2004

NORM
NORM-VET

**Usage of Antimicrobial
Agents and Occurrence of
Antimicrobial Resistance
in Norway**

CONTRIBUTORS AND PARTICIPANTS

Editors:

Hilde Kruse	NORM-VET, National Veterinary Institute
Gunnar Skov Simonsen	NORM, Dept. of Microbiology, University Hospital of North Norway and Norw. Inst. of Public Health

Authors:

Hege Salvesen Blix	Norwegian Institute of Public Health
Kari Grave	VETLIS / Norwegian School of Veterinary Science
Merete Hofshagen	NORM-VET, National Veterinary Institute
Bjørn Iversen	Norwegian Institute of Public Health
Hilde Kløvstad	Norwegian Institute of Public Health
Hilde Kruse	NORM-VET, National Veterinary Institute
Jørgen Lassen	Norwegian Institute of Public Health
Turid Mannsåker	Norwegian Institute of Public Health
Madelaine Norström	NORM-VET, National Veterinary Institute
Gunnar Skov Simonsen	NORM, Dept. of Microbiology, University Hospital of North Norway and Norw. Inst. of Public Health
Martin Steinbakk	NORM, Dept. of Microbiology, University Hospital of North Norway and Akershus Univ. Hospital
Marianne Sunde	National Veterinary Institute

Institutions participating in NORM-VET:

Norwegian Food Safety Authority	
National Veterinary Institute, Norwegian Zoonosis Centre	Madelaine Norström / Hilde Kruse / Merete Hofshagen
National Veterinary Institute, Section of Bacteriology	Hanne Tharaldsen / Marianne Sunde
Norwegian Institute of Public Health	Jørgen Lassen / Trine-Lise Stavnes

Institutions participating in NORM:

Aker University Hospital, Department of Bacteriology	Signe H. Ringertz / Bitten Rasmussen
Akershus University Hospital, Department of Microbiology	Martin Steinbakk / Siri Haug
Bærum Hospital, Central Laboratory, Section of Microbiology	Pål A. Jenum / Merriam Sundberg
Central Hospital of Buskerud, Department of Microbiology	Hjørdis Iveland / Ellen Grimstad
Central Hospital of Nordland, Department of Microbiology	Liisa Mortensen / Hege Elisabeth Larsen
Central Hospital of Nord-Trøndelag, Levanger, Department of Microbiology	Arne Mehl / Anne-Kristine Lorås
Central Hospital of Oppland, Lillehammer, Department of Microbiology	Einar Aandahl / Viggo Hasseltvedt / Kari Ødegaard
Central Hospital of Hedmark, Elverum, Laboratory of Microbiology	Linda Schildman / Lene Bjøntegård
Central Hospital of Østfold, Department of Microbiology	Eivind Ragnhildstveit / Anne Cathrine Hollekim
Stavanger University Hospital, Department of Microbiology	Elisebet Haarr / Tone Roa
Central Hospital of Sogn og Fjordane, Department of Microbiology	Reidar Hjetland / Astrid Vedde
Central Hospital of Vest-Agder, Department of Microbiology	Sølvi Noraas / Torill S. Larsen
Central Hospital of Vestfold, Department of Microbiology	Rolf Schøyen / Astrid Lia
Central Hospital of Hordaland, Haugesund, Department of Microbiology	Liv J. Sønsteby / Pirrko-Liisa Kellokumpu
County Hospital of Møre og Romsdal, Molde, Department of Microbiology	Einar Vik / Jytte Scheby
County Hospital of Møre og Romsdal, Ålesund, Department of Microbiology	Reidar Hide / Anna Haugen
Haukeland Univ. Hospital, Department of Immunology and Microbiology	Asbjørn Digranes / Torunn Sneide Haukeland
National Hospital, University of Oslo, Institute of Medical Microbiology	Fredrik Müller / Magli Bøvre
National Cancer Hospital, Laboratory of Microbiology	Jørgen Lassen / Inchis Engelstad / Merete R. Ueland
Norwegian Institute of Public Health, Laboratory of Enteric Pathogens	Jørgen Lassen / Trine-Lise Stavnes
Telelab A/S, Skien	Yngvar Tveten / Monica Kollstrøm
Ullevål University Hospital, Department of Microbiology	Gaute Syversen / Thea Bergheim
University Hospital of North Norway, Department of Microbiology	Gunnar Skov Simonsen / Siv-Heidi Barkhald
University Hospital of Trondheim, Department of Microbiology	Trond Jacobsen / Sissel Kristiansen Stubø
Laboratory of Clinical Medicine, Oslo	Heidi Svanevik / Wibeke Aasnæs

NORM reference group in 2004:

Inger Sofie Samdal Vik	Norwegian Institute of Public Health
Fredrik Müller	National Hospital, University of Oslo, Institute of Medical Microbiology
Astrid Lia	Central Hospital of Vestfold, Department of Microbiology
Olav Natås	Central Hospital of Rogaland, Department of Microbiology
Arne Broch Brantsæter	Norwegian Institute of Public Health
Elisabeth von der Lippe	Ullevål University Hospital, Department of Infectious Diseases
Mark Fagan	Froland Community Health Center

CONTENTS

I. INTRODUCTION

Antimicrobial resistance is an emerging problem worldwide. It reduces the effectiveness of antimicrobial treatment of infectious diseases in humans and animals thereby leading to increased morbidity and mortality, as well as higher costs. It is well established that there is a strong association between the usage of antimicrobial agents and the occurrence of resistance. The selective pressure exerted by antimicrobial usage is a key issue in the epidemiology of resistance. Moreover, resistance can be disseminated through the spread of resistant pathogenic bacteria themselves or by horizontal transfer of resistance genes from one type of bacteria to another. Such transfer is not limited to closely related bacteria; it can also take place between bacteria of different evolutionary origins and/or ecological sources. Thus, antimicrobial usage and resistance in one compartment can have consequences for the occurrence of resistance in another compartment. When addressing antimicrobial resistance – the occurrences, causes, consequences and preventive measures – a holistic approach is needed, encompassing both usage and resistance in human and veterinary medicine, as well as in food production.

Several countries have implemented monitoring programmes for antimicrobial resistance and antimicrobial usage in recent years. Some programmes focus primarily on human usage and resistance in human pathogens, whereas others also include data concerning veterinary medicine and food production. The EU supported this broad approach in 1998 through the Copenhagen recommendations and at later follow-up conferences in Sweden, Belgium, Luxembourg and Italy. The World Health Organization (WHO), the Food and Agriculture Organization (FAO) and the World Animal Health Organization (OIE) have through several expert consultations emphasised the importance of monitoring antimicrobial usage and resistance in both human and veterinary medicine and have published several reports and recommendations in this regard.

In response to the growing concern about antimicrobial resistance, the Norwegian Ministry of Health and Social Affairs issued a national action plan against antimicrobial resistance (2000–2004) in March 2000. Again, the importance of monitoring both the human and veterinary sectors, including food production, was emphasized. The action plan recognized the need for ongoing surveillance as a fundamental component of the strategy for containment of antimicrobial resistance. The NORM and NORM-VET programmes were consequently established in order to provide and present microbiologically and epidemiologically valid data on the occurrence and distribution of antimicrobial resistance over time. The national action plan formally expired by the end of 2004. However, a conference organized in September 2004 by the Norwegian Institute of Public Health and supported by the Norwegian government issued a report which will form the basis for containment of antimicrobial resistance in the years to come. The need for surveillance of both resistance and drug usage was again emphasized.

The NORM surveillance programme for antimicrobial resistance in human pathogens was established in 1999 and is coordinated by the Department of Microbiology at the University Hospital of North Norway in Tromsø. The NORM-VET monitoring programme for antimicrobial resistance in the veterinary and food production sectors was established in 2000 and is coordinated by the Norwegian Zoonosis Centre at the National Veterinary Institute. The usage of antimicrobial agents in humans and animals is based on wholesalers' data reported to the Norwegian Institute of Public Health. This reporting was made mandatory from January 1, 2002. Data on the usage of feed additives, including antimicrobial and coccidiostatic growth promoters, are collated at the Norwegian Food Safety Authority.

