

DANMAP: monitoring antimicrobial resistance in Denmark

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Abstract

The objectives of the Danish Integrated Antimicrobial Resistance Monitoring and Research Programme (DANMAP) are to monitor trends in resistance among bacteria from animals, food and humans, to monitor the consumption of antimicrobial agents and to determine the association between consumption and occurrence of resistance and to model transmission of resistance from animals to humans. DANMAP is based on the examination of representative bacterial isolates of animal and human pathogens, of zoonotic bacteria and of indicator bacteria. For food animals, both diseased and the healthy populations are studied. Isolates from all three reservoirs are examined for their susceptibility to a basic panel of antibiotics that includes representatives of the major classes of compounds, making comparison of resistance levels in the reservoirs possible. Isolates are stored in a strain collection and are available for further study. The data are stored in databases as MIC values or mm inhibition zones with all identifiers. A system for recording all use of the veterinary medicines, VETSTAT, is currently under implementation. For production animals, the consumption will be recorded for each herd, providing a basis for detailed modelling of the effect of consumption on resistance. © 2000 Elsevier Science B.V. and International Society of Chemotherapy. All rights reserved.

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1. Background

In 1994 and 1995 concerns arose that the use of the growth promoting antimicrobial avoparcin might be associated with the occurrence of vancomycin resistant *Enterococcus faecium* in humans [1–3]. In Denmark, there was also considerable public concern about a perceived overuse of antibiotics in Danish pig production and the effects on antimicrobial resistance.

The Danish government therefore decided to fund a programme to monitor antimicrobial resistance and the consumption of antimicrobial agents. The programme, Danish Integrated Antimicrobial Resistance Monitoring and Research Programme (DANMAP) represents a close collaboration between veterinary, food and health authorities in order to provide comparable data for food animals, food and humans. The objectives of the programme are to provide data on trends in the occurrence of antimicrobial resistance, to monitor consumption of antimicrobial agents and to explore any association between the occurrence of resistance and the use of antimicrobials.

The bacterial species included in the programme comprise human and animal pathogens, zoonotic bacteria and indicator bacteria. All isolates from animals and from foods as well as the human isolates of zoonotic bacteria are kept in a strain collection and are available for further study.

The results of the monitoring programme are reported annually [4–6], while the results of detailed studies are published internationally in relevant scientific journals.

The present paper describes the design of the programme.

2. Sampling procedures

2.1. Food animals

Samples from animals are collected from slaughter animals as well as from the diseased population. The former is sampled by collection of caecal contents (pigs), cloacal swabs (broilers) or rectal contents (young bulls). The samples are collected monthly by meat inspection staff (pigs and cattle) or weekly (broilers) and the number of samples per plant determined in

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proportion to the annual throughput of the plant. Only one sample per herd or flock is collected on any one sampling occasion. The pig and broiler slaughter plants included in the programme account for between 95 and 98% of the total national production of these species, while for cattle the figure is about 80%. Formal randomisation is not carried out, and while the procedure resembles a stratified sampling plan there is a bias towards inclusion of large herds in preference to small. Except for cattle, the herd identity is recorded at sampling and when the data are processed, it is possible to ensure that any one herd is included only once.

Isolates of animal pathogens are collected as a systematic random sample among isolates from diagnostic submissions to the Danish Veterinary Laboratory or to the industry-owned Cattle Health Laboratory at Ladelund and the laboratory run by the Federation of Danish Pig Producers and Slaughterhouses at Kjellerup. Between them, these laboratories process close to 100% of diagnostic samples from food animals in Denmark.

The bacteria included from food animals and the target number of isolates are shown in Table 1.

2.2. Foods

Most samples from foods originate from a nationwide, continuous monitoring of food borne pathogens (*Salmonella*, *Campylobacter*, *Listeria monocytogenes*, *Yersinia enterocolitica* and *Staphylococcus aureus*), carried out by all 32 Municipal Food and Environmental Laboratories (MFELs). A non-randomised subset of the isolates are selected by the Danish Food Directorate for susceptibility testing and inclusion in DANMAP. The isolates of indicator bacteria (*Escherichia coli* and enterococci) result from dedicated sampling of predetermined foodstuffs. This sampling is carried out by 21 of the MFELs under the direction of the Food Directorate.

