/L.

Table ENT IV. Number of *Enterococcus faecalis* resistant to three or more antimicrobials, presented by resistance phenotype,dogs 2006. "R" in shaded fields indicates resistance.

	E. faecalis						
	Resistance pattern[a]						
2006 n=135	Tc	Em	Sm	Km	Cm	Gm	Na
1	R	R	R	R	R	R	
1	R	R	R	R	R		
2	R	R	R	R			
1	R	R	R				
1	R	R				R	
1	R	R					R
1		R	R	R		R	
8 (6%)	Total number of multiresistant isolates						

[a] Tc: tetracycline; Em: erythromycin; Sm: streptomycin; Km: kanamycin; Cm: chloramphenicol; Gm: gentamicin; Na: narasin.

Broilers

VRE were isolated from 29 (28%) of 102 samples cultured on vancomycin-supplemented media (16 mg/L). For all 29 isolates, MIC for vancomycin was >128 mg/L. In addition, they were resistant to narasin (MIC 4–16 mg/L) and 27 isolates to erythromycin (MIC 8–32 mg/L). Ten isolates examined by PCR all carried the *van*A-gene.

Comments

In agreement with the results for *E. coli*, resistance is rare in enterococci from dairy cows. The results tally with the survey in calves/yearling cattle in 2000 and show that selection towards resistance in the commensal flora of adult cattle is small. This is probably because adult cattle mainly are treated with penicillin as discussed above. Although treatment (for mastitis) is common, the impact on resistance in the commensal enteric flora is low in agreement with the narrow antibacterial spectrum of penicillin. Occurrence of resistance in other species of commensal bacteria in cattle deserves further study, however.

In dogs, resistance in enterococci was more common than in isolates from cattle and of similar magnitude as in enterococci from pigs and poultry. The situation is similar to that in *E. coli* (see above) and signifies a substantial selection pressure by use of antimicrobials. Resistance to tetracycline and erythromycin were the most common traits in agreement with a high use of these antimicrobials (see Use of antimicrobials). In contrast, despite the common use of aminopenicillins in dogs (see Use of antimicrobials), ampicillin resistance was not observed among *E. faecium* or *E. hirae*. This indicates that ampicillin resistant enterococcal clones are rare among healthy Swedish dogs.

The prevalence of VRE among broilers (28%), studied by use of selective media, is numerically lower than in 2005 (41%) and thus the increase in prevalence observed in the last years (SVARM 2005) seems to have abated. In previous years, phenotyping of VRE by the PhenePlate™ system has indicated clonality of isolates from Swedish broilers (SVARM 2005). This year, isolated VRE were not typed that way, but the resistance phenotype of the isolates, including vancomycin, narasin, and erythromycin, is the same as in previous years, which suggest that the dominant clone still prevails.

Table ENT V. Distribution of MICs for *Enterococcus faecalis* from dairy cows (n=13) and dogs (n=135) year 2006. Data for calves/yearling cattle (n=22) from SVARM year 2000 are given for comparison.

Antimicrobial	Animal species	Year	Resistance (%)	Distribution (%) of MICs[a] (mg/L)															
				≤0.12	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**1024**	**2048**	**> 2048**
Ampicillin	Dairy cows	-06	0			23.1	61.5	15.4											
	Calves/ Yearlings	-00	0		13.6	18.2	59.1		9.1										
	Dogs	-06	<1			8.9	85.2	4.4	0.7	0.7									
Bacitracin[b]	Dairy cows	-06	0					7.7		38.5	46.2	7.7							
	Calves/ Yearlings	-00	0			9.1	9.1	9.1	27.3	27.3	4.5	13.6							
	Dogs	-06	1				2.2	3.7	13.3	60.7	16.3	2.2		0.7	0.7				
Chloramphenicol	Dairy cows	-06	0						30.8	69.2									
	Calves/ Yearlings	NT[c]	-																
	Dogs	-06	7				1.5	2.2	23.0	66.7			5.2	1.5					
Erythromycin	Dairy cows	-06	0			30.8	23.1	23.1	23.1										
	Calves/ Yearlings	-00	5		13.6	18.2	9.1	31.8	22.7				4.5						
	Dogs	-06	14			17.8	21.5	21.5	25.2	3.0	3.7	1.5	0.7	5.2					
Gentamicin	Dairy cows	-06	0						15.4	23.1	61.5								
	Calves/ Yearlings	-00	0				4.5	4.5	9.1	40.9	40.9								
	Dogs	-06	<1					4.4	3.7	31.9	57.0	2.2				0.7			
Kanamycin	Dairy cows	-06	0									7.7	84.6		7.7				
	Calves/ Yearlings	NT	-																
	Dogs	-06	4								5.2	37.0	50.4	2.2	0.7			0.7	3.7
Linezolid	Dairy cows	-06	0					84.6	15.4										
	Calves/ Yearlings	NT	-																
	Dogs	-06	0			0.7	3.7	93.3	2.2										
Narasin	Dairy cows	-06	8		7.7	38.5	30.8	15.4		7.7									
	Calves/ Yearlings	-00	0	9.1	31.8	40.9	13.6	4.5											
	Dogs	-06	1	0.7	17.8	57.0	17.0	5.9	1.5										
Streptomycin	Dairy cows	-06	0									15.4	76.9	7.7					
	Calves/ Yearlings	-00	14								4.5	13.6	40.9	27.3			9.1	4.5	
	Dogs	-06	9							3.0		3.7	37.0	47.4				8.9	
Tetracycline	Dairy cows	-06	15			76.9	7.7					7.7	7.7						
	Calves/ Yearlings	-00	14			13.6	27.3	45.5			4.5	9.1							
	Dogs	-06	32			51.1	14.8	2.2			1.5	7.4	22.2	0.7					
Vancomycin	Dairy cows	-06	0				38.5	30.8	30.8										
	Calves/ Yearlings	-00	0				27.3	63.6	9.1										
	Dogs	-06	0				17.0	62.2	20.7										
Virginiamycin	Dairy cows	-06	0					15.4	23.1		53.8	7.7							
	Calves/ Yearlings	-00	0			9.1	9.1	18.2	13.6	22.7	22.7	4.5							
	Dogs	-06	0			1.5	0.7	3.0	3.7	19.3	52.6	19.3							
				≤0.12	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**1024**	**2048**	**>2048**

[a] 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 cut-off values defining resistance; [b] MIC in U/mL, see Appendix 3 for details; [c] Not tested.

Table ENT VI. Distribution of MICs for *Enterococcus faecium* from dairy cows (n=98) and dogs (n=29) year 2006. Data for calves/yearling cattle (n=71) from SVARM year 2000 are given for comparison.

Antimicrobial	Animal species	Year	Resistance (%)	Distribution (%) of MICs[a] (mg/L)															
				≤0.12	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**1024**	**2048**	**>2048**
Ampicillin	Dairy cows	-06	0		10.2	30.6	45.9	13.3											
	Calves/ Yearlings	-00	1		4.2	1.4	12.7	63.4	16.9	1.4									
	Dogs	-06	0		13.8	13.8	48.3	20.7	3.4										
Bacitracin[b]	Dairy cows	-06	1				1.0	1.0	3.1	16.3	66.3	11.2	1.0						
	Calves/ Yearlings	-00	1			2.8	25.4	21.1	5.6	12.7	12.7	18.3	1.4						
	Dogs	-06	3				3.4	10.3	6.9	6.9	37.9	31.0			3.4				
Chloramph.	Dairy cows	-06	0					1.0	24.5	72.4	2.0								
	Calves/ Yearlings	-00	NT[c]																
	Dogs	-06	0				3.4	3.4	31.0	62.1									
Erythromycin	Dairy cows	-06	7			29.6	24.5	13.3	25.5	7.1									
	Calves/ Yearlings	-00	6		5.6	53.5	14.1	9.9	11.3		2.8		2.8						
	Dogs	-06	28			20.7	3.4	27.6	20.7	10.3	10.3	3.4		3.4					
Gentamicin	Dairy cows	-06	0					3.1	33.7	57.1	6.1								
	Calves/ Yearlings	-00	0					4.2	5.6	56.3	31.0	2.8							
	Dogs	-06	0					13.8	20.7	55.2	10.3								
Kanamycin	Dairy cows	-06	0								4.1	36.7	28.6	20.4	7.1	3.1			
	Calves/ Yearlings	-00	NT																
	Dogs	-06	0								3.4	17.2	37.9	31.0	6.9	3.4			
Linezolid	Dairy cows	-06	0				2.0	78.6	19.4										
	Calves/ Yearlings	-00	NT																
	Dogs	-06	0				13.8	44.8	41.4										
Narasin	Dairy cows	-06	0		1.0	36.7	55.1	7.1											
	Calves/ Yearlings	-00	1	5.6	26.8	23.9	39.4	2.8	1.4										
	Dogs	-06	7		10.3	10.3	55.2	17.2	3.4		3.4								
Streptomycin	Dairy cows	-06	0								3.1	34.7	62.2						
	Calves/ Yearlings	-00	0							1.4	2.8	52.1	38.0	5.6					
	Dogs	-06	0							3.4	6.9	27.6	55.2	6.9					
Tetracycline	Dairy cows	-06	3			62.2	34.7					3.1							
	Calves/ Yearlings	-00	7		1.4	5.6	62.0	23.9	1.4			2.8	2.8						
	Dogs	-06	17			79.3	3.4					6.9	10.3						
Vancomycin	Dairy cows	-06	0				75.5	10.2	14.3										
	Calves/ Yearlings	-00	1				77.5	16.9	4.2	1.4									
	Dogs	-06	0				86.2	6.9	6.9										
Virginiamycin	Dairy cows	-06	1			25.5	16.3	18.4	38.8	1.0									
	Calves/ Yearlings	-00	7			21.1	21.1	33.8	16.9	5.6			1.4						
	Dogs	-06	0			20.7	27.6	6.9	44.8										
				≤0.12	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**1024**	**2048**	**>2048**

[a] 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] MIC in U/mL, see Appendix 3 for details. [c] Not tested.

Table ENT VII. Distribution of MICs for *Enterococcus hirae* from dairy cows (n=147) and dogs (n=22) year 2006. Data for calves/yearling cattle (n=127) from SVARM year 2000 are given for comparison.