This report, which is the fifth annual joint report from NORM and NORM-VET, presents data for 2004. In addition to resistance data, the NORM/NORM-VET reports present data on the usage of antimicrobial agents in humans and animals in Norway. The present report, together with earlier reports and reports from the years to come, form a basis for the detection, interpretation and evaluation of trends regarding antimicrobial usage and occurrence of resistance in Norway. The NORM and NORM-VET programmes are valuable tools for setting policies, assessing risks and evaluating interventions.

The editors would like to thank the Reference Center for Detection of Antimicrobial Resistance in Tromsø for fruitful cooperation and all those who contributed to data collection and the writing of this report for excellent work.

Tromsø / Oslo, September 2005

II. SAMMENDRAG

Dette er den femte felles rapporten fra Norsk overvåkings-system for antibiotikaresistens hos mikrober (NORM) og Norsk overvåkingsprogram for antibiotikaresistens hos fôr, dyr og næringsmidler (NORM-VET). Rapporten presenterer data over forekomst av antibiotikaresistens og forbruk av antibiotika til mennesker og dyr i 2004. Data fra relevante prosjekter som ikke er en del av de kontinuerlige overvåkingsprogrammene er også presentert. Både NORM og NORM-VET programmene er en del av regjeringens tiltaksplan mot antibiotikaresistens som ble offentliggjort i 2000. NORM koordineres av Mikrobiologisk avdeling, Universitetssykehuset i Tromsø og NORM-VET koordineres av Norsk zoonosesenter ved Veterinærinstituttet, Oslo. De to programmene har et godt samarbeid og utgir en felles årsrapport.

Forbruk av antibiotika til dyr

Forbruket av antibiotika i norsk husdyrproduksjon og akvakultur er lavt. Totalsalget av antibiotika til terapeutisk bruk på landdyr i 2004 var 5 752 kg. Fra 1995 til 2001 ble salget av veterinære antibiotika til landdyr redusert med 40%. Etter dette har forbruket holdt seg konstant på noenlunde samme nivå. Forbruksmønsteret har i denne perioden utviklet seg mer og mer i gunstig retning, det vil si at andelen penicillinbruk har økt. Penicilliner utgjorde 42% av salget av veterinære antibiotika til landdyr i 2004, og av dette var 83% beta-laktamase følsomme penicilliner. Forbruket av tetracykliner utgjorde kun 3%. Nedgangen i antibiotikaforbruket og endringene i forskrivnings-mønsteret skyldes først og fremst at husdyrnæringen i andre halvdel av 1990-tallet gjennomførte systematiske kampanjer for å redusere forbruket av antibiotika til dyr samt kampanjer for riktig bruk av antibiotika.

Totalsalget av veterinære antibiotika til terapeutisk bruk til oppdrettsfisk i Norge var i 2004 på 1159 kg aktiv substans. Kinoloner utgjorde 90% av dette salget. Forbruket av antibiotika i oppdrettsnæringen har blitt redusert med 98% de siste 18 årene. Denne betydelige reduksjonen kan tilskrives innføringen av effektive vaksiner samt sykdomsforebyggende tiltak, herunder bedrede miljøforhold.

Avoparcin ble brukt som antibakteriell vekstfremmer i norsk broiler- og kalkunproduksjon fra 1986 inntil det ble forbudt i 1995. Samme år innførte husdyrnæringene et selvpålagt forbud mot bruk av alle antibakterielle vekstfremmere. Koksidiostatika som fôrtilsetningsstoff brukes fremdeles i norsk fjørfeproduksjon. Salgstallene, i kg aktiv substans, er noe høyere enn før forbudet mot bruk av antibakterielle vekstfremmere, noe som kan forklares ved økt produksjon av broilere. Forbruksmønstret for koksidiostatika er vesentlig endret siden 1996, fra monensin til narasin, som nå utgjør hovedparten av forbruket av de ionofore koksidiostatika.

Forbruk av antibiotika hos mennesker

Totalsalget av antibiotika til systemisk bruk hos mennesker var i 2004 17,2 DDD/1000 innbyggere/dag. Det samlede forbruket har vært forholdsvis stabilt gjennom mange år, men det har skjedd en gradvis forskyvning mellom de ulike undergruppene. Salget av penicilliner er stabilt mens forbruket av kinoloner øker og tetracyklinforbruket synker.

I 2004 utgjorde penicilliner 43% av det totale antibiotika-forbruket i Norge. Det har skjedd en forskyvning mot bredspektrede penicilliner samtidig med at bruken av penicillinase følsomme penicilliner sank med 3%. Undergruppene av bredspektrede og penicillinase stabile penicilliner økte med henholdsvis 3% og 7%. Tetracykliner utgjorde 17% av totalforbruket og har sunket jevnt siden 1993. Makrolider utgjorde 11% av totalforbruket i 2004. Makrolidforbruket økte med 24% i 2001 og 2002 sammenliknet med 2000. Dette kan ha sammenheng med utbrudd av kikhoste i Norge i denne perioden. Salget av cefalosporiner, monobaktamer og karbapenemer var begrenset til 3,6% av totalsalget, men er økende. Det har gjennom de senere år skjedd en overgang til tredje generasjons cefalosporiner og karbapenemer. Det har vært en markant økning i forbruket av fluorokinoloner. Denne gruppen utgjorde kun 3% av totalforbruket i 2004, men dette er en økning på 60% siden 1998.

Forbruket av antibiotika utenfor sykehus utgjorde 92% av totalforbruket i 2004. Penicilliner utgjorde 47% av forbruket i sykehus og 40% i allmenpraksis. De mest brukte gruppene i allmenpraksis utenom penicilliner var tetracykliner (18%) og makrolider og linkosamider (11%). På sykehus var cefalosporiner den vanligste gruppen nest etter penicilliner (16%), etterfulgt av metronidazol (6%) og kinoloner (6%).

I Norge kan antibiotika kun utleveres mot resept. Fra 1 januar 2004 har det vært etablert en nasjonal reseptdatabase. Data fra dette registeret vil bli gjort tilgjengelige i løpet av 2005.

Resistens hos kliniske isolater fra dyr

De kliniske isolatene inkludert i 2004 var fra diagnostiske prøver fra førstegangs hud- og øreinfeksjoner hos hund (*Staphylococcus intermedius*) og fra sepsis hos fjørfe og enteritt eller ødemsyke hos svin (pathogene *Escherichia coli*).

Forekomsten av resistens blant *S. intermedius* fra hud- og øreinfeksjoner hos hund var relativt høy. Kun 18,3% av isolatene var følsomme for alle antibiotika som inngikk i undersøkelsen. Resistens ble hyppigst observert for følgende antibiotika; penicillin (70%), fusidinsyre (50%) og oxytetracyklin (42%). Forekomsten av resistens blant sykdomsfremkallende *E. coli* fra svin og fjørfe var moderat. Blant svineisolatene ble resistens hyppigst påvist mot streptomycin (47%) og oxytetracyklin (24%), mens blant fjørfeisolatene dominerte fluorokinolonresistens. Til tross for et begrenset prøveantall, er det indikasjoner på en økende fluorokinolonresistens (26%) hos patogene *E. coli* fra fjørfe sammenlignet med NORM/NORM-VET 2002; 2%.

Resistens hos indikatorbakterier

Forekomsten av ervervet antibiotikaresistens blant bakterier som utgjør den normale tarmfloraen, kan tjene som en indikator på selektivt antibiotikapress i ulike populasjoner. I NORM-VET benyttes *Escherichia coli* og *Enterococcus* spp. som indikatorbakterier. I 2004 ble indikatorbakterier fra hund, svin og broiler inkludert.