Table 1
Bacteria from food animals included in the DANMAP programme^a

	Pigs	Cattle	Poultry
<i>Actinobacillus pleuropneumoniae</i>	100		
<i>Staphylococcus hyicus</i>	100		
Coagulase negative staphylococci		200	
<i>Staphylococcus aureus</i>		200	
<i>Salmonella</i> spp.	300	100	100
<i>Campylobacter</i>	100	100	100
<i>Escherichia coli</i> (diagnostic submissions)	100	100	100
<i>Escherichia coli</i> (normal population)	300	100	200
<i>Enterococcus faecium/faecalis</i>	300	100	200

^a Figures represent the target number of isolates.

2.3. Humans

Bacterial isolates from humans include *Salmonella*, *Campylobacter* and *Y. enterocolitica*. These are recovered from diagnostic submissions from general practice and from hospitals to the department of gastro-intestinal infections at Statens serum Institut (SSI). SSI also makes results from the national monitoring of antimicrobial susceptibility of *Streptococcus pneumoniae* and *S. aureus* available to DANMAP. In addition, a number of regional hospital laboratories provide results of susceptibility testing. *E. coli* and coagulase negative staphylococci from diagnostic submissions. There is no routine collection of community samples from humans.

All bacterial isolates from DANMAP are stored in a strain collection, which is available for further studies and for retrospective studies to determine the occurrence of resistance to new types of antimicrobials. Human pathogens examined by the participating regional hospital laboratories and for which results are available are not included in the strain collection.

3. Susceptibility testing

Most susceptibility testing of the bacteria from food animals is carried out using broth micro dilution in Sensititre (Trek Diagnostic Systems Ltd.). However, *Campylobacter* are tested using plate dilution which is also used to test for susceptibility to a number of growth promoting antimicrobials that for technical reasons have not been included in the Sensititre panel. Isolates of *Actinobacillus* until now have been examined using tablet diffusion on Müller–Hinton agar. All susceptibility testing is carried out according to NCCLS guidelines.

Bacterial isolates from foods used to be examined using tablet diffusion. However, from 1999 onwards testing has been carried out with broth micro dilution.

Isolates from humans are tested by either tablet or disk diffusion. SSI carries out performance checking on a continuous basis to ensure that susceptibility testing done at the participating hospitals provides comparable results. In addition, SSI carries out performance testing among the three DANMAP partners.

The antimicrobial panels used include representatives of the main groups of therapeutic agents as well as antimicrobial growth promoters. A complete list of the antimicrobials included in the panel may be found in the DANMAP 98 report [6].

4. Data handling

The susceptibility data for all bacteria from animals and foods and those for zoonotic bacteria from humans

are stored in databases as continuous variables with all identifiers.

The results for food animals are stored in an ORACLE database. They are accessed and analysed using SAS version 6.12. The database also includes information about samples from slaughter animals that have been analysed for a bacterial species but with a negative result. This provides useful denominator information that may be used to estimate the prevalence of specific resistance profiles in the animal population. Information is also available about the herd of origin, identified by an official farm-ID number. This ID-number enables additional information about the farm to be accessed such as the number of animals and the animal species present on the farm as well as the geographical co-ordinates (GIS) of the farm. Additional information in the database include the name of the animal and the bacteria species, including subtype, as well as the results of susceptibility testing.

For food isolates, the data includes details about the food analysed in addition to information about bacterial species and the results of susceptibility testing. The data are stored in a PARADOX database and analysed using EXCEL spreadsheets.

5. Recording consumption of antimicrobials

Phenotypic expressions of resistance may occur as a result of co-selection by unrelated compounds because of resistance genes that are linked or be due to cross-resistance to related antimicrobial agents. Therefore, resistance monitoring alone does not provide the information required for well-targeted intervention. Focused intervention to control antimicrobial resistance must be based on detailed information about the use of antimicrobials, in addition to resistance data. Therefore, an important part of the DANMAP programme is the monitoring of antibiotic consumption.