Antimicrobial	Animal species	Year	Resistance (%)	Distribution (%) of MICs[a] (mg/L)															
				≤0.12	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**1024**	**2048**	**>2048**
Ampicillin	Dairy cows	-06	0		10.9	11.6	30.6	46.3	0.7										
	Calves/ Yearlings	-00	2		11.8	3.9	15.0	46.5	21.3	1.6									
	Dogs	-06	0		13.6	31.8	36.4	18.2											
Bacitracin[b]	Dairy cows	-06	0				30.6	57.1	4.1	3.4	2.7	2.0							
	Calves/ Yearlings	-00	0				27.6	41.7	24.4	2.4	2.4	1.6							
	Dogs	-06	0				13.6	40.9	27.3	9.1	9.1								
Chloramph.	Dairy cows	-06	0				0.7	3.4	78.2	17.7									
	Calves/ Yearlings	-00	NT[c]																
	Dogs	-06	0					9.1	72.7	18.2									
Erythromycin	Dairy cows	-06	<1			93.2	3.4	2.0	0.7	0.7									
	Calves/ Yearlings	-00	0		36.2	60.6	0.8	2.4											
	Dogs	-06	14			72.7	4.5		9.1	4.5	9.1								
Gentamicin	Dairy cows	-06	0					3.4	6.8	50.3	35.4	4.1							
	Calves/ Yearlings	-00	0					4.7	8.7	52.8	27.6	6.3							
	Dogs	-06	0					27.3	18.2	40.9	13.6								
Kanamycin	Dairy cows	-06	0								4.1	32.7	55.8	6.1	1.4				
	Calves/ Yearlings	-00	NT																
	Dogs	-06	5								4.5	18.2	36.4	27.3	9.1				4.5
Linezolid	Dairy cows	-06	0			0.7	5.4	90.5	3.4										
	Calves/ Yearlings	-00	NT																
	Dogs	-06	0				9.1	81.8	9.1										
Narasin	Dairy cows	-06	2		8.2	59.2	27.2	3.4	0.7	1.4									
	Calves/ Yearlings	-00	2		15.0	36.2	44.1	3.1	1.6										
	Dogs	-06	5		18.2	9.1	27.3	40.9	4.5										
Streptomycin	Dairy cows	-06	0							0.7		13.6	76.9	8.8					
	Calves/ Yearlings	-00	0								6.3	22.8	60.6	10.2					
	Dogs	-06	5							9.1	31.8	4.5	45.5	4.5				4.5	
Tetracycline	Dairy cows	-06	0			85.7	14.3												
	Calves/ Yearlings	-00	3			8.7	43.3	44.9	2.4				0.8						
	Dogs	-06	14			81.8	4.5				4.5	4.5	4.5						
Vancomycin	Dairy cows	-06	0				86.4	13.6											
	Calves/ Yearlings	-00	0				88.2	11.8											
	Dogs	-06	0				95.5		4.5										
Virginiamycin	Dairy cows	-06	0			10.2	7.5	35.4	46.9										
	Calves/ Yearlings	-00	15			20.5	9.4	47.2	7.9	15.0									
	Dogs	-06	5			4.5	31.8	36.4	22.7		4.5								
				≤0.12	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**64**	**128**	**256**	**512**	**1024**	**2048**	**>2048**

[a] 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] MIC in U/mL, see Appendix 3 for details. [c] Not tested.

Resistance in animal pathogens

ISOLATES TESTED are from clinical submission of samples to SVA if not stated otherwise. For these samples, information on the indications for sampling is not available but the vast majority of clinical submissions are likely from diseased animals. Therefore, data are probably biased towards samples from treated animals or from herds where antimicrobial treatments are common. Any assessment of trends is based on the assumption that this bias is inherent throughout the observation period.

In SVARM 2006, some cut-off values defining resistance used in SVARM 2000–2005 have been changed. To facilitate comparisons, data on prevalence of resistance from these reports were recalculated using current cut-off values if not otherwise stated (see Appendix 3 for an overview of cut-off values).

Pig

Isolates included

Isolates of *Escherichia coli* from years 1992–2006 are from diagnostic submissions of samples from the gastro-intestinal tract (intestinal content, faecal samples or mesenteric lymph nodes), while data from 1989–1991 include all *E. coli* isolated from pigs, irrespective of material type. Isolates of *Brachyspira hyodysenteriae* are from clinical submissions of faecal samples from pigs.

Escherichia coli

As in previous years, resistance to ampicillin, streptomycin, tetracycline or trimethoprim-sulphonamides was common in 2006 (Table Pig I). Statistical analyses indicate that over the last six years (2001–2006) prevalence of resistance to ampicillin has increased (P=0.03, Chi-square for trend), and resistance to tetracycline decreased (P=0.004, Chi-square for trend). Over the last three years, however, prevalence of resistance has been stable for both antimicrobials.

In the 70s and 80s, six and seven percent, respectively, of *E. coli* from diarrhoeic pigs were resistant to ampicillin (Franklin, 1976; Franklin, 1984). In the early 90s, prevalence of ampicillin resistance increased gradually to 22% in 2004 and still remains at this level (Figure Pig I). However, 93% of isolates resistant to ampicillin was resistant also to at least one other antimicrobial. Use of aminopenicillins will therefore probably co-select for resistance to other substances. The most common combination of traits was resistance to ampicillin and trimethoprim-sulphonamides (80% of the ampicillin resistant isolates), and 78% of isolates resistant to trimethoprim-sulphonamides were resistant to ampicillin. This indicates that the genes coding for resistance to ampicillin and trimethoprim-sulphonamides are linked. For trimethoprim-sulphonamides, the prevalence of resistance has increased from 10% in the beginning of the 80s (break-point for resistance >8mg/L; Franklin 1984) to a peak figure of 27% in 2004. The combination trimethoprim-sulphonamides has been used since 1974, amongst others, in treatment of diarrhoea in piglets.

Multiresistance (i.e. resistance to three or more antimicrobials) occurred in 17% of the isolates and of those, 17% were resistant to five or more antimicrobials (3% of the whole material). In multiresistant isolates, the most frequent combination, found in 56% of the multiresistant isolates, was resistance to ampicillin, trimethoprim-sulphonamides, and streptomycin, and 48% of these were resistant also to tetracycline.

The extent of use of aminopenicillins or trimethoprim-sulphonamides for Swedish pigs is not known, as data on use of antimicrobials are not yet fully available per animal species. However, the extent of use of tetracycline to pigs is know and has increased by 26% since year 2000 (see Use of antimicrobials), but is not reflected in the prevalence of tetracycline resistance.

Table Pig I. Occurrence of resistance among *Escherichia coli* from pigs 1998-2006 and distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of faecal samples or samples taken post mortem from the gastro-intestinal tract.

	Resistance (%)								Distribution (%) of MICs[a] (mg/L)									
Antimicrobial	1989-1991 n=248	1992-1994 n=431	1995-1997 n=1244	1998-2000 n=1074	2001-2003 n=935	2004 n=386	2005 n=325	2006 =298	≤0.12	0.25	0.5	1	2	4	8	16	32	>32
Ampicillin	6	10	9	11	17	22	22	21				10.7	54.4	14.4		20.5		
Ceftiofur	-	-	-	-	0[e]	<1	<1	<1		37.2	57.7	4.4		0.7				
Enrofloxacin	1[c,e]	7[c]	5[c]	6[c]	8[c]	9	9	8	91.6	2.3	3.0	0.3	2.7					
Florfenicol	-	-	-	-	<1[f]	0	0	<1					1.3	35.7	57.4	5.4	0.3	
Gentamicin	1[d]	1[d]	<1[d]	1[d]	4[d]	2	<1	1					91.6	7.4	0.7		0.3	
Neomycin	17	14	9	6	5[g]	4	3	3						93.0	3.7	0.7	0.3	2.3
Streptomycin	44	44	32	30	30	28	30	25						9.7	37.6	20.8	6.7	25.2
Tetracycline	28	35	31	33	30	27	24	26				25.5	36.9	10.4	1.3	25.8		
Trim/Sulph.[b]	17	15	13	14	19	27	24	21			77.2	1.7			21.1			

[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 cut-off values defining resistance; [b] Concentration of trimethoprim given, tested in concentration ratio 1/20 (trimethoprim/sulphametoxazole); [c] Cut-off value was >0.25 mg/L until year 2001; [d] Cut-off value was >8 mg/L until year 2002; [e] 227 isolates tested; [f] 688 isolates tested; [g] 926 isolates tested.

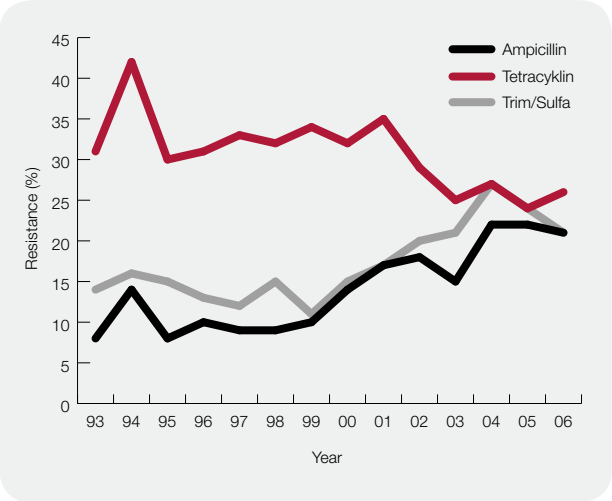

Figure Pig I. Occurrence of resistance to selected antimicrobials among *Escherichia coli* from pigs 1992-2006. Isolates are from diagnostic submissions of faecal samples or samples taken post mortem from the gastro-intestinal tract. Around 300 isolates were tested each year.

Brachyspira hyodysenteriae

All isolates of *B. hyodysenteriae* were susceptible to tiamulin (Table Pig II). Sweden has a programme for controlling swine dysentery by three strategies; nucleus and multiplying herds are tested for *B. hyodysenteriae* twice a year, eradication of the bacteria in infected herds and tracing the source of infection. Nevertheless, it is imperative that all herds where treatment failure is suspected are thoroughly investigated.

Resistance to tylosin has increased dramatically over the last decade. In 1988–90 only 20% of *B. hyodysenteriae* were classified as resistant to tylosin when tested with an agar dilution technique (Gunnarsson *et al.*, 1991), but from 2001 the prevalence of resistance has been around 80% (Table Pig II).

Brachyspira pilosicoli

In 2001, the first isolates of *B. pilosicoli* resistant to tiamulin were confirmed in Sweden. These isolates were associated with treatment failure in a Swedish pig herd with spirochaetal diarrhoea (See SVARM 2003). Since then, tiamulin resistant strains have been isolated every year but there is no apparent increasing trend in prevalence of resistance (Table Pig III). The frequency of resistance to tylosin seems to rise, however, and this year about two thirds of the isolates are resistant to this antimicrobial (Table Pig III).

Resistance to both antimicrobials occurred in 11% of all isolates and 17% of *B. pilosicoli* resistant to tylosin was resistant also to tiamulin. Although such isolates may be susceptible to other antimicrobials, only tiamulin and tylosin are currently licensed for treatment of spirochaetal diarrhoea in pigs in Sweden. The findings stress the need for susceptibility testing of *B. pilosicoli* from herds, where tiamulin is to be used.

Cattle

Isolates included

Escherichia coli are from the gastro-intestinal tract of cattle. *Klebsiella* spp. were isolated from diagnostic submissions of milk samples from dairy cows. Each strain of *Klebsiella* spp. is from a unique herd.

Escherichia coli

Resistance to ampicillin, enrofloxacin, neomycin, streptomycin, tetracycline or trimethoprim-sulphonamides was common in 2006 as in previous years (Table Cattle I). The proportions of *E. coli* resistant to ampicillin, enrofloxacin, neomycin, tetracycline, or trimetoprim-sulphonamides were numerically higher than in 2005. The prevalence of multiresistance (26%) was also higher than in 2005 (13%). However, the small number of isolates tested precludes conclusions on trends.