Forekomsten av resistens blant *E. coli* og *Enterococcus* spp. i 2004 var moderat og i et internasjonalt perspektiv relativt lavt. Totalt var 85,3%, 67,6% og 61,2% av *E. coli*

isolatene fra hund, svin og broilere følsomme mot alle antibiotika som inngikk i undersøkelsen. Hyppigst ble resistens observert mot de antibiotika som brukes terapeutisk hos disse dyrene; streptomycin, sulfa, oxytetracyklin og ampicillin.

Andelen enterokokker som var resistente mot minst ett antibiotikum varierte fra 27,3% til 94% avhengig av den spesifikke *Enterococcus* spp. (*E. faecalis, E. faecium*), dyreart og prøvetype. Resistens mot narasin ble hyppigst observert fra broilere, mens oxytetracyklinresistens ble hyppigst observert blant isolatene fra hund og svin.

Resistens hos zoonosebakterier og andre enteropatogene bakterier

Salmonella spp., med unntak av *S. enterica* subsp. *diarizonae* fra sau, påvises sjelden hos matproduserende dyr i Norge. I 2004 ble kun fem tilfeller av *S.* Typhimurium-infeksjon (to storfe og tre svin) og ett tilfelle av *S.* Senftenberg infeksjon (fjørfe) påvist i det nasjonale overvåkingsprogrammet. Disse isolatene var følsomme for alle antibiotika som inngikk. De andre isolatene som ble resistenstestet (n=42) var fra diagnostiske innsendelser fra kjæledyr, vilt, ville fugler og reptiler. I den siste kategorien var det kun ett isolat, *S.* Newport fra en slange (dyrehage), som var resistent mot både enrofloxacin og nalidixinsyre. Resultatene indikerer at resistens ikke er utbredt blant *Salmonella*-isolater som en sjelden gang blir isolert fra norske dyr.

Av de humane salmonellose-tilfellene som ble rapportert i 2004, var 72% oppgitt å ha blitt smittet i utlandet. Andelen *S.* Typhimurium isolater som var følsomme for alle antibiotika, var høyere for kategorien "smittet i Norge" (79,5%) enn for kategorien "smittet i utlandet" (48,3%). Multiresistens (resistens mot mer enn to antibiotika) ble hyppigere påvist i sistnevnte kategori (24,7%) enn førstnevnte (14,1%). En signifikant forskjell mellom disse kategoriene ble observert i forhold til kinolonresistens; 24% av isolatene fra pasienter smittet utenlands var resistente mot nalidixinsyre mot 2,6% av isolatene fra personer smittet innenlands. Ingen av isolatene, uavhengig av smittested, viste nedsatt følsomhet for ciprofloxacin.

Andelen *S.* Enteritidis-isolater resistente mot de ulike antibiotika inkludert var lavere enn for *S.* Typhimurium bortsett fra resistens mot nalidixinsyre. Totalt var 26,6% av *S.* Enteritidis isolatene resistente mot nalidixinsyre. Kun 0,1% var resistente mot ciprofloxacin, mens en redusert følsomhet mot ciprofloxacin var observert hos 1,6% av isolatene. Forekomsten av resistens hos *S.* Typhimurium og *S.* Enteritidis i NORM/NORM-VET 2004 er noenlunde tilsvarende som tidligere år. Dataene antyder likevel at isolater fra importerte tilfeller er hyppigere multiresistente og at nalidixinsyreresistens er økende sammenlignet med i NORM/NORM-VET 2003.

Resultatene fra 2004 viser at forekomsten av resistens hos *Campylobacter jejuni* fra norske broilere fremdeles er lav og stabil. Totalt 93,5% av isolatene var følsomme for alle antibiotika som inngikk i undersøkelsen. Totalt var 6,5% resistente mot kun ett antibiotikum, enten ampicillin eller oxytetracyklin. Ingen fluorokinolonresistens ble påvist. Nivået av resistens og resistensmønstrene for *C. jejuni* fra norske broilere samsvarer med *C. jejuni* fra mennesker smittet i Norge med unntak av en høyere forekomst av kinolonresistens, blant humanisolatene. Dette forholdet ble

også påvist i NORM/NORM-VET 2001, 2002 og 2003. Resistens var betydelig mer utbredt blant *C. jejuni* fra pasienter smittet i utlandet (80,5% resistente mot minst ett antibiotikum) enn pasienter smittet i Norge (11,5%). Fluorokinolonresistens var mer vanlig blant isolater fra pasienter smittet utenlands enn fra innenlands smittede; 68,8% mot 9,6%.

Forekomsten av resistens blant *Yersinia enterocolitica* isolater er fortsatt lav, men har økt sammenlignet med det som er blitt observert i tidligere rapporter og kan for en stor del forklares med en økt andel av infeksjoner ervervet utenlands. Spesielt kan resistens mot nalidixinsyre forklares av forekomsten av resistens blant de utenlands smittede, hvor 9,1% av isolatene var resistente mot nalidixinsyre, mens ingen kinolonresistens ble observert blant isolater fra personer smittet innenlands.

De aller fleste *Shigella*-isolatene var fra pasienter smittet utenlands. I likhet med hva som rapporteres fra andre land, var resistens utbredt.

Resistens hos kliniske isolater fra mennesker

Forekomsten av antibiotikaresistens i kliniske isolater fra mennesker i Norge var fortsatt meget lav i 2004, og det ble kun påvist mindre endringer fra 2003 til 2004. Fire av 660 *Staphylococcus aureus* isolater fra blodkultur (0,6%) og seks av 1 136 *S. aureus* isolater fra sår (0,5%) ble verifisert som methicillinresistente (MRSA) ved *mecA* og *nuc* PCR. Den lave forekomsten av MRSA ble bekreftet av det nyetablerte meldesystemet for systemiske *S. aureus* isolater fra laboratorienes rutinediagnostikk (8/1088, 0,7%). Meldesystemet for infeksjonssykdommer (MSIS) registrerte 221 tilfeller av MRSA infeksjoner i 2004 hvilket er praktisk talt uendret fra 216 tilfeller i 2003. Det gjenstår å se om dette utgjør et brudd med den gradvise økningen i MRSA tilfeller over de siste åtte år. Det må bemerkes at antallet meldinger av sepsis med MRSA ble doblet slik at den økende overvekten av lokaliserte infeksjoner ble korrigert. Forhåpentligvis vil vedvarende tilbakeholdenhet med forskrivning av antibiotika og økende fokus på smitteverntiltak innenfor og utenfor sykehus hindre videre spredning av MRSA i Norge. Fra 2005 er både MRSA kolonisering og infeksjon meldepliktig til MSIS. Sett i sammenheng med bedre molekylær karakterisering av bakterieisolatene på nasjonal basis vil den forbedrede overvåkingen utgjøre et viktig verktøy i arbeidet med å opprettholde Norge som lavprevalens-område. Forekomsten av resistens mot andre antibiotikagrupper enn beta-laktamer i *S. aureus* var uendret. Det vesentligste funnet var 25,0% resistens mot fusidin mot 20,8% i 2001 og 23,0% i 2003.