5.1. Human medicine

In Denmark, all antibiotics are prescription-only medicines distributed exclusively through pharmacies. Physicians do not sell antibiotics but will issue prescriptions for redemption at a pharmacy. All medical formulations are available only in defined pack sizes, each of which is identified by an official numerical code. This code identifies precisely the active ingredients of the medicine, the strength and the size of the pack, as well as other items of interest such as the identity of the person receiving treatment and the identity of the doctor issuing the prescription.

The Danish Medicines Agency (DMA) is responsible for monitoring the consumption of all medicines in Denmark. The agency has established a recording sys-

tem where the processing of all prescriptions at all pharmacies is logged electronically. The records are transmitted to the DMA on a monthly basis and collated into a statistic describing the consumption in defined daily dosages (DDD) per 1000 persons. These statistics are made available for use in the DANMAP reports.

5.2. Veterinary medicine

As for human medicine all antibiotics for treatment of animals are prescription-only medicines, sold exclusively through pharmacies. Veterinarians must obtain medicines for use in practice from a pharmacy. They can legally sell the medicines to farmers however, their profit is by law limited to 5% which is deemed sufficient to cover expenses. Consequently, about 80% of all medicines used in veterinary practice are sold on prescriptions, which are redeemed by farmers at a pharmacy.

Until now, recording the use of antibiotics for treatment of animals has been based on compulsory reporting to the DMA by importers and manufacturers of the quantities sold to wholesalers and pharmacies. This means that the figures provide an estimate only of the quantities actually consumed in herds and there is no information on the consumption in the individual animal species.

The Danish Plant Directorate monitors the consumption of coccidiostats and antimicrobial growth promoters. These agents can be sold to feed mills only in the form of premixes. Producers of such premixes must be licensed and are required to report to the Plant Directorate the quantities of active ingredients used.

It is a result of this dual reporting system for therapeutics and growth promoters that DANMAP is able to discriminate between consumption of compounds licensed for both of these two separate purposes. Examples include tylosin, which is used as a growth promoter and for the treatment of pigs and monensin, which is licensed for use as growth promoter in cattle and as a coccidiostat in poultry.

5.3. VETSTAT

VETSTAT is a programme to carry out a continuous detailed monitoring of the use of all prescription medicine in animals, in addition to coccidiostats and growth promoters, at the individual herd level. The programme is now being planned in detail and will become effective during the year 2000. The programme utilises the infrastructure used for recording consumption of human medicines, but in addition, veterinarians will be required by law to report their sale of medicines to a central database. Among the information recorded by VETSTAT is the identity of the farm receiving the

medicine (using the farm-ID number described above), the species of animal and the age group for which the medicine is intended as well as the reason for prescribing. The data will be collated to form a national statistic on the consumption of veterinary medicine. The results will be used for detailed studies on the association between antimicrobial consumption and occurrence of resistance but will also be used to form a basis for advice to veterinarians and farmers about prudent use of antimicrobials.

6. Discussion

DANMAP forms an integrated programme for monitoring trends in antimicrobial resistance in bacteria from animals, foods and humans allows us to compare occurrence of resistance for each circumstance in the chain. DANMAP also forms a foundation for specific research projects, for example determination of the genetic basis for resistance profiles of particular interest.

Overall, DANMAP has confirmed the association between the occurrence of resistance and the quantities of antimicrobials used. However, DANMAP has also shown the importance of co-selection on the occurrence of specific types of resistance. Furthermore, results indicate that non-antimicrobial mediated transmission of resistant subtypes within a bacterial genus may play an important role. So far, DANMAP has not provided the data necessary to quantify the role of antibiotics in the transmission of animal pathogens however, the merging of results from the DANMAP and the VET-STAT programmes is expected substantially to extend the range of knowledge.

Results of the DANMAP programme are published annually. The reports may be accessed at the Zoonosis Centre homepage on the internet at, www.svs.dk.

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