Table Pig II. Occurrence of resistance among *Brachyspira hyodysenteriae* from pigs 2001- 2006 and distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of faecal samples.

	Resistance (%)					Distribution (%) of MICs[a] (mg/L)													
Antimicrobial	2001 n=75	2002 n=109	2003 n=100	2005 n=31	2006 n=26	≤0.016	0.031	0.063	0.125	0.25	0.5	1	2	4	8	16	32	64	128	>128
Tiamulin	0	0	0	0	0			23.1	69.2		7.7									
Tylosin	83	73	89	81	85								3.8		11.5					84.6

[a] 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.

Table Pig III. Occurrence of resistance among *Brachyspira pilosicoli* from pigs 2002-2006 and distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of faecal samples.

	Resistance (%)			Distribution (%) of MICs[a] (mg/L)														
Antimicrobial	2002-03 n=93	2005 n=57	2006 n=72	≤0.016	0.031	0.063	0.125	0.25	0.5	1	2	4	8	16	32	64	128	>128
Tiamulin	14	16	12			19.4	27.8	16.7	12.5	11.1			2.8	9.7				
Tylosin	50[b]	63	67								5.6	15.3	11.1	1.4	4.2	2.8	2.8	56.9

[a] 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; [b] 86 isolates tested.

Table Cattle I. Occurrence of resistance among *Escherichia coli* from cattle 1992-2002, 2005, 2006 and from calves 2004. Distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of faecal samples or samples taken post mortem from the gastro-intestinal tract.

	Resistance (%)				Distribution (%) of MICs[a] (mg/L)									
Antimicrobial	1992-02 n=220	2004 n=87[f]	2005 n=39	2006 n=24	≤0.12	0.25	0.5	1	2	4	8	16	32	>32
Ampicillin	24	29	31	33				12.5	33.3	20.8		33.3		
Ceftiofur	0[c]	0	0[g]	0		20.8	58.3	12.5	8.3					
Enrofloxacin	10[d]	14	10	17	83.3	8.3		4.2	4.2					
Florfenicol	0[c]	0	0	0					4.2	20.8	70.8	4.2		
Gentamicin	5[e]	0	0	0					87.5	12.5				
Neomycin	8	7	10	17						75.0	8.3			16.7
Streptomycin	42	48	54	54						4.2	33.3	8.3		54.2
Tetracycline	31	37	46	54				8.3	25.0	12.5		54.2		
Trim/Sulph.[b]	11	10	18	25			66.7	8.3			25.0			

[a] 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 cut-off values defining resistance; [b] Concentration of trimethoprim given, tested in concentration ratio 1/20 (trimethoprim/sulphametoxazole); [c] 16 isolates tested; [d] Cut-off value >0.25 mg/L; [e] Cut-off value >8 mg/L until year 2001; [f] 1/3 of the isolates were from calves with diarrhoea; [g] 38 isolates tested.

Table Cattle II. Occurrence of resistance among *Klebsiella* spp. from dairy cows 2002-03 and 2006 and distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of milk samples.

	Resistance (%)		Distribution (%) of MICs[a] (mg/L)														
Antimicrobial	2002-03 n=33	2006 n=24	≤0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	256	512	>512
Ampicillin	NR[b]	NR									16.7	29.2	12.5	41.7				
Cefotaxime	-	0		100														
Ceftiofur	0	0			12.5	45.8	37.5	4.2										
Chloramphenicol	0	0						4.2	41.7	0.5	4.2							
Enrofloxacin	0	4	8.3	79.2	4.2	4.2		4.2										
Florfenicol	0	0								75.0	25.0							
Gentamicin	0	0					83.3	16.7										
Nalidixic acid	0	4							8.3	83.3	4.2					4.2		
Neomycin	0	0							100									
Streptomycin	14	13							4.2	75.0	8.3			8.3		4.2		
Sulphamethoxazole	9	4										16.7	41.7	37.5				4.2
Tetracycline	7	0					16.7	45.8	37.5									
Trimethoprim	2	0				4.2	50.0	37.5	4.2	4.2								

[a] 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 cut-off values defining resistance; [b] Not relevant as the genus is inherently resistant to ampicillin.

Klebsiella

During 2006, 24 *Klebsiella* spp. (14 *Klebsiella pneumoniae* and 10 *Klebsiella oxytoca*) isolated from bovine milk samples were tested for antimicrobial susceptibility. Twenty-one *Klebsiella* spp. were isolated from sub-clinical mastitis and only three from clinical mastitis.

Apart from ampicillin, to which *Klebsiella* spp. is inherently resistant, the isolates were susceptible to most antimicrobials, including the two cephalosporins tested (Table Cattle II). Thus, none of the isolates were extended spectrum beta-lactamase (ESBL) producers. The results are in full agreement with a previous study on *Klebsiella* spp. from cases of acute clinical mastitis in dairy cows (SVARM 2003).

Of the tested antimicrobials, only enrofloxacin, tetracycline, and the combination trimethoprim-sulphonamides are approved for treatment of mastitis in cattle in Sweden. The result of treatment of mastitis caused by *Klebsiella* spp. is generally considered poor, even though resistance is rare. Factors related to the host, the bacterium or to the pharmacodynamics of these antimicrobials may explain the poor treatment results.

Horse

Isolates included

Escherichia coli are from the genital tract of mares, while isolates of *Streptococcus zooepidemicus* are from the respiratory tract.

Escherichia coli

Resistance to trimethoprim-sulphonamides or streptomycin were the most common resistance traits (Table Horse I). Trimethoprim-sulphonamides resistance is probably a consequence of the frequent use of this antimicrobial combination in horses (see Use of antimicrobials). Moreover, this usage probably co-selects for streptomycin resistance, since 10% of all isolates were resistant to both streptomycin and trimethoprim-sulphonamides. Trimethoprim-sulphonamides were introduced on the Swedish market as an oral formulation for horses in the late 80s. In the period 1992 to 1994, only 2% of *E. coli* were resistant to this combination, but from the mid-90s, the resistance level rose to the current level of about 15%. In 2006, the prevalence of resistance to trimethoprim-sulphonamides or streptomycin was numerically lower than in the previous six years, but there are no significant trends in the period (data not shown, P>0.05, Chi-square for trend).

The prevalence of ampicillin resistance was similar to the levels the last six years (Table Horse I). Prevalence of gentamicin resistance was higher than last year but still low (4%) despite the use of gentamicin in extenders for semen and in solutions for uterine douching in equine stud practice.

Multiresistance (i.e. resistance to three or more antimicrobials) occurred in 8 % of the isolates. Most of the multiresistant isolates were resistant to four or more antimicrobials. Resistance to ampi-

Table Horse I. Occurrence of resistance among *Escherichia coli* from horses 1992-2006 and distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of samples from the female genital tract.

	Resistance (%)							Distribution (%) of MICs[a] (mg/L)									
Antimicrobial	1992-1994 n=48	1995-1997 n=216	1998-2000 n=222	2001-2003 n=457	2004 n=188	2005 n=161	2006 n=124	≤0.12	0.25	0.5	1	2	4	8	16	32	>32
Ampicillin	15	17	10	9	10	4	7				0.8	53.2	38.7		7.3		
Ceftiofur	-	-	-	0[e]	1	0	0		11.3	77.4	11.3						
Enrofloxacin	8[c]	3[c]	3[c]	2[c,f]	3	4	5	95.2		0.8	2.4	1.6					
Florfenicol	-	-	-	0[e]	0	0	0					0.8	35.5	62.1	1.6		
Gentamicin	0[d]	3[d]	6[d]	6	2	2	4					93.5	2.4	0.8		3.2	
Neomycin	4	5	5	3[g]	5	2	5						95.2		0.8		4.0
Streptomycin	31	24	21	19	21	19	14						5.6	53.2	25.0	2.4	13.7
Tetracycline	6	5	9	6	10	6	6				30.6	58.9	4.0		6.5		
Trim/Sulph.[b]	2	15	17	17	20	16	13			85.5	0.8	0.8		12.9			

[a] 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 cut-off values defining resistance; [b] Concentration of trimethoprim given, tested in concentration ratio 1/20 (trimethoprim/sulphametoxazole); [c] Cut-off value >0.25 mg/L until year 2002; [d] Cut-off value >8 mg/L; [e] 353 isolates tested; [f] 456 isolates tested; [g] 455 isolates tested.

Table Horse II. Occurrence of resistance among *Streptococcus zooepidemicus* from horses 1992-2006 and distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of samples from the respiratory tract.

	Resistance (%)							Distribution (%) of MICs[a] (mg/L)									
Antimicrobial	1992-1994 n=218	1995-1997 n=402	1998-2000 n=409	2001-2003 n=505	2004 n=185	2005 n=175	2006 n=174	≤0.12	0.25	0.5	1	2	4	8	16	32	>32
Ampicillin	0	<1	0	0	0	0	0				100						
Enrofloxacin	-	-	-	NR	NR	NR[e]	NR		0.6	3.4	96.0						
Florfenicol	-	-	-	1[d]	2	0	0					92.0	6.3	1.7			
Gentamicin	NR[c]	NR	NR	NR	NR	NR	NR					2.3	1.1	16.1	63.2	17.2	
Neomycin	NR	NR	NR	NR	NR	NR	NR						2.3	0.6	2.9	26.4	67.8
Penicillin	0	<1	0	0	0	0[e]	0	98.9	0.6	0.6							
Spiramycin	<1	1	0	1	1	0[f]	1						97.7	2.7			0.6
Streptomycin	NR	NR	NR	NR	NR	NR	NR						1.1	0.6	4.0	61.5	32.8
Tetracycline	4	3	4	5	3	3	2				63.2	30.5	3.4	0.6	2.3		
Trim/Sulph.[b]	1	11	57	36	49	41	36			55.2	4.0	3.4	2.7	35.6			

[a] 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 cut-off values defining resistance; [b] Concentration of trimethoprim given, tested in concentration ratio 1/20 (trimethoprim/sulphametoxazole); [c] NR= Not relevant as the inherent susceptibility is such that the MIC range is above concentrations that can be obtained during therapy; [d] 370 isolates tested; [e] 174 isolates tested; [f] 172 isolates tested.

cillin, tetracycline, streptomycin and trimetoprim-sulphonamides was found in 60% of the multiresistant isolates. Gentamicin resistance occurred only in isolates resistant to three or more antimicrobials, and it is possible that gynaecological use of gentamicin selects for multiresistant *E. coli*.

Streptococcus zooepidemicus

In all years studied, *Streptococcus zooepidemicus* have been uniformly susceptible to penicillin (Table Horse II). Occurrence of resistance to trimethoprim-sulphonamides, however, increased dramatically in the 90s. In the last eight years, about one third to half of the isolates have been resistant to this antimicrobial combination. This is probably due to a concurrent increase in use of trimethoprim-sulphonamides in horses. Resistance to antimicrobials other than trimethoprim-sulphonamides is rare. *Streptococcus zooepidemicus* has a low inherent susceptibility to aminoglycosides (i.e. gentamicin, neomycin and streptomycin) and it can be observed that MIC ranges are above the concentrations that can be obtained during systemic therapy with these antimicrobials (Table Horse II).