Blodkulturisolater av *E. coli* og *Klebsiella* spp. var som tidligere generelt følsomme for bredspektrede antibiotika inkludert cefotaxim, ceftazidim, cefpirom, meropenem, ciprofloxacin og piperacillin/tazobactam. Det ble imidlertid påvist en økning i nedsatt følsomhet for gentamicin fra 0,7% i 2003 til 1,4% i 2004. Dette må overvåkes i fremtiden da en slik resistensutvikling kan true det tradisjonelle aminoglykosid baserte regimet som ofte brukes ved sepsis i Norge. Syv av 982 *E. coli* isolater (0,7%) og to av 359 *Klebsiella* spp. isolater (0,6%) produserte bredspektrede beta-laktamaser (ESBL). Selv om forekomsten fortsatt er lav gir disse ESBL-produserende Gram-negative stavbakteriene grunn til

bekymring, og det første sykehusutbruddet med en slik stamme i Norge er allerede beskrevet. Forekomsten av resistens mot kinoloner var fortsatt lav men fortsatte å øke i 2004. Ved bruk av de reviderte brytningspunktene fra 2005 økte forekomsten av nedsatt følsomhet og resistens mot ciprofloxacin fra 2,4% til 3,3% i *E. coli* og fra 1,0% til 2,5% i *Klebsiella* spp. Den nedsatte følsomheten for kinoloner i disse bakterieartene gjenspeiler økningen i forbruket av ciprofloxacin fra 0,29 DDD/1000 innbyggere/ dag i 2000 til 0,47 DDD/1000 innbyggere/dag i 2004.

Det ble ikke påvist klinisk signifikant vankomycinresistens i enterokokker i 2004. Forekomsten av nedsatt følsomhet for ampicillin økte imidlertid til 74,5% i *Enterococcus faecium*, og høygradig gentamicin-resistens økte dramatisk i både i *E. faecalis* (22,0%) og *E. faecium* (19,6%). Det mest bekymringsverdige er at 17,6% av systemiske *E. faecium* isolater nå er resistente mot begge komponentene i det tradisjonelle beta-laktam/ aminoglykosid kombinasjonsregimet som ofte brukes ved sepsis på norske sykehus

Streptococcus pneumoniae fra blodkulturer var generelt følsomme for alle relevante antibiotika. Det ble imidlertid påvist nedsatt følsomhet for penicillin G i 12 av 628 isolater (2,0%) hvilket er en økning fra de to foregående år. Tre av disse isolatene hadde også nedsatt følsomhet for cefalosporiner og var høygradig resistente mot makrolider og tetracykliner. Forekomsten av nedsatt følsomhet for erytromycin fortsatte å øke fra 6,0% i 2003 til 9,7% i 2004.

Haemophilus influenzae fra luftveisprøver ble første gang overvåket i 2000. Forekomsten av beta-laktamase produksjon økte svakt fra 7,1% i 2000 og 7,0% i 2001 til 8,8% i 2004. Det ble også påvist en økning av beta-laktamase negative, ampicillin resistente *H. influenzae* (BLNAR) fra 1,2% i 2000 og 1,3% i 2001 til 3,1% i 2004 slik at den samlede forekomsten av ampicillinresistens var 11,9% i 2004. Prevalensen av nedsatt følsomhet for trimetoprim/sulfametoxazol økte fra 9% i 2000 og 7% i 2001 til 18,7% i 2004 ved bruk av samme brytnings-punkter.

Beta-hemolytiske streptokokker gruppe A (*S. pyogenes*) fra sår og luftveisprøver ble første gang undersøkt i NORM i 2002. Alle isolatene fra 2004 var fullt følsomme

for penicillin G. Forekomsten av tetracyklinresistens var litt lavere i 2004 (11,7%) enn i 2002 (15,6%). Dette kan ha sammenheng med det synkende forbruket av tetracykliner i norsk allmenpraksis. Som i 2002 var forekomsten av resistens høyere i sårprøver (15,1%) enn i luftveisprøver (8,0%). Forekomsten av erytromycin-resistens sank fra 3,8% i 2002 til 2,0% i 2004 hvilket er overraskende sett i et internasjonalt perspektiv.

Forekomsten av resistens i *E. coli* fra urin var i det vesentlige uendret bortsett fra enkelte ESBL-positive isolater (2/1101, 0,2%) og en svak økning av nedsatt følsomhet for ciprofloxacin fra 2,6% i 2003 til 3,7% i 2004.

I alt 302 tilfeller av tuberkulose ble meldt til MSIS i 2004. 282 av dem hadde ikke tidligere blitt behandlet med tuberkulostatika, og resistensbestemmelse ble utført på 234 *Mycobacterium tuberculosis* isolater fra disse pasientene. Til sammen fire isolater ble klassifisert som multiresistente; to isolater med smittested Afrika og to isolater med smittested Europa utenom Norge. Resistensbestemmelse ble også gjort på *M. tuberculosis* isolater fra 20 pasienter som tidligere var behandlet for tuberkulose. Ett isolat fra en asiatisk pasient var monoresistent mot rifampicin, og en pasient født i Norge hadde et isolat som var monoresistent mot streptomycin.

Konklusjon
Antibiotikaresistens er fortsatt et begrenset problem i Norge både når det gjelder mennesker og dyr. Det lave forbruket av antibiotika og det fordelaktige forbruksmønsteret må opprettholdes for å bevare den gunstige situasjonen. Resultatene som presenteres i denne rapporten, viser at norske strategier når det gjelder antibiotikabruk og antibiotikaresistens hittil har vært vellykkede både i husdyrholdet og i helsevesenet. Faren for økende resistensproblemer er imidlertid til stede i form av økt antibiotikaforbruk i Norge og import av resistens fra andre land. Det er derfor nødvendig med fortsatt aktiv innsats for å sikre at vi også i fremtiden kan gi tilfredsstillende antibiotikabehandling til dem som trenger det. NORM/NORM-VET-rapporten er et viktig bidrag til arbeidet med å forebygge utvikling og spredning av antibiotikaresistens.

III. SUMMARY

This is the fifth joint report from the NORM surveillance programme for antimicrobial resistance in human pathogens and the NORM-VET monitoring programme for antimicrobial resistance in the veterinary and food production sectors. The report presents data on the occurrence of antimicrobial resistance and the usage of antimicrobial agents in humans and animals for the year 2004. Data from specific surveys and projects that are not part of the continuous monitoring through NORM or NORM-VET are also presented. The NORM and NORM-VET programmes are parts of the Norwegian Government's Action Plan against Antimicrobial Resistance issued in 2000. NORM is coordinated by the Department of Microbiology, University Hospital of North Norway, Tromsø. NORM-VET is coordinated by the Norwegian Zoonosis Centre, National Veterinary Institute, Oslo. Successful collaboration has been established between NORM and NORM-VET and a joint report is issued annually.

Usage of antimicrobial agents in animals

The usage of antimicrobials in Norwegian animal production and aquaculture is low. In 2004, the total sale of antimicrobial drugs approved for therapeutic use in animals (excluding fish) in Norway was 5752 kg. The annual usage of veterinary antimicrobial drugs decreased gradually by 40% from 1995 to 2001, and has thereafter remained stable. The patterns of use have gradually become more favourable as the proportion of penicillin use has increased. In 2004, the proportion accounted for by penicillins was 42% of which beta-lactamase sensitive penicillins constituted 83%. The usage of tetracyclines amounted to only 3% of the total usage. The reduced antimicrobial use as well as the favourable prescribing patterns is mainly explained by a successful campaign on prudent use of antimicrobials completed by the Norwegian husbandry organizations during the second part of the 1990s.

In 2004, the total sale of antimicrobial drugs for therapeutic use in farmed fish was 1159 kg of active substance. Quinolones accounted for 90% of this. The usage of antimicrobials in aquaculture has decreased by 98% during the last 18 years, which is mainly attributed to the introduction of effective vaccines as well as improved health management.

The antimicrobial growth promoter avoparcin was used in Norwegian broiler and turkey production from 1986 and until it was prohibited in 1995. The same year, the Norwegian food animal production industries voluntarily abandoned the use of all antimicrobial growth promoters.

In 2004, the total sale of coccidiostatic feed additives, in kilograms of active substance, was slightly higher than before the ban on antimicrobial growth promoters was implemented. This is explained by increased production of broilers. While monensin was the most frequently used ionophore in poultry in 1995, the usage of coccidiostats is now dominated by narasin.

Usage of antimicrobial agents in humans

In 2004, the overall sales of antibacterials for systemic use in humans represented 17.2 DDD/1000 inhabitants/day. Total sales of antibacterials have remained relatively unchanged for many years. However, within subgroups of antibacterials, usage trends have changed. The penicillin group is stable; the subgroup of quinolones is steadily increasing, while the subgroup of tetracyclines is decreasing.

In 2004, the penicillins represented 43% of the total antimicrobial usage in Norway. There has been a shift towards more broad-spectered penicillins while beta-lactamase sensitive penicillins is decreasing - by 3% in 2004. The subgroups of penicillins with extended specter and beta-lactamase resistant penicillins increased by 3% and 7%, respectively. Tetracyclines represented 17% of total usage and has decreased steadily since 1993. Macrolides represented 11% of total use in 2004. An increase of 24% was found in 2001 and 2002 as compared to 2000. This may have been due to epidemics of e.g. pertussis within the Norwegian population. The sales of cephalosporins, monobactams and carbapenems, although limited, have also increased, now representing 3.6% of the total sales of antibacterials. Over the years, there has been a shift towards 3rd generation cephalosporins and carbapenems. There has been a marked increase in quinolone use. Quinolones represented only 3% of total antibacterial sales in 2004, but this still accounts for a 60% increase since 1998.

The use of antibacterials outside hospitals represented 92% of the total human sales of antimicrobials in 2004. Penicillins represented around 47% and 40% of the use in hospitals and in ambulatory care, respectively. The most important other groups in ambulatory care are tetracyclins (18%) and macrolides and lincosamides (11%). In hospitals, the cephalosporins (16%) was the most used group after the penicillins, followed by metronidazole (6%) and the quinolones (6%).

In Norway, antibiotics are prescription-only drugs. From 1 January 2004, a national prescription database has been established. Data from this register will be available in 2005.

Resistance in clinical isolates from animals

The clinical isolates included in 2004 were from diagnostic samples from "first time" skin infections including otitis externa in dogs *(Staphylococcus intermedius)* and from septicaemia in poultry, enteritis and oedema disease in swine (pathogenic *Escherichia coli*).

The prevalence of resistance in *S. intermedius* from dogs was relatively high. In total, only 18.3% of the isolates were susceptible to all the antimicrobials included in the monitoring. Resistance was most frequently observed for the following antimicrobials; penicillin 70%, fusidic acid 50% and oxytetracycline 42%.

The prevalences of resistance in pathogenic *E. coli* from swine and poultry were moderate. Resistance was most commonly observed to streptomycin (47%) and oxytetracycline (24%) in the isolates from swine. Resistance to fluoroquinolones accounted for the main proportion of the resistance in the isolates from poultry. Although the sample size was limited, there are indications of an increased occurrence of fluoroquinolone resistance in clinical isolates from broilers (26%) as compared to previous years (NORM/NORM-VET 2002; 2%).

Resistance in indicator bacteria

The prevalence of acquired antimicrobial resistance among certain bacteria of the normal enteric microflora can serve as an indicator of the selective antimicrobial pressure in the various populations. In NORM-VET, *Escherichia coli* and *Enterococcus* spp. serve as indicator bacteria. In 2004, indicator bacteria from dogs, swine and broilers were included.

The occurrences of resistances in *E. coli* and *Enterococcus* spp. in 2004 were moderate, and relatively low in an international perspective. In total, 85.3%, 67.6% and 61.2% of the *E. coli* isolates from dogs, swine and broilers, respectively were susceptible to all antimicrobial agents included in the monitoring. Resistance was most commonly observed to antimicrobial agents commonly used for therapeutic treatment; streptomycin, sulfamethoxazole, oxytetracycline and ampicillin. Fluoroquinolone resistance was rarely observed.

The proportion of *Enterococcus* spp. resistant to at least one antimicrobial agent varied from 27.3% to 94%, depending on the particular *Enterococcus* spp. (*E. faecalis, E. faecium*), the animal species and the sample types included. Resistance to narasin was most frequently observed in isolates from broilers, whereas resistance to oxytetracycline was most commonly observed in the isolates from dogs and swine.

Resistance in zoonotic and non-zoonotic enteropathogenic bacteria

Salmonella spp., apart from *S. enterica* subsp. *diarizonae* in sheep, is rarely isolated from food producing animals in Norway. In 2004, only five cases of *S.* Typhimurium-infection (two cattle and three pigs) and one isolate of *S.* Senftenberg-infection (poultry) were detected in the national surveillance programme. The other *Salmonella* isolates (n=42) were from the surveillance programme or from diagnostic submission from pets, wild animals, wild birds and reptiles. Only one isolate, *S.* Newport from a snake (from a zoo), was resistant to enrofloxacin and nalidixid acid. All other isolates were susceptible to all antimicrobials included in the monitoring. The data, although very limited, indicate that antimicrobial resistance is not very widespread among those *Salmonella* that sometimes are isolated from Norwegian animals.

In 2004, 72% of the human cases of salmonellosis were reported as being infected abroad. The proportion of *S.* Typhimurium isolates susceptible to all antimicrobial agents was higher for the category "infected in Norway" (79.5%) than for the "infected abroad" category (48.3%). Moreover, multiresistance (resistance to more than two antimicrobial agents) was more common in the category "infected abroad" (24.7%) as compared to the category "infected in Norway" (14.1%). A significant discrepancy for the two categories was observed for quinolones; in the category "infected abroad", 24.0% of the isolates were resistant to nalidixic acid as opposed to 2.6% among those from patients "infected in Norway". None of the isolates, regardless of origin, showed reduced susceptibility to ciprofloxacin. The proportion of *S.* Enteritidis isolates resistant to the different antimicrobial agents included in the monitoring was considerably lower than for *S.* Typhimurium except for nalidixic acid. In total, 26.6% of the isolates of *S.* Enteritidis were resistant to nalidixid acid. Only 0.1% were resistant to ciprofloxacin, whereas

reduced susceptibility to ciprofloxacin was observed in 1.6% of the isolates. The resistance frequencies observed for *S.* Typhimurium and *S.* Enteritidis in 2004 are quite similar to those observed in previous reports. There may be some indications that imported cases of *S.* Typhimurium may more often be multiresistant and that resistance to nalidixic acid is increasing as compared to 2003.

The results obtained in 2004 show that the prevalence of resistance in *Campylobacter jejuni* from Norwegian broilers is still low and stable. A total of 93.5% of the isolates were susceptible to all antimicrobials included in the monitoring. Altogether 6.5% were resistant to only one antimicrobial (ampicillin or oxytetracycline). No fluoroquinolone resistance was observed. The level of resistance and the resistance patterns for *C. jejuni* isolated from Norwegian broilers correspond quite well with what was observed for *C. jejuni* isolated from humans infected within Norway, except for a higher prevalence of resistance to quinolones among the human isolates. This relationship was also observed in 2001, 2002 and 2003. Resistance was significantly more widespread in the *C. jejuni* isolates derived from patients infected abroad (80.5% resistant to at least one antimicrobial) than patients infected in Norway (11.5%). Fluoroquinolone resistance was more common in isolates from infections acquired abroad than from domestic cases; 68.8% versus 9.6%.

The occurrence of resistance in isolates of *Yersinia enterocolitica* is still low, but has increased compared to what has been observed in previous reports and is explained by a higher proportion of infections acquired abroad than in earlier years. In particular, the resistance to nalidixic acid was explained by infections acquired abroad; 9.1% of these isolates were resistant to nalidixic acid, as opposed to none of the isolates from domestic cases.

The vast majority of the *Shigella* isolates tested originated from patients infected abroad. As is the case in reports from other countries, resistance was widespread.