MRSI – Methicillin-resistant *Staphylococcus intermedius*

IN THE AUTUMN OF 2006, methicillin resistant *S. intermedius* (MRSI) were isolated from several cases of post-operative wound infection in dogs. Over a period of six months, ten isolates have been confirmed as *mec*A-positive. The isolates were obtained from dogs that had been hospitalised at two large animal hospitals. Apart from being methicillin resistant, and thus resistant to all beta-lactam antibiotics, the isolates had similar resistance patterns and were only susceptible to fusidic acid and tetracycline (See Table 1). Macrorestriction profiling and pulse-field gel electrophoresis (PFGE) using *sma*I restriction enzyme showed that the most MRSI isolates were indistinguishable or closely related. These findings indicate a nosocomial infection with MRSI and also a dissemination of an identical or closely related bacterial clones in the dog population

Methillin-resistance is a rare finding in Sweden. Since 2000, the prevalence of resistance in *S. intermedius* from dogs has been monitored in SVARM with data from 1992. Resistance to cephalothin or oxacillin has been observed only occasionally, but no isolate has been confirmed by PCR to have the *mec*A gene. During 2006, 299 healthy dogs were sampled by perineal swabs and cultured for staphylococci (see also Methicillin-resistant *Staphylococcus aureus*). *Staphylococcus intermedius* was isolated from 135 dogs, but only one isolate was methicillin resistant. The PFGE-pattern differed markedly from those described above, i.e. this isolate was not related to the hospital-related clone.

All infected dogs were treated with antimicrobials on one or several occasions before isolation of MRSI. A multiresistant nosocomial pathogen is, of course, favoured by the excessive use of antimicrobials to dogs, and should be prevented by, for instance, increased awareness of hand hygiene and sanitary actions. Treatment options for infection with this MRSI are drastically limited. It is probably only a matter of time before these strains have acquired resistance to both tetracycline and fusidic acid, which is common among other *S. intermedius*. An emerging problem is approaching if these strains of MRSI will spread further in the dog population.

Table 1. Antibiogram of ten methicillin resistant *Staphylococcus intermedius*.

Antimicrobial	MIC (mg/L)	S/R[a]
Penicillin	>4	R
Cephalothin	>8	R
Cefoxitin	8	R
Oxacillin (+ 2% NaCl)	>16	R
Erythromycin	>32	R
Chloramphenicol	64	R
Clindamycin	>32	R
Tetracycline	<=0.5	S
Fusidic acid	0.25	S
Gentamicin	32	R
Kanamycin	>32	R
Ciprofloxacin	>4	R
Trimethoprim	>32	R

[a] Susceptible (S) or resistant (R) for cut-off values see Appendix 3.

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Dog

Isolates included

Isolates of *E. coli* are from urine samples, submitted either as urine or as dip-slide cultures. *Staphylococcus intermedius* are from skin samples.

Escherichia coli

Table Dog I shows that the proportions of resistant *E. coli* have remained stable during the years studied. Resistance to ampicillin was around 20% and the prevalence of resistance to enrofloxacin, streptomycin, tetracycline, and trimethoprim-sulphonamides were all above or around 10%.

The high proportion of *E. coli* resistant to enrofloxacin throughout the study period is partly explained by the use of a low cut-off value for resistance (>0.12 mg/L), compared to the clinical break-point recommended by CLSI (2004), which is >1 mg/L. Nevertheless, isolates with MIC >0.12 mg/L have decreased susceptibility. Such phenotypes are likely to be explained by at least one mutation in one of the genes encoding the target enzymes of this class of drugs. If an infection caused by such a strain is treated with any fluoroquinolones, there is a risk of further mutations resulting in decreased susceptibility (Drlica, 2003).

Multiresistance occurred in 12% of the isolates, which is somewhat higher than in 2005 (8%). The number of isolates resistant to five or more antimicrobials was 3% in 2006. Of the multiresistant isolates, 66% were resistant to ampicillin, streptomycin and trimethoprim-sulphonamides and 36% were resistant to these antimicrobials and to tetracycline.

Uncomplicated cystitis in dogs is commonly treated with aminopenicillins, and aminopenicillins are by far the most commonly prescribed antimicrobials for dogs (See SVARM 2005 "Antimicrobials prescribed for dogs"). This could explain the proportion of *E. coli* resistant to ampicillin. However, streptomycin is barely prescribed for dogs at all (unpublished data 2006) and trimethoprim-sulphonamides are rarely prescribed, only 3% of all antimicrobial prescriptions for systemic treatment of dogs (See SVARM 2005 "Antimicrobials prescribed for dogs"). Yet, resistance to these substances has been above 10% most years. This could probably be explained by co-resistance between resistance to ampicillin, streptomycin and trimethoprim-sulphonamides. Resistance to these three antimicrobials are also common for commensal *E. coli* (See Resistance in indicator bacteria). Among the isolates from urinary tract infections that were resistant to streptomycin, 83% was also resistant to ampicillin, and for isolates resistant to trimethoprim-sulphonamides, 89% was resistant to ampicillin. The excessive use of aminopenicillins therefore selects for resistance to the other two substances.

Besides aminopenicillins, urinary tract infections are often treated with fluoroquinolones, and occasionally with trimethoprim-sulphonamides. Four percent of the isolates were resistant to all these three antimicrobial groups, which is higher than last year. Of the multiresistant isolates, 34% was resistant to ampicillin, enrofloxacin and trimethoprim-sulphonamides. These figures emphasise the need for culture and susceptibility testing before treatment of recurrent or non-responding urinary tract infections.

Staphylococcus intermedius

This year as many as 91% of the isolates were resistant to penicillin due to production of β-lactamases (penicillinase), this is a peak rate (Table Dog II). Already in the late 70s, 70% of *S. intermedius* were resistant to penicillin (Franklin, 1978) and during the last decade the resistance rate has been around 80%. Besides penicillin, resistance to clindamycin, erythromycin, fusidic acid, streptomycin or tetracycline was common in 2006. Resistance rates for these substances, except for tetracycline, have been stable over the past six years (data not shown, P>0.05, respectively, Chi-square for trend). Noteworthy, resistance to trimethoprim-sulphonamides is low, possibly because this combination currently constitutes only 3% of all antimicrobial prescriptions to dogs (see SVARM 2005 "Antimicrobials prescribed for dogs") and consequently the selective pressure is low. Resistance to tetracycline has increased over the past six years (P<0.03, Chi-square for trend), which probably can be explain by co-selection through clindamycin use (see discussion below).

Table Dog I. Occurrence of resistance among *Escherichia coli* from dogs 1992-2006 and distribution of MICs for isolates from 2006. Isolates are from diagnostic submissions of urinary tract samples.

	Resistance							Distribution (%) of MICs[a] (mg/L)									
Substance (%)	**1992-1994** n=245	**1995-1997** n=296	**1998-2000** n=418	**2001-2003** n=621	**2004** n=247	**2005** n=304	**2006** n=366	**≤0.12**	**0.25**	**0.5**	**1**	**2**	**4**	**8**	**16**	**32**	**>32**
Ampicillin	18	18	18	18	19	17	20				1.4	42.1	34.4	1.9	20.2		
Enrofloxacin	9[c]	9[c]	10[c]	9[c]	12	9	10	89.9	1.4	2.5	1.4	5.2					
Gentamicin	2[d]	1[d]	2[d]	2[d]	1	1	2					92.9	5.2	0.6	0.6	0.8	
Nitrofurantoin	3	3	1	2	1	2[h]	2								96.2	1.9	1.9
Streptomycin	16	18	15[e]	15	13	14	14						6.6	45.4	32.0	2.2	13.9
Tetracycline	16	14	12	11[f]	13	7	10				28.7	54.1	7.1	0.3	9.8		
Trim/Sulph.[b]	9	8	11	11[g]	17	8	12			85.8	1.4	0.8	0.3	11.8			

[a] 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 cut-off values defining resistance; [b] Concentration of trimethoprim given, tested in concentration ratio 1/20 (trimethoprim/sulphamethoxazole); [c] Cut-off value for enrofloxacin is >0.25 mg/L until 2002; [d] Cut-off value for gentamicin >8 mg/L until 2002; [e] 417 isolates tested; [f] 617 isolates tested; [g] 620 isolates tested; [h] 302 isolates tested.

Table Dog II. Occurrence of resistance among *Staphylococcus intermedius* from dogs 1992-2006 and distribution of MICs for the isolates from 2006. Isolates are from diagnostic submissions of samples from skin.

	Resistance (%)							Distribution (%) of MICs[a] (mg/L)									
Antimicrobial	1992-1994 n=304	1995-1997 n=322	1998-2000 n=433	2001-2003 n=382	2004 n=159	2005 n=126	2006 n=89	≤0.12	0.25	0.5	1	2	4	8	16	32	>32
Cephalothin	<1	<1	0	1	2	1	0					100					
Clindamycin	12	20	21	18	21	18	16				80.9		3.4	15.7			
Enrofloxacin	-	-	-	2[e]	3	3	1	61.8	33.7	3.4	1.1						
Erythromycin	21	28	27		30	22	25			74.2	1.1			24.7			
Fusidic acid	9	14	20[d]	20[f]	27	25	23					73.0	4.5	22.5			
Gentamicin	<1	<1	<1	0	1	1	0					100					
Nitrofurantoin	1	1	<1	1	0	1	0								100		
Oxacillin	1	2	1	2	2	1	0			100							
Penicillin[b]	79	80	80	80	80	84	91										
Streptomycin	-	-	-	22[e]	31	28	29						66.3	3.4		1.1	29.2
Tetracycline	24	12	28	25[g]	29	31	37				59.6	2.2	1.1		37.1		
Trim/Sulph[c]	1	2	1	3	10	6	1			59.6	36.0	3.4		1.1			

[a] 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] Denotes β-lactamase production; [c] Concentration of trimethoprim given, tested in concentration ratio 1/20 (trimethoprim/sulphamethoxazole); [d] 421 isolates tested; [e] 273 isolates tested; [f] 346 isolates tested; [g] 381 isolates tested.

Multiresistance occurred in 38% of the isolates, which is higher than in 2003 (26%) and 2005 (33%), and of the same magnitude as in 2004. Resistance to penicillin, clindamycin and erythromycin was the most common phenotype, occurring in 41% of multiresistant isolates. In 2006, 10 isolates (11%) were resistant to five or more antimicrobials. Of these, nine were resistant to clindamycin, erythromycin, penicillin, streptomycin and tetracycline. Interestingly, resistance to enrofloxacin occurred only in multiresistant phenotypes. In *S. intermedius*, resistance to macrolides is commonly mediated by erm-genes, and if these genes are constitutively expressed, the bacteria will be resistant also to lincosamides (clindamycin) and streptogramin B. In this material, 63% of isolates resistant to erythromycin were also resistant to clindamycin.