Resistance in clinical isolates from humans

The prevalence of antimicrobial resistance in human clinical isolates was still very low in Norway in 2004, and only minor changes were observed from 2003 to 2004. Four of 660 *Staphylococcus aureus* blood culture isolates (0.6%) and six of 1136 *S. aureus* wound isolates (0.5%) were verified as methicillin resistant (MRSA) by *mecA* and *nuc* PCRs. The low prevalence of MRSA was confirmed by the newly established notification of all systemic *S. aureus* infections from routine laboratory data (8/1088, 0.7%). The Norwegian Surveillance System for Communicable Diseases (MSIS) registered 221 cases of MRSA infections in 2004 which was practically unchanged from the 216 cases in 2003. It remains to be seen whether this represents a lasting change of the upward trend from the last eight years. It is interesting to note that the number of MRSA septicaemias was doubled in 2004, thus correcting the steadily increasing proportion of MRSA isolates recovered from wounds and soft tissue infections. Hopefully, the continuing prudence of prescribers and increasing focus on infection control in hospitals and nursing homes will succeed in preventing

further spread of MRSA in Norway. From 2005, both clinical cases and MRSA colonization will be notifiable to MSIS. In conjunction with intensified molecular characterization of isolates on a national basis, this improvement of the MRSA surveillance will provide important tools for the continuing efforts to maintain Norway as a low-prevalence area. The resistance to non beta-lactam antimicrobials was essentially unchanged in *S. aureus* with 25.0% resistance to fusidic acid in wound isolates (20.8% in 2001 and 23.0% in 2003) as the most important finding.

Blood culture isolates of *E. coli* and *Klebsiella* spp. were, as in previous years, generally susceptible to broad-spectrum antimicrobials including cefotaxime, ceftazidime, cefpirome, meropenem, ciprofloxacin and piperacillin/tazobactam. However, it should be noted that the prevalence of non-susceptibility to gentamicin in *E. coli* increased from 0.7% in 2003 to 1.4% in 2004. This issue will be closely watched as it may threaten the traditional aminoglycoside-based empirical treatment regimens often used for septicaemia in Norway. Seven out of 982 *E. coli* isolates (0.7%) and two out of 359 *Klebsiella* spp. isolates (0.6%) produced extended spectrum beta-lactamases (ESBL). Although the numbers are small, the appearance of ESBL in Gram-negative enterics is a cause for great concern, and the first hospital outbreak of such a strain in Norway has already been documented. The prevalence of intermediate susceptibility and resistance to quinolones was still low but increased further in 2004. By use of the revised 2005 breakpoints, the overall prevalence of ciprofloxacin non-susceptibility increased from 2.4% to 3.3% in *E. coli* and from 1.0% to 2.5% in *Klebsiella* spp. The reduced susceptibility to quinolones in these species parallels the increasing use of ciprofloxacin from 0.29 DDD/1,000 inhabitants/day in 2000 to 0.47 DDD/1000 inhabitants/day in 2004.

Clinically significant vancomycin resistance was not detected in enterococci in 2004. However, the prevalence of non-susceptibility to ampicillin has now reached 74.5% in *Enterococcus faecium*, and high-level resistance to gentamicin is dramatically increasing in both *E. faecalis* (22.0%) and *E. faecium* (19.6%). Most worrisome, 17.6% of systemic *E. faecium* isolates are now resistant to both components of the traditional beta-lactam/aminoglycoside combination regimen used for empirical treatment of septicaemiae in Norwegian hospitals.

Streptococcus pneumoniae from blood cultures were generally susceptible to all relevant antimicrobials. However, 12 of 628 blood culture isolates (2.0%) displayed reduced susceptibility to penicillin G which is an increase from the two previous years. Furthermore, three of these isolates also displayed varying levels of resistance to cephalosporins and all were highly resistant to erythromycin and doxycycline. The prevalence of non-susceptibility to erythromycin continued to increase from 6.0% in 2003 to 9.7% in 2004.

Haemophilus influenzae isolates from respiratory tract specimens were surveyed for the first time in 2000. The prevalence of beta-lactamase production increased slightly from 7.1% in 2000 and 7.0% in 2001 to 8.8% in 2004. Furthermore, the prevalence of beta-lactamase negative *H. influenzae* with reduced susceptibility to ampicillin

(BLNAR) increased from 1.2% in 2000 and 1.3% in 2001 to 3.1% in 2004. In total, 11.9% of the isolates were non-susceptible to ampicillin. Non-susceptibility to trimethoprim/sulfamethoxazole increased from 9% in 2000 and 7% in 2001 to 18.7% in 2004 when comparable breakpoints were used.

Group A streptococci (*S. pyogenes*) from wounds and the respiratory tract were first introduced into the NORM programme in 2002. All isolates from 2004 were fully susceptible to penicillin G. The overall prevalence of resistance to doxycycline was slightly lower in 2004 (11.7%) than in 2002 (15.6%). This may be related to the decreasing usage of tetracyclines among out-patients in Norway. As in 2002, resistance was more widespread among isolates originating from wounds (15.1%) than among respiratory tract isolates (8.0%). Remarkably, the prevalence of erythromycin resistance decreased from 3.8% in 2002 to 2.0% in 2004.

The results for *E. coli* isolates from the urinary tract remained essentially unchanged from previous years except for occasional ESBL isolates (2/1101, 0.2%) and a slight increase in non-susceptibility to ciprofloxacin from 2.6% in 2003 to 3.7% in 2004.

A total of 302 cases of tuberculosis were reported to MSIS in 2004. 282 of them had not previously been treated with antituberculosis drugs, and susceptibility tests were performed on 234 *Mycobacterium tuberculosis* isolates from these patients. A total of four isolates were classified as multidrug resistant. Two of the isolates originated from Africa whereas the remaining two were from Europe outside Norway. Susceptibility tests were also performed on *Mycobacterium tuberculosis* isolates from 20 patients who had previously received antituberculosis drug treatment. One isolate from an Asian patient was monoresistant to rifampicin and one Norwegian born patient had an isolate monoresistant to streptomycin.

Conclusion

Antimicrobial resistance is still a limited problem in Norway. The relatively low usage of antimicrobial agents as well as the advantageous patterns of use must be maintained to preserve this favourable situation. The data presented in this report show that Norwegian antimicrobial policies in food production and healthcare have succeeded. However, the situation may rapidly change if the use of antimicrobial agents in Norway increases or if resistant clones are imported from abroad. A continued effort is needed to prevent the development and spread of antimicrobial resistance and thus ensure the effectiveness of antimicrobials when such treatment is needed. The NORM/NORM-VET report is a vital component of the work aimed at preventing the development and spread of antimicrobial resistance.

IV. POPULATION STATISTICS

Population statistics for human and animal populations are presented in order to facilitate comparison of Norwegian data with corresponding figures from other countries. The data are collected by Norwegian authorities as shown in the various tables below.

TABLE 1. Human population in Norway as of January 1, 2005.
(Data provided by Statistics Norway).

Age group	All	Males	Females
0 to 4 years	289 146	147 752	141 394
5 to 14 years	620 172	318 491	301 681
15 to 24 years	562 949	287 410	275539
25 to 44 years	1 315 455	667 462	647 993
45 to 64 years	1 140 921	577 566	563 355
65 years and older	677 720	285 389	392 331
All age groups	4 606 363	2 284 070	2 322 293

TABLE 2. Livestock population in Norway and the number of slaughtered animals in 2004.

Animal category	Herds	Number of Animals[*]	Slaughtered animals[*]
Cattle	22.500[1]	936.600[1]	334.100[2]
Dairy cow**	15.677[1]	253.200[1]	-
Suckling cow**	3.793[1]	44.700[1]	-
Combined production (cow)**	934[1]	25.200[1]	-
Goat	1.090[1]	71.000[1]	18.400[2]
Dairy goat**	568[1]	44.650[1]	-
Sheep	-	2.412.700[1]	1.264.200[2]
Breeding sheep > 1 year**	17.439[1]	918.500[1]	-
Swine	3.762[1]	800.400[1]	1.469.200[2]
Breeding animal > 6 months**	2.199[1]	61.800[1]	-
Fattening pigs for slaughter	3.344[1]	424.100[1]	-
Poultry			-
Egg laying hen (> 20 weeks of age)	2.650[1]	3.432.100[1]	2.469.200[2]
Flocks > 250 birds**	916[1]	-	-
Broiler	489[2]	-	42.851.700[2]
Turkey, ducks and geese for slaughter	191[1]	365.800[1]	1.035.200[2]
Flocks > 25 birds**	67[1]	-	-
Ostrich	18[1]	192[1]	-

Data from: [1)] Register of Production Subsidies as of July 31, 2004; [2)] Register of Slaughtered Animals; [3)] Directorate of Fisheries, amount in metric tons, ungutted fish, preliminary data.
* Numbers > 100 rounded to the nearest ten, numbers > 1000 rounded to the nearest hundred.
** Included in above total.

TABLE 3. Animals (excluding fish) imported to Norway in 2004. *(Data provided by KOORIMP).*

Animal species	Live animals* Individuals	Consignments	Semen Doses	Embryos
Cattle			40.000	69
Swine			200	
Goat	26	2		
Sheep	11	2	750	
Reindeer live animals for slaughter	350	2		
Fur animals	213	1		
Poultry – day old chicks	157.357	16		
Turkey – day old chicks	14.326	7		
Ducks and geese	840	2		

V. USAGE OF ANTIMICROBIAL AGENTS

A. USAGE IN ANIMALS

Antimicrobial and coccidiostatic growth promoters

Data on the usage of various substances and categories of feed additives were obtained through annual reports from the Norwegian Agricultural Inspection Service (1995-2002) and The Norwegian Food Safety Authority (2003-2004). Table 4 summarizes total sales of antimicrobial growth promoters and coccidiostat feed additives in Norway in 1995–2004.

The glycopeptide avoparcin was licensed for the Norwegian market as a growth promoter in broilers and turkeys in 1986. It was prohibited in 1995 due to a reported association between its use and the occurrence of vancomycin resistant enterococci in animal husbandry. The same year, the Norwegian food animal production industry voluntarily abandoned the use of all antimicrobial growth promoters. These measures resulted in an immediate reduction in the usage of these substances

(Table 4). In 1998, the streptogramin virginiamycin was officially prohibited due to reports from other countries of an association between its use and the occurrence of enterococci that were resistant to quinupristin-dalfopristin, a streptogramin combination preparation used in human medicine. No antimicrobial growth promoters have been used in animals in Norway since 1998.

Coccidiostats as feed additives are still used in Norwegian poultry production. The total sales of coccidiostats, in kilograms of active substance, are slightly higher than before the ban on antimicrobial growth promoters. During the same time the production of broilers has increased. However, the pattern of usage has changed (Table 4). While monensin was the most frequently used ionophore in the poultry industry in 1995, the usage of coccidiostats is now almost totally dominated by narasin.

TABLE 4. Total sales, in kilograms of active substance, of antimicrobial growth promoters and of coccidiostats as feed additives in Norway 1995-2004. Data were obtained through annual reports from the Norwegian Agricultural Inspection Service (1996-2002) and The Norwegian Food Safety Authority (2003-2004).

Active substances	Total sales in kg active substance									
	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004
Avoparcin*	419	Prohib.	Prohib.	Prohib.	Prohib.	Prohib.	Prohib.	Prohib.	Prohib.	Prohib.
Zincbacitracin	129	64	27	0	0	0	0	0	0	0
Virginiamycin	0	0	0	0	Prohib.	Prohib.	Prohib.	Prohib.	Prohib.	Prohib.
Total antimicrobial growth promoters	**548**	**64**	**27**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
Lasalocid	996	480	471	193	208	80	96	514	108	173
Monensin	3 422	891	561	485	557	776	629	521	717	817
Salinomycin	214	27	0	0	27	233	12	0	0	0
Narasin	24	3 508	3 343	3 530	4 062	4 486	4 195	4 470	5 067	5 270
Total ionophore coccidiostats	**4 656**	**4 906**	**4 375**	**4 208**	**4 854**	**5 575**	**4 932**	**5 505**	**5 892**	**6 260**
Amprolium/etopabat	156	116	582	174	201	135	159	74	42	0.8
Total other coccidiostats	**156**	**116**	**582**	**174**	**201**	**135**	**159**	**74**	**42**	**0.8**

*Prohibited since May 31, 1995.

Therapeutic usage of veterinary antimicrobial drugs

Sales data for veterinary antimicrobial agents represent sales from drug wholesalers to Norwegian pharmacies. The majority of substances included are approved as pharmaceutical formulations for food animals, including horses, and/or dogs and cats. Thus, the figures represent overall sales data for veterinary antimicrobial agents. Antimicrobial agents authorized for human use, but prescribed for animals, are not included. Such drugs are primarily used in small animal practices.

Table 5 summarizes the sales (in kg of active substance) in 2004 of veterinary antimicrobial drugs approved for therapeutic use in domestic animals in Norway. The data are organized according to the main therapeutic substance

groups (ATCvet groups) and show the total usage for the various routes of administration. The total sale of veterinary antimicrobial agents is given in Figure 1. Figure 2 illustrates the proportion of the total sale of the various groups of antimicrobial substances. Both figures present annual sales data for the period 1995–2004. In 2004, the sales of veterinary antimicrobial agents approved for therapeutic use in animals in Norway amounted to 5752 kg of active substance (Table 5). The annual usage of veterinary antimicrobial agents decreased gradually by 40% from 1995 to 2001. Since then, the annual usage has remained on a relatively constant level.

TABLE 5. Sales in 2004 (in kilograms of active substance) of veterinary antimicrobial agents approved in Norway for therapeutic use in animals (farmed fish not included, see Table 6). The data were obtained from the Norwegian Institute of Public Health and represent sales from drug wholesalers to Norwegian pharmacies.

Groups of substances	ATCvet code	Active substance or combinations of substances	Gastro-intestinal (QA07)	Uterine (QG01)	Systemic indiv. (QJ01)	Systemic herds (QJ01)	Intra-mammary (QJ51)
Tetracyclines	QG01AA07	Oxytetracycline		2	101	87	
	QJ01AA02	Doxycycline			0.1		
	QJ01AA06	Oxytetracycline					
Beta-lactams	QJ01CA04	Amoxicillin			94	108	
	QJ01CE09	Benzylpenicillinprocain*			1 986		5
	QJ01CE90/QJ51CE90	Penethamate hydroiodide*			7		
	QJ01CR02/QJ51RV01	Amoxicillin+clavulanic acid			190		9
Sulfonamides	QJ01EQ06	Sulfanilamid			22		
Sulfonamides and	QJ01EQ10	Sulfadiazine+trimethoprim**			1 126		
trimethoprim**	QJ01EQ13	Sulfadoxine+trimethoprim			121		
Lincosamides	QJ01FF01	Clindamycin			10		
	QJ01FF02	Lincomycin			5		
Aminoglycosides	QA07AA01	Neomycin	31				
	QA07AA90	Dihydrostreptomycin (DHS)	131				
Quinolones	QJ01MA90	Enrofloxacin			27		
	QJ01MA96	Ibafloxacin			1		
Others	QJ01XX92	Tiamulin			11	122	
Combinations	QG01AE99	Sulfadimidine+procaine penicillin*+DHS		206			
	QJ01RA01	Benzylpenicillinprocain* +DHS			559		766
	QJ51RC25	Penethamate hydroiodide* + DHS					25
Total per route of administration			**162**	**208**	**4 260**	**317**	**805**
Total (kg)							**5 752**

*Calculated as benzylpenicillin
**Includes small amounts of baquiloprim

The proportion accounted for by pure penicillin preparations rose from 25% in 1995 to 42% in 2004. Altogether 83% of the veterinary penicillin preparations sold in 2004 were beta-lactamase sensitive penicillins. From 1995 to 2002, the sale of sulfonamides in combination with trimethoprim or baquiloprim increased from 11% to 24% of the total sales, while this figure was 22% in both 2003 and 2004. This reduction was mainly due to a reduced sale for use in dogs and cats. The proportion of sale of the combination preparations of penicillins and aminoglycosides decreased from 35% to 23% from 1995-2004. The corresponding figure for the sulfonamides decreased from 14% in 1995 to 0.4% in 2004. The proportion accounted for by tetracyclines declined from 5% to 3% during the same period. The reduced use as well as the favourable prescribing patterns is mainly explained by a successful campaign on prudent use of antimicrobials conducted by the Norwegian husbandry organizations during the second part of the 1990s.