Since the tested isolates are from diagnostic submissions of samples from skin, there is a high probability of bias towards dogs with recurrent skin infections, previously treated with antimicrobials. A prospective study by Holm *et al.*, (2002) showed higher levels of multiresistance among isolates from recurrent compared to those from first-time pyoderma. This probably explains the high levels of resistance in this material. Clindamycin and cephalosporins are commonly used to treat pyoderma in dogs. With the high proportion of multiresistant isolates treatment with e.g. clindamycin will co-select for resistance to erythromycin, streptomycin, and tetracycline, despite the fact the two latter substances are rarely used in treatment of pyoderma.

Resistance to cephalothin or oxacillin was recorded only occasionally and is probably due to high production of β-lactamases, and not to the presence of *mec*A gene. At SVA, all isolates with high MIC of oxacillin (>2 mg/L) are retested at a lower temperature (33–34 °C) and with 2% NaCl added to the broth, according to CLSI (2006). If oxacillin MIC still is high, the isolates are examined for *mec*A gene with PCR (See Highlight on methicillin resistant *S. intermedius*).

SVARMpat –getting settled in field practice

IN 2005, incorporating SVARMpat into SVARM strengthened monitoring of antimicrobial resistance in specific animal pathogens. The purpose of SVARMpat is to increase the knowledge on resistance in animal pathogens from farm animals (For more information see SVARM 2005). The programme is run in co-operation between the National Veterinary Institute (SVA) and the Swedish Animal Health Service and is financed by the Swedish Board of Agriculture. Results will be reported yearly in the SVARM report, and in addition three times yearly in newsletters directly to veterinary practitioners. The purpose of the newsletters is to continuously inform practitioners on activities and results but also to deepen their knowledge on antimicrobials, antimicrobial treatment and resistance.

Hitherto, an important activity in SVARMpat has been to encourage practitioners and pathologists to submit samples for microbiological culture and susceptibility testing. Special focus has been given to sampling of calves, pigs, or sheep with pneumonia and/or diarrhoea and swine with dermatitis. This is because currently, the number of tested *Escherichia coli* from cattle and sheep, *Brachyspira* spp and *Staphylococcus hyicus* from pigs and Pasteurella and Mannheimia spp. from cattle, pigs and sheep is too small for valid conclusions on prevalence of resistance and trends.

Cat

Isolates included

Isolates of *E. coli* are from urine samples, submitted either as urine or as dip-slide cultures.

Escherichia coli

Resistance to ampicillin, streptomycin, or tetracycline were the most common resistance traits. The levels of resistance vary over the study period but there are no discernible trends (Table Cat I).

As for dogs, the high proportion of *E. coli* resistant to enrofloxacin throughout the study period is partly explained by the low cut-off value for resistance (>0.12 mg/L), which is chosen for fluoroquinolones in SVARM, compared to the break-point recommended by e.g. CLSI (2004), which is >1 mg/L. As mentioned above, strains with MIC >0.12 mg/L are less susceptible and there is a risk for further mutations during fluoroquinolone treatment.

Seventeen percent of the isolates were multiresistant, a higher figure than in 2005. Sixty-two percent of the multiresistant isolates were resistant to ampicillin, tetracycline, and streptomycin. This year two isolates (2% of all) were resistant to more than five antimicrobials. Urinary tract infections in cats are often treated with aminopenicillins or fluoroquinolones. This year, 3% of the isolates were resistant to both these antimicrobials. The observed high levels of resistance in *E. coli* from cats show that the choice of antimicrobials for treatment may be severely limited and must be based on culture and susceptibility tests.

Table Cat I. Occurrence of resistance among *Escherichia coli* from cats 1992-2006, and distribution of MICs for the isolates from 2006. Isolates are from diagnostic submissions of urine samples.

	Resistance (%)						Distribution (%) of MICs[a] (mg/L)									
Antimicrobial	1992-1997 n=61	1998-2000 n=74	2001-2003 n=135	2004 n=55	2005 n=74	2006 n=95	≤0.12	0.25	0.5	1	2	4	8	16	32	>32
Ampicillin	26	34	27	18	20	26					52.6	20.0	1.1	26.3		
Enrofloxacin	5[c]	8[c]	13[c]	5	11	5	94.7		1.1	2.1	2.1					
Gentamicin	0[d]	3[d]	5	0	0	1					90.5	8.4	1.1			
Nitrofurantoin	2	2	1	2	4	2								97.9		2.1
Streptomycin	25	18	21	9	18	19						7.4	52.6	21.1		18.9
Tetracycline	28	16	16	13	12	15				32.6	48.4	4.2		14.7		
Trim-Sulph.[b]	7	10	13	13	3	6			91.6	2.1			6.3			

[a] White fields denote the 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 Concentration of trimethoprim given, tested in concentration ratio 1/20 (trimethoprim/sulphamethoxazole); [c] Cut-off value >0.25 (mg/L) until year 2002; [d] Cut-off value >8 mg/L.

In addition, specific and more elaborated, studies have been initiated within the framework of SVARMpat:

- Comparison of antimicrobial resistance in indicator bacteria (*E. coli* and *Enterococcus* spp.) from healthy cattle of different ages from organic and conventional dairy farms.
- Comparison of antimicrobial resistance in indicator bacteria (*E. coli* and *Enterococcus* spp.) from healthy young pigs (<12 weeks of age) in relation to incidence of post-weaning diarrhoea and antimicrobial use.
- Investigation of prevalence of MRSA among 500 finisher pigs in 100 different herds. This study is completed and the results presented in the highlight on MRSA.
- Improved routine diagnostics of *E. coli* from pigs by PCR analysis for genes coding virulence factors (toxin production and adhesion factors). Today *E. coli* isolated from clinical submissions are only serotyped and tested for antimicrobial susceptibility. After evaluation of a PCR for toxin and adhesions factors, *E. coli* will in the future be routinely examined for these factors and only isolates positive for virulence factors will be serotyped and tested for antimicrobial susceptibility. Such knowledge could help practitioners to initiate treatment only when "true" pathogens are present, i.e. *E. coli* that produce toxin and have adhesions factors.
- Investigation of antimicrobial resistance in indicator bacteria (*E. coli* and *Enterococcus* spp.) from different age groups of healthy sheep.
- Investigation of prevalence of udder pathogens, including antimicrobial susceptibility, from ewes with acute, clinical mastitis and dairy cows with subclinical mastitis.

Appendix 1: Demographic data

AGRICULTURAL STATISTICS are provided by Statistics Sweden in collaboration with the Board of Agriculture and published annually as a Yearbook of Agricultural Statistics and continuously as Statistical Messages (SM). The Yearbook and Statistical Messages are is available on the Internet via the websites for Statistics Sweden (www.scb.se) or the Board of Agriculture (www.sjv.se).

Annual figures on number of animals and holdings are given in Table AP1 I & II, and on numbers and volumes of animals slaughtered in Table AP1 III & IV. For details on methodology, see the respective sources of the statistics.

Over the last two decades, the total number of dairy cows, pigs, and laying hens has decreased notably concomitantly with an increase in herd size. In the same period, the number of beef cows and sheep as well as the number of broilers slaughtered has increased.

Table AP1 I. Number of livestock and horses (in thousands) 1980-2006 (Yearbook of Agricultural Statistics, Sweden 2006 and Statistical Message JO 20 SM 0602).

Animal Species	1980[a]	1985[a]	1990	1995	2000	2004	2005	2006
Cattle								
Dairy cows	656	646	576	482	428	404	393	388
Beef cows	71	59	75	157	167	172	177	178
Other cattle >1 year	614	570	544	596	589	539	527	530
Calves <1 year	595	563	524	542	500	514	508	496
Total, cattle	**1 935**	**1 837**	**1 718**	**1 777**	**1 684**	**1 629**	**1 605**	**1 590**
Pigs								
Boars & sows	290	260	230	245	206	195	188	187
Fattening pigs >20 kg [b]	1 254	1 127	1 025	1 300	1 146	1 094	1 085	1 002
Piglets <20kg [c]	1 170	1 113	1 009	769	566	528	539	492
Total, swine	**2 714**	**2 500**	**2 264**	**2 313**	**1 918**	**1 818**	**1 811**	**1 680**
Sheep								
Ewes and rams	161	173	162	195	198	220	222	244
Lambs	231	252	244	266	234	246	249	262
Total, sheep	**392**	**425**	**406**	**462**	**432**	**466**	**471**	**505**
Laying hens								
Hens	5 937	6 548	6 392	6 100	5 670	4 995	5 065	4 524
Chickens reared for laying	2 636	2 159	2 176	1 812	1 654	1 625	1 697	1 646
Total, hens	**8 573**	**8 708**	**8 568**	**7 912**	**7 324**	**6 620**	**6 762**	**6 170**
Turkeys								
Total, turkeys	-[d]	-	-	-	-	-	**121**	-
Horses								
Total, horses	-	-	-	-	-	**283**	-	-

[a] For 1980 and 1985 only cattle and sheep at premises with more than 2 ha counted; [b] Before 1995, the figure denotes pigs above 3 months of age; [c] Before 1995, the figure denotes pigs below 3 months of age; [d] Data not available.

Table AP1 II. Number of holdings with animals of different types, 1980-2006 (Yearbook of Agricultural Statistics, Sweden 2006 and Statistical Message JO 20 SM 0602).

Animal Species	1980	1985	1990	1995	2000	2004	2005	2006
Cattle								
Dairy cows	44 143	35 063	25 921	17 743	12 676	9 147	8 548	8 027
Beef cows	12 436	10 310	10 883	17 069	13 861	13 013	12 821	12 447
Other cattle >1 year	63 179	52 652	42 696	39 160	30 457	26 291	24 808	23 700
Calves <1 year	62 314	52 001	41 986	36 542	27 733	24 116	22 888	21 752
Total holdings with cattle	**70 503**	**58 872**	**47 292**	**41 990**	**32 063**	**27 626**	**26 179**	**25 054**
Sheep	10 238	10 595	9 749	10 037	8 089	8 239	7 653	9 152
Pigs	26 122	19 937	14 301	10 753	4 809	3 194	2 794	2 414
Laying hens	23 603	17 531	12 900	9 593	5 678	5 376	4 916	4 877
Chickens reared for laying	5 093	2 714	1 875	1 405	715	803	634	528
Broilers	-[a]	-	-	-	-	237	234	192
Turkeys	-	-	-	-	-	-	383	-
Horses	-	-	-	-	-	56 000	-	-

[a] Data not available.

Table AP1 III. Number of animals slaughtered (in thousands) at slaughterhouses, 1980-2006. (Yearbook of Agricultural Statistics, Sweden 1981, 1986, 1991 & 2006 and Statistical Message JO 48 SM 0702).

Animal Species	1980	1985	1990	1995	2000	2004	2005	2006
Cattle								
Cattle >1 year	574	584	523	502	490	458	433	433
Calves < 1 year	130	152	70	30	39	34	33	33
Total, cattle	**704**	**736**	**593**	**532**	**529**	**492**	**466**	**466**
Pigs	4 153	4 283	3 653	3 743	3 251	3 365	3 159	3 037
Sheep	302	328	280	189	202	193	206	213
Broilers	40 466[a]	36 410[a]	38 577[a]	61 313	68 617	69 628	73 458	72 906

[a] Data supplied by the National Food Administration.

Table AP1 IV. Quantity of livestock slaughtered (in 1000 tonnes) at slaughterhouses, 1990-2006 (Yearbook of Agricultural Statistics, Sweden 1991 & 2006 and Statistical Message JO 48 SM 0702).

Animal Species	1990	1995	2000	2004	2005	2006
Cattle						
Cattle >1 year	139.5	140.1	145.4	137.8	131.4	132.9
Calves < 1 year	6.8	3.2	4.4	4.6	4.5	4.5
Total, cattle	**146.3**	**143.3**	**149.8**	**142.4**	**135.9**	**137.4**
Pigs	293.1	308.8	277.0	294.5	275.1	265.6
Sheep	5.0	3.5	3.9	3.8	4.1	4.2
Broilers	44.0[a]	73.6[a]	89.9	91.2	96.2	95.5

[a] Data supplied by the National Food Administration.

Appendix 2: Materials and methods, use of antimicrobials

Source for the statistics

The antimicrobial drugs used in veterinary medicine in Sweden are only available on veterinary prescription. Furthermore, antimicrobial drugs are dispensed through pharmacies only. Sales statistics are available from Apoteket AB (The National Corporation of Swedish Pharmacies). From year 2003, statistics on drug sales is based on electronic records of amount of drugs dispensed at or from pharmacies, i.e. sales statistics. Data for previous years are the amount of antimicrobial products sold from the wholesalers to the pharmacies.

Sweden has a long tradition in drug consumption statistics. Apoteket AB, former Apoteksbolaget AB, has since 1976 monitored the consumption of drugs for use in humans mainly by using wholesalers´ statistics. In the case of drugs for animal use, SVA and Apoteket AB have collaborated over the years and data on the total use of antimicrobials for animals in Sweden are available since 1980. For a review of the figures from 1980–2000 as well as references to publications on which that review is based, see SVARM 2000.

Classification of drugs

Veterinary medicinal drugs are classified according to the Anatomical Therapeutic Chemical veterinary classification system (ATCvet) (WHO, Guidelines for ATCvet classification). The system is based on the same main principles as the ATC classification system for substances used in human medicine. In both the ATC and ATCvet systems, drugs are divided into groups according to their therapeutic use. First, they are divided into 15 anatomical groups, classified as QA-QV in the ATCvet system (without Q in the system for human drugs), on basis of their main therapeutic use. Thereafter subdivision is made according to therapeutic main groups, which is followed by a further division in chemical/therapeutic subgroups.

Antimicrobials are classified in the QJ group – general antiinfectives for systemic use. However, antimicrobials can also be found in other groups such as QA (alimentary tract and metabolism), QD (dermatologicals), QG (genito-urinary system) and QS (sensory organs) depending on the therapeutic use.

Inclusion criteria

All veterinary antibacterial drugs authorised for use in animals except dermatologicals, ophtalmologicals and otologicals (i.e., ATCvet codes QA, QG and QJ) were included. Veterinary drugs are preparations authorised for use in animals. Human drugs may be authorised not only for humans, but for animals as well. This latter category is not included in the statistics. However, no such drugs are authorised for use in the major food producing animal species, and the volume sold is very limited.

Drugs with antibacterial activity can also be found in other groups, notably among the antiprotozoals (QP51). Of these, the nitroimidazoles were included earlier but no such substances are presently authorised for use in animals. Sulfaclozine is licensed for treatment of coccidiosis only and has therefore not been included.

The ionophoric antibiotics are presently regulated as feed additives and not sold through pharmacies and are therefore not included in the wholesalers' statistics. However, the Board of Agriculture collects figures on sales of ionophores from the feed mills as a part of the feed control system. As the source differs, data on ionophores are given only in Table AC III.

Distribution of veterinary medicines in Sweden

Marketing of drugs in Sweden is regulated by the Medicinal Products Act, which applies both to human and veterinary drugs. According to the Act, a medicinal product may not be sold until it has been granted marketing authorisation by the Medical Products Agency (MPA). The MPA has issued provisions concerning authorisation, distribution and prescription of veterinary medicinal products. In case there are no authorised veterinary medicinal products for a certain condition, the MPA can also permit special license prescription for a drug.

The state-owned Apoteket AB has exclusive rights regarding retail sales of medicines in Sweden. Apoteket AB operates according to guidelines set out in an agreement with the State. According to the Act only pharmacies run by Apoteket AB are permitted to sell drugs. This implies that veterinarians in Sweden are not permitted to sell drugs, although they may for practical reasons hand over medicines for emergency use. Veterinarians are, however, under no conditions permitted to make a profit from dispensing medicines.

Appendix 3: Materials and methods, resistance monitoring

Sampling strategy

Zoonotic bacteria

Salmonella

Isolates of *Salmonella* from warm-blooded animals (wild and domesticated) are included. Salmonellosis in animals is a notifiable disease in Sweden. It is mandatory that at least one isolate from each notified incident, including incidents detected in the Swedish *Salmonella* control programme, is confirmed at SVA. The first isolate of each serovar, and when appropriate phagetype, from each food animal species in each notified incident is included in the material presented in SVARM. The same inclusion criteria are also used for isolates from other warm blooded animal species, unless the epidemiological situation in a particular year is judged unusual. In year 2006, *Salmonella* was isolated from a total of 77 cats and of these; every fifth isolate in consecutive order was selected for testing (total number of isolates 15).

Campylobacter

Campylobacter were isolated from colon content of healthy dairy cows sampled at abattoirs for isolation of indicator bacteria (see below). All samples collected (n=470) were cultured for *Campylobacter*.

Campylobacter were also isolated from faecal samples collected by convenience from 36 piglet-producing herds from all parts of Sweden in the period April to June 2006. In each herd, one suckling (<5 weeks old) and one weaned piglet (5–12 weeks old) were sampled.

Methicillin resistant *Staphylococcus aureus* (MRSA)

To screen for MRSA from pigs, five pigs in each of 100 pig producing units distributed across the country were sampled. Swabs were taken from the nostrils and samples from each production unit were pooled before analysis.

Indicator bacteria

Indicator bacteria, *Escherichia coli* and *Enterococcus* spp., were isolated from colon content of dairy cows sampled at slaughter by meat inspection staff or abattoir personnel. Eleven abattoirs participated in the collection of samples. These abattoirs are geographically separated and accounted for 84% of the total volume of dairy cows slaughtered in Sweden during 2005.

At each abattoir, an equal number of samples were collected during each of four periods (March, May, September and November). The number of samples collected at each abattoir was proportional to the respective annual volume of dairy cows slaughtered and each sample represents a unique herd. By these measures, bacterial isolates included are from randomly selected dairy cows of Swedish herds.

Indicator bacteria were also isolated from faecal samples from healthy dogs. During the fall of 2006, a total of 299 rectal swabs were taken from healthy dogs attending shows, and from dogs belonging to personnel and veterinary students. In all cases, the swabs were taken from the dogs with their owner's consent.

The rate of colonisation by VRE among broiler chickens was investigated in a separate study on samples obtained through the Swedish *Campylobacter* programme. From these samples, 51 caeca collected at slaughter were selected in order of arrival at SVA in spring and 51 caeca in the fall. Samples selected for culture were from unique flocks but not necessarily from unique production sites.

Animal pathogens

Isolates of animal pathogens included emanate from routine bacteriological examinations of clinical submissions or post-mortem examinations at SVA.

Escherichia coli from pigs and cattle are from the gastro-intestinal tract (gut content, faecal samples or mesenteric lymph nodes) and *Brachyspira* spp. from faecal samples from pigs. *Escherichia coli* from horses are from the genital tract of mares and *Streptococcus zooepidemicus* from the respiratory tract. From dogs and cats *E. coli* isolated from samples of urine are included and in addition, from dogs, *Staphylococcus intermedius* isolated from skin samples.

Isolation and identification of bacteria

Zoonotic bacteria

Salmonella

Salmonella were isolated and identified at the Dept. of Bacteriology, SVA or at regional laboratories in accordance with standard procedures. All samples within official control programmes are cultured according to the procedures detailed by the Nordic Committee on Food Analysis (NMKL Nr 71 5th ed., 1999). Confirmatory identification and serotyping of isolates was performed at the Dept. of Bacteriology, SVA according to the standard procedures of Kaufmann and White. The Dept. of Bacteriology, SVA is accredited for isolation, identification and serotyping of *Salmonella*.

Most isolates of *Salmonella* Typhimurium and *S.* Enteritidis were phagetyped by the Swedish Institute for Infectious Disease Control (SMI), Stockholm using the Colindale scheme.

Campylobacter

Campylobacter spp. from dairy cows and pigs were isolated and identified at SVA according to standard procedures. Briefly, samples from dairy cows were cultured for thermophilic *Campylobacter* spp. by a modified NMKL method (NMKL Nr 119, 1990) using enrichment in Preston broth followed by culture on Preston selective agar and incubation at 42°C. Samples from piglets and weaning pigs were cultured directly on Preston

agar. Identification was based on colony morphology, microscopic appearance including motility and the following phenotypic characteristics: production of oxidase, catalase, hippurate hydrolysis reaction and indoxyl-actetate reaction (Nachamkin, 1999). With these tests, hippurate-positive *C. jejuni* can be identified whereas other isolates are described as hippurate-negative thermophilic *Campylobacter* spp.

Methicillin resistant Staphylococcus aureus (MRSA)

The five swabs from each herd were pooled in 15–20 mL tryptone soy broth (TSB) with 6.5% NaCl and 75 mg/L of aztreonam. After incubation in 37°C for 18–20h, 10 µL was subcultured on mannitol salt agar with lithium chloride (MAST DM 160, Oxoid) and cefoxitin 4 mg/L. After incubation at 37°C over night, colonies with morphology consistent with *S.* aureus were subcultured to bovine blood agar. Identification was based on morphology, production of haemolytic toxins, coagulation of rabbit plasma, DNAse production and aerobic acid production from maltose and trehalose on bromcresol-purple agar.

Indicator bacteria

Escherichia coli

For samples from cattle, approximately 0.5 g of colon content was diluted in 4.5 mL saline. After thorough mixing, 0.1 mL of this suspension was spread on MacConkey agar. Swab samples from dogs were spread directly on MacConkey agar. After incubation overnight at 37°C, one lactose positive colony with morphology typical for *E. coli* was sub-cultured on horse-blood agar (5% v/v), after which the isolate was tested for production of tryptophanase (indole) and β-glucuronidase (p-nitrophenyl-β-D- glucopyranosiduronic acid, PGUA). Only lactose-positive isolates with typical morphology and positive reactions in both tests were selected for susceptibility tests.

Enterococci

Colon content from dairy cows was diluted as described for *E. coli* and cultured on solid media without selective antibiotics. Caecal content from broilers was prepared the same way but cultured on selective plates with vancomycin (16 mg/L).

Culture without selective antibiotics: For samples from cows, 0.1 mL the diluted colon content was spread onto Slanetz-Bartley (SlaBa) agar. Swab samples from dogs were spread directly on SlaBa agar. The plates were incubated for 48 h at 37°C. One colony, randomly chosen, was sub-cultured on bile-esculin agar and blood agar (37°C, 24 h). Colonies with a morphology consistent with enterococci, and with a positive reaction on bile-esculin agar were tested for antimicrobial susceptibility and identified to species level according to Devriese *et al.* (1993) by use of the following biochemical tests: mannitol, sorbitol, arabinose, saccharose, ribose, raffinose and methyl-α-D-glucopyranoside.

Selective culture for vancomycin resistant enterococci: Diluted colon content (0.1 mL) was also cultured on SlaBa with vancomycin (16 mg/L). From plates showing growth of colonies typical for enterococci, at least one colony of each morphological type was sub-cultivated on bile-esculin agar and blood agar (37°C, for 24 h). Identification of presumptive enterococci was performed as above.

Animal pathogens

Animal pathogens were isolated and identified at the Dept. of Bacteriology, SVA with accredited methodology, following standard procedures.

Susceptibility testing

The Dept. of Antibiotics or the Dept. of Bacteriology performed antimicrobial susceptibility tests, with accredited methodology, using dilution methods in cation adjusted Mueller-Hinton broth (CAMBH). Tests were performed following the standards for microdilution of the Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS). The microdilution panels used, VetMIC™, are produced at the Dept. of Antibiotics, SVA. Different panels were used depending on the bacterial species tested and the original purpose of the investigation (monitoring or clinical diagnostics). Minimum inhibitory concentration (MIC) was recorded as the lowest concentration of the antimicrobial that inhibits bacterial growth.

For susceptibility testing of *Brachyspira hyodysenteriae*, a broth dilution method was used (Råsbäck *et al*, 2005). The antimicrobials were dried in serial twofold dilutions in the tissue culture trays with 48 wells per plate. The wells were filled with 0.5 mL of a suspension of bacteria in brain heart infusion broth with 10% foetal calf serum ($1x10^6$–$5x10^6$ CFU/ml). The trays were incubated in an anaerobic atmosphere for four days on a shaker.

Screening for methicillin resistance was performed with microdilution according to CLSI 2006, testing oxacillin with 2% NaCl added to the broth, and in addition oxacillin without added NaCl and cefoxitin. Presence of the *mec*A gene in both *S. aureus* or *S. intermedius* was tested in isolates with a phenotype indicating methicillin resistance at the by polymerase chain reaction (PCR) according to Smyth *et al*. (2001).

For every fifht consecutive enterococcal isolate with MICs of vancomycin above >128 mg/L, the resistance genotype was confirmed with PCR for the *van*A- and *van*B-genes (Dutka-Malen *et al*., 1995).

For interpretation of results for zoonotic bacteria (*Salmonella* and *Campylobacter*) and indicator bacteria (*E. coli* and enterococci) epidemiological cut-off values recommended by the European Committee on Antimicrobial Susceptibility Testing (EUCAST) were used (http://www.escmid.org). When no cut-off value was available, or the range of concentrations tested was inappropriate for the recommended value, a cut-off value was defined on basis of the actual MIC distributions obtained in the SVARM programme. The same approach was used when recommended cut-off values would have cut through distributions of MIC in a manner not in agreement with the concept of wild-type distributions, causing an erroneously high frequency of resistance in single a year(s). This applies to ciprofloxacin and gentamicin in *E. coli*.

Also for animal pathogens the principle of epidemiological cut-off values were used, but the clinical breakpoints recommended for animal pathogens by CLSI, 2002 were also taken into consideration.

Bacitracin values in this report are given in units/mL. In an attempt to convert unit/mL to mg/L we discovered that there appears to be some confusion in the matter. The bacitracin compound used in SVARM is obtained from Sigma and meets the standards set by the United States Pharmacopoeia (USP), stating that one unit is equivalent to 26 µg of the US standard. However, according to the International Standard Preparations, one international unit is equivalent to 13.51 µg. On the other hand, if the bacitracin is of a very high degree of purity, though unstable, it correspond to 66 (-70) units/mg, that is, one unit is equivalent to approximately 15 µg. Feedingstuff grade of bacitracin correspond to 42–50 units/mg (one unit=20–24 µg) (Otten *et al*., 1975).

Quality assurance system

The Dept. of Antibiotics and Dept. of Bacteriology are accredited according to SS-EN ISO/IEC 17025 by the Swedish Board for Accreditation and Conformity Assessment (SWEDAC) to perform antimicrobial susceptibility tests with microdilution methods. The Dept. of Bacteriology is also accredited for isolation and identification of animal pathogens and *Salmonella* according to the same standard.

For susceptibility tests of zoonotic and indicator bacteria, *Escherichia coli* ATCC 25922, *Enterococcus faecalis* ATCC 29212 and *Campylobacter jejuni* CCUG 11284 (analogue to *Campylobacter jejuni* ATCC 33560) were included as quality controls. Relevant control strains were also included and evaluated at least once weekly for animal pathogens. The *B. hyodysenteriae* type strain B78T ATCC 27164T were used for internal performance control.

The Dept. of Antibiotics participates in several proficiency tests for antimicrobial susceptibility testing. These are arranged either by the Community Reference Laboratory (CRL) or as national studies. Likewise, the Dept. of Bacteriology participates in proficiency tests concerning isolation and identification of *Salmonella* spp. and general clinical veterinary bacteriology and susceptibility tests.

Data handling

Records on *Salmonella* and animal pathogens such as source of cultured sample, identification results, antimicrobial susceptibility etc. are routinely registered in an Oracle database at SVA. From this, relevant data were extracted to an Access database.

For indicator bacteria, data on animal species, date of sampling, abattoir and herd or flock of origin were recorded in an Access database on arrival of samples, and the results of culture identification and susceptibility tests were recorded on completion of testing.

Calculations and analysis of data were performed in the computer programs Access, Excel, or EpiInfo.

Concerning confidence limits

When the prevalence of antimicrobial resistance is close to zero, e.g. when one out of 120 isolates is resistant, the question arises how to calculate the prevalence of resistance and its confidence intervals. In the example, the prevalence could be estimated to 0.83% while the 95% confidence interval is trickier. The normal approximation to the binomial distribution would give a lower confidence of -0.8% and an upper confidence limit of 2.5%. The lower limit is nonsensical and indicates the unsuitability of the normal approximation in this case.

There are several ways out of the dilemma; one is to calculate the exact binomial confidence limits, which would be possible in some cases (small number of isolates). Another alternative is to run Monte-Carlo simulations based on the beta-distribution which is possible but quite laborious for a huge set of data since each prevalence estimate has to be simulated 10 000 times. Finally the relationship between the F-distribution, the beta-distribution and the binomial distribution can be used. This gives the formulae that enable calculations of the confidence interval (Rao, 1965). Using this approach, the confidence intervals in the example would be 0.021% and 4.6%.

In conclusion, the normal approximation to the binomial distribution might be unsuitable when the prevalence is close to 0% or close to 100% since the approximation might lead to confidence intervals lower than 0% or higher than 100%. Moreover, when the prevalence of resistance is less than 5% using the link between the F-distribution and the binomial distribution yield different confidence intervals compared to those obtained from the normal approximation and should accordingly be preferred.

Table AP3 I. Cut-off values (mg/L) defining resistance. Values in bold lettering are concordant with current (April 2007) epidemiological cut-off values presented by EUCAST, values in italic lettering deviate from values presented by EUCAST and for values in normal lettering no EUCAST value is available (See "Susceptibility testing" above for details).

Antimicrobial	*Brachyspira* spp.	*Campylobacter jejuni*	*Campylobacter* spp.	*Enterococcus* spp. (indicator)	*Escherichia coli* (indicator)	*Escherichia coli* (pathogen; pig, cattle, horse)	*Escherichia coli* (pathogen; dog, cat)	*Klebsiella* spp.	*Salmonella* spp.	*Staphylococcus intermedius*	*Streptococcus zooepidemicus*
Ampicillin		**>8**	**>16**	**>4**	**>8**	**>8**	**>8**		**>4**		>8
Bacitracin[a]				**>32**							
Cefotaxime					**>0.25**			**>0.12**	**>0.5**		
Ceftiofur					**>1**	*>2*		>2	**>2**		
Cefoxitin										>4	
Cephalothin										>2	
Chloramphenicol				**>32**	**>16**			>16	**>16**		
Clindamycin										*>4*	
Ciprofloxacin					*>0.06*				**>0.06**	**>1**	
Enrofloxacin		>0.5	>0.5		**>0.12**	**>0.12**	**>0.12**	>0.25	>0.25	>0.5	
Erythromycin		**>4**	**>16**	**>4**						*>4*	
Florfenicol					**>16**	**>16**		>16	**>16**		>8
Fusidic acid										>4	
Gentamicin		**>1**	**>2**	**>32**	*>4*	*>4*	*>4*	**>2**	**>2**	>4	
Kanamycin				>1024	>16				>16		
Linezolid				**>4**							
Nalidixic acid		**>16**	**>32**		**>16**			>16	**>16**		
Narasin				>2							
Neomycin					**>8**	**>8**		>8	**>4**		
Nitrofurantoin							*>32*			>32	
Oxacillin										**>1**	
Penicillin										[e]	>1
Spiramycin											>16
Streptomycin				**>512/>128**[c]	**>16**	*>32*	*>32*	>32	>32	>32	
Sulphamethoxazole					>256			>256	>256		
Tetracycline		**>2**	**>2**	**>2**	**>8**	**>8**	**>8**	>8	**>8**	>8	>8
Tiamulin	>2										
Trimethoprim					**>2**			>8	**>2**		
Trimethoprim & sulphamethoxazole[b]						*>4*	*>4*		>0.5	>2	>4
Tylosin	>16										
Vancomycin				**>4**							
Virginiamycin				**>32/>4**[d]							

[a] MIC in U/mL; [b] Concentration of trimethoprim given, tested with sulphamethoxazole in concentration ration 1/20; [c] *Enterococcus faecalis* >512 mg/L, other enterococci >128 mg/L; [d] E. faecalis >32 mg/L, E. faecium and E. hirae >4 mg/L; [e] β-lactamase production.

Appendix 4: Antimicrobial agents licensed

ANTIMICROBIAL AGENTS licensed for therapy in veterinary medicine in Sweden year 2006 are listed in Table AP4 I.

Only substances licensed for systemic, oral, intrauterine or intramammary use are included (ATCvet codes QJ, QG, QA and QP). Data from FASS VET. 2006. For explanation of ATCvet code, see Appendix 2.

Table AP4 I. Antimicrobial agents authorised for therapeutic use in cattle, sheep, pigs, poultry, horses, dogs and cats in Sweden, 2006. Routes of administration are indicated [a].

		Animal species						
Antimicrobial agent	**ATCvet code**	**Cattle**	**Sheep**	**Pigs**	**Poultry**	**Horses**	**Dogs**	**Cats**
Tetracyclines								
Doxycycline	QJ01A A02			O			O	O
Oxytetracycline	QJ01A A06, QG01A A07	I O U	I U	I O U	O		O	O
beta-lactams, penicillins								
Ampicillin	QJ01C A01	O		O		O	O	O
Amoxicillin	QJ01C A04	I		I			I O	O
Amoxicillin/Clavulanic acid	QJ01C R02			I			I O	I O
Penicillin G, sodium	QJ01C E01/QJ51C E09	I M		I		I		
Penicillin G, procaine	QJ01C E09	I	I	I		I	I	I
Penicillin G, penetamathydroiodide	QJ01C E90	I						
beta-lactams, cephalosporins								
Cephalexin	QJ01D B01						O	O
Cefadroxil	QJ01D B05						O	O
Ceftiofur	QJ01D D90	I						
Sulphonamides /Trimethoprim								
Sulphadiazine/Trimethoprim	QJ01E W10	I	I	I		I O	O	
Sulphadoxine/Trimethoprim	QJ01E W13	I		I		I		
Sulphonamides								
Formosulphatiazole	QA07A B90	O	O	O		O	O	
Sulphaclozin	QP51A G04				O			
Macrolides								
Spiramycin	QJ01F A02	I						
Tylosin	QJ01F A90	I		I O	O		I	I
Lincosamides								
Clindamycin	QJ01F F01						O	O
Pirlimycin	QJ51F F90	M						
Aminoglycosides								
Gentamicin	QJ01G B03					I U	I	
Dihydrostreptomycin (DHS)	QA07A A90	O U	O U	O U		O U	O	O
Fluoroquinolones								
Enrofloxacin	QJ01M A90	I		I	O		I O	I O
Danofloxacin	QJ01M A92	I		I				
Marbofloxacin	QJ01M A93						O	O
Orbifloxacin	QJ01M A95						O	
Ibafloxacin	QJ01M A96						O	O
Pleuromutilins								
Tiamulin	QJ01X X92			I O				
Valnemulin	QJ01X X94			O				
Combinations								
Penicillin G, procaine/DHS	QJ01R A01, QJ51R C23	I M	I	I		I	I	I
Penicillin G, benzatin/DHS	QJ51R C24	M						
Penicillin G, ester/Framycetin	QJ51R C25	M						
Penicillin G, ester/DHS	QJ51R C25	M						

[a] O = oral; I = injection; U = intrauterine; M = intramammary.

Appendix 5: References

CLSI. Performance Standards for Antimicrobial Disc and Dilution Susceptibility Tests for Bacteria Isolated from Animals; Informational Supplement. NCCLS document M31-S1. [ISBN 1-56238-534-8]. NCCLS, Wayne Pennsylvania, USA. 2004.

CLSI. Performance Standards for Antimicrobial Susceptibility Testing; Sixteenth Informational Supplement. CLSI document M100-S16 [ISBN 1-56238-588-7]. Clinical and Laboratory Standards Institute, Wayne Pennsylvania, USA, 2006.

Devriese, LA., Pot, B. and Collins, MD. Phenotypic identification of the genus *Enterococcus* and differentiation of phylogenetically distinct enterococcal species and species groups. J Appl Bacteriol 1993, 75:399-408.

Drlica, K. The mutant selection window and selection of resistance. J Antimicrob Chemother 2003, 52:11-17.

Dutka-Malen, S., Evers S. and Courvalin, P. Detection of glycopeptide resistance genotypes and identification to the species level of clinically relevant enterococci by PCR. J Clin Microbiol 1995, 33:24-27.

EUCAST – the European Committee on Antimicrobial Susceptibility Testing. "Data from the European Committee on Antimicrobial Suseptibility Testing (EUCAST) website 2006-03-16, http://www.eucast.org".

FASS VET. 2006 (Swedish list of permitted veterinary drugs). Läkemedelsindustriföreningen, Stockholm, Sweden, 2006. ISSN 0347-1136.

Franklin, A. [Stafylokocker från hud. Biokemi och antibiotikaresistens] Staphylococci from skin. Biochemical tests and antibiotic resistance. In proceedings from: Nordic Veterinary Congress, Åbo, Finland, 1978, p 355.

Franklin, A. Antibiotikakänslighet hos *Escherichia coli*-stammar isolerade från spädgrisar i Sverige 1964-68 samt 1974-75 [Antibiotic susceptibility of *Escherichia coli*-strains isolated from piglets in Sweden 1964-68 and 1974-75]. Svensk VetTidn 1976, 28:845-852.

Franklin, A. Antimicrobial drug resistance in porcine enterotoxigenic *Escherichia coli* of O-group 149 and non-enterotoxigenic *Escherichia coli*. Vet Microbiol 1984, 9:467-475.

Gunnarsson, A., Franklin, A., Horn af Rantzien, M. and Landén, A. Resistensundersökning av svenska isolat av Treponema hyodysenteriae. Svensk VetTidn. 1991, 43:349-352.

Holm, B., Petersson U., Mörner A., Bergström K., Franklin A., and Greko C. Antimicrobial resistance in staphylococci from canine pyoderma: a prospective study of first-time and recurrent cases. Vet Rec 2002, 151:600-605.

Jesse, T.W., Englen, M.D., Pittenger-Alley, L.G., Fedorka-Cray, P.J., 2006, Two distinct mutations in *gyrA* lead to ciprofloxacin and nalidixic acid resistance in *Campylobacter coli* and *Campylobacter jejuni* isolated from chickens and beef cattle. J Appl Microbiol 100, 682-688.

Landin, H., 2006, Treatment of mastitis in Swedish dairy production (in Swedish with English summary). Svensk Veterinärtidning 58, 19-25.

Nachamkin, I. Manual of Clinical Microbiology, 7th ed. 1999, p 716-726.

Otten, H., Plempel, M. and Siegenthaler, W. Antibiotika-Fibel. Antibiotika und Chemotherapeutika Therapie mikrobieller Infektionen. George Thieme Verlag, Stuttgart, 1975, pp 542-545.

Rao, CR. Linear statistical inference and its applications. John Wiley and Sons, 1965.

Råsbäck, T., Fellström, C., Bergsjo, B. Cizek, A., Collin, K., Gunnarsson, A., Jensen, S.M.,Mars, A., Thomson, J., Vyt, P., Pringle, M. Assessment of diagnostics and antimicrobial susceptibility testing of Brachyspira species using a ring test. Vet Microbiol 2005, 109:229-243

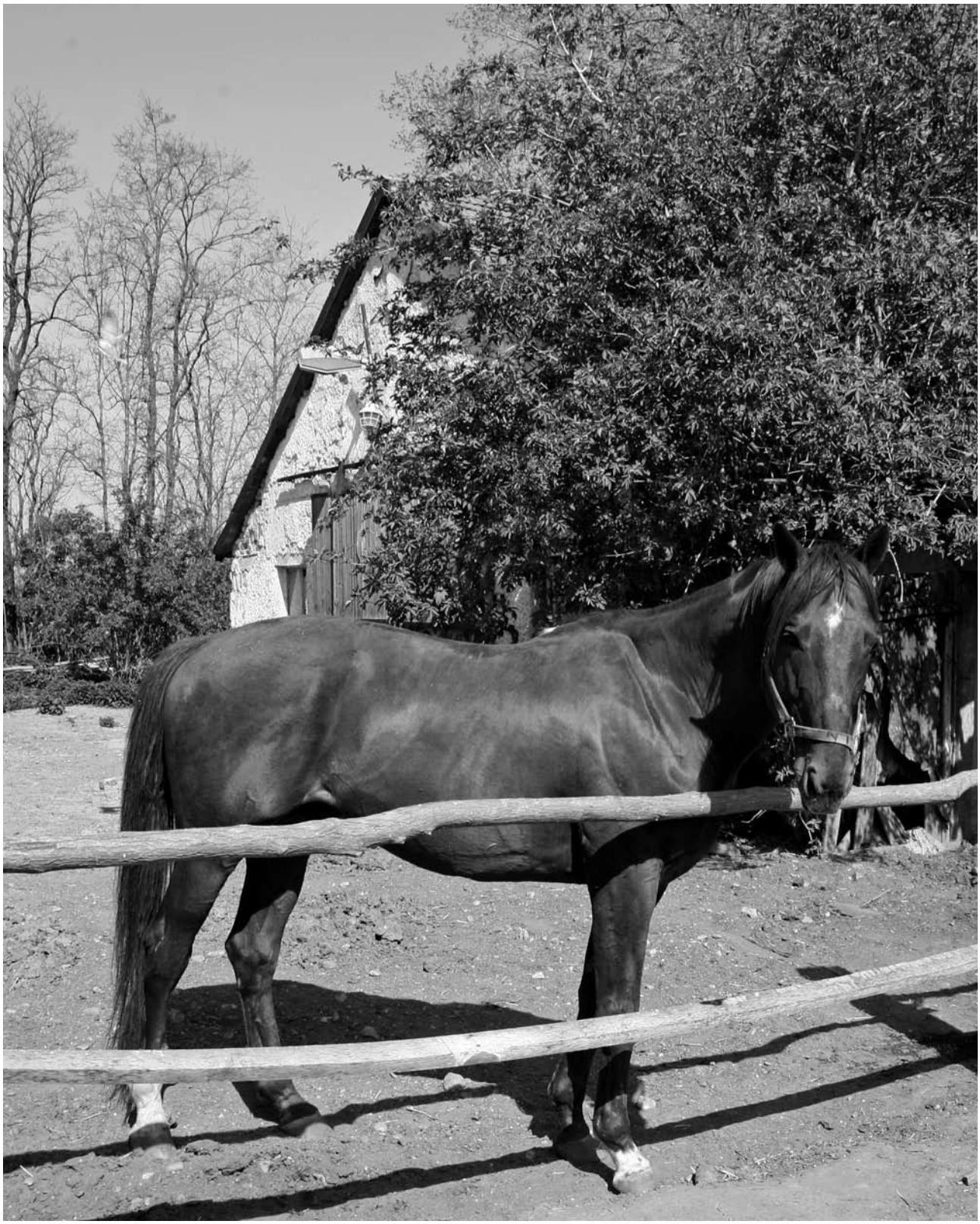
Smyth, R.W., Kahlmeter, G., Liljequist, B.O., Hoffman, B. Methods for identifying methicillin resistance in *Staphylococcus aureus*. J Hosp Inf 2001, 48:103-107.

SVARM, Swedish Veterinary Antimicrobial Resistance Monitoring. The National Veterinary Institute (SVA), Uppsala, Sweden. ISSN 1650-6332 (available at http://www.sva.se).

SWEDRES 2006. A report on Swedish antibiotic utilisation and resistance in human medicine. The Swedish strategic programme for the rational use of antimicrobial agents (STRAMA) and the Swedish institute for infectious disease control. ISSN 1400-3473 (available at http://www.strama.se).

WHO. Guidelines for ATCvet classification, 4th ed. WHO Collaborating Centre for Drug Statistics Methodology. 2002. Oslo, Norway. ISBN 82-90312-41-5 8.

Zwensson, J. [Antibacterial resistance in bacteria isolated from healthy Swedish dogs – indicator bacteria – *E. coli* and *Enterococcus* spp and Staphylococcus spp isolated from skin] in Swedish, English summary. 2007. Swedish university of Agricultural Sciences, Uppsala, Sweden. ISSN 1652-8697 (also available at http://www.epsilon.slu.se).



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