In Norway, premixes for farmed fish are approved by the drug authorities and classified as pharmaceutical specialities. Sales figures, in kg of active substance, of medicated feeds sold through authorized feed mills and of premixes containing antimicrobial agents sold through pharmacies are presented in Table 6.

TABLE 6. Total sales (in kilograms of active substance) of veterinary antimicrobial agents for therapeutic use in farmed fish in Norway 1995-2004. The data were obtained from the Norwegian Institute of Public Health and represent sales from drug wholesalers to Norwegian pharmacies and sales by feed mills.

Groups of substances/active substance	ATCvet code	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004
Tetracyclines											
Oxytetracycline	QJ01AA06	70	27	42	55	25	15	12	11	45	9
Amphenicols											
Florfenicol	QJ01BA90	64	64	123	135	65	148	109	205	154	111
Quinolones											
Flumequine	QJ01MB07	182	105	74	53	7	52	7	5	60	4
Oxolinic acid	QJ01MB91	2 800	841	507	436	494	470	517	998	546	1 035
Total		**3 116**	**1 037**	**746**	**679**	**591**	**685**	**645**	**1 219**	**805**	**1 159**

Altogether, 1159 kg of veterinary antimicrobial agents for therapeutic use in farmed fish were sold in 2004. The antimicrobial class most commonly used in farmed fish in Norway is quinolones. In 2004, the quinolones accounted for 90% (in kg) of the total usage of antimicrobial agents in fish. The annual usage of antimicrobial agents in farmed fish declined by 98% from 1987 to 2004. In the same period, the total production of farmed fish increased massively. This significant decrease in the usage of antimicrobial agents in Norwegian aquaculture is mainly attributed to the introduction of effective vaccines as well as to improved health management.
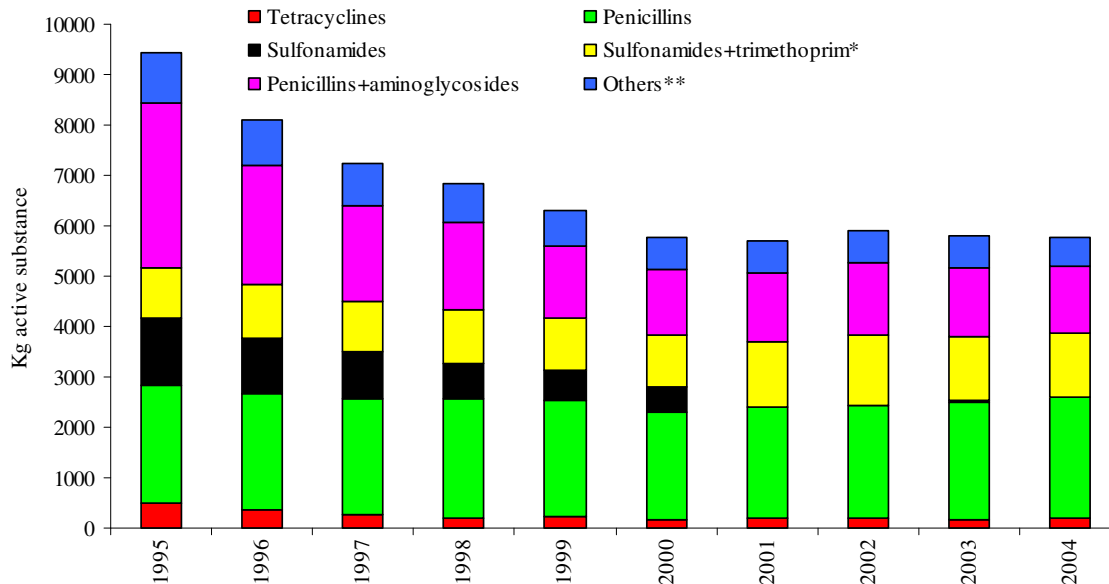
FIGURE 1. Sales (in kilograms of active substance) of veterinary antimicrobial agents (QA07AA, QG01AA, QG01AE, QJ01, QJ51) for therapeutic use in Norway in 1995–2004, fish not included.
*Includes small amounts of baquiloprim. **Includes ATCvet codes: QA07AA01; QA07AA51; QA07AA90; QG01AE99; QJ01EQ06; QJ01FA02; QJ01FF01; QJ01FF02; QJ01MA90; J01RA91; QJ01XX92.
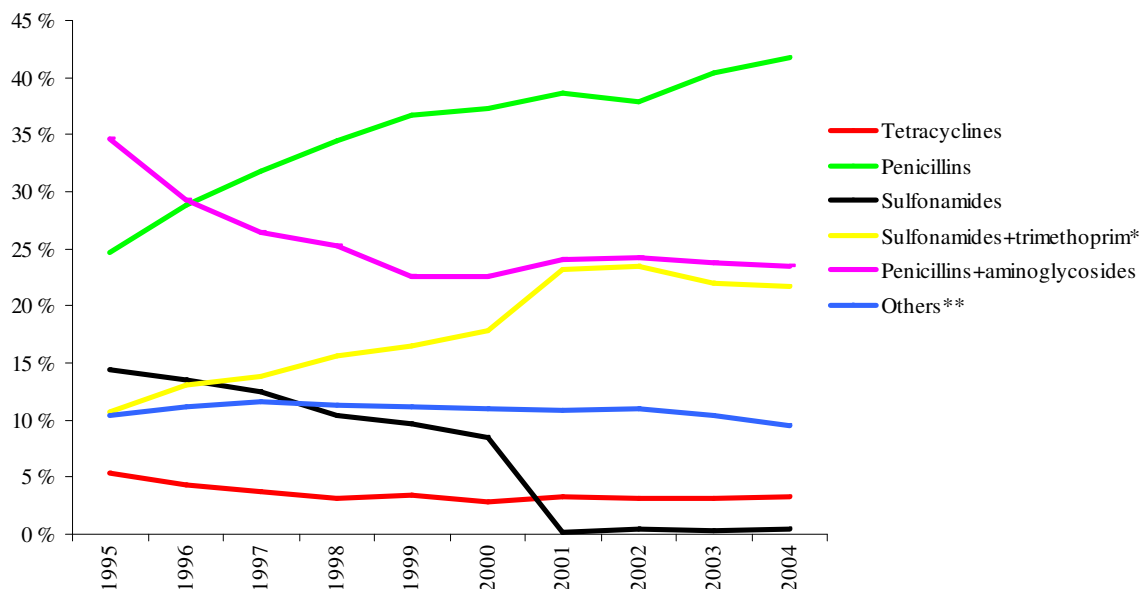
FIGURE 2. Sales (as percentage of total sales) of veterinary antimicrobial agents (QA07AA, QG01AA, QG01AE, QJ01, QJ51) in Norway in 1995–2004, fish not included.
*Includes small amounts of baquiloprim. **Includes ATCvet codes: QA07AA01; QA07AA51; QA07AA90; QG01AE99; QJ01EQ06; QJ01FA02; QJ01FF01; QJ01FF02; QJ01MA90; QJ01RA91; QJ01XX92.

B. USAGE IN HUMANS

In 2004, the overall sales of antibacterials for systemic use in humans represented 17.2 DDD/1000 inhabitants/day. Total sales of antibacterials have remained relatively unchanged for many years. However, within subgroups of antibacterials, usage trends have changed over the years. The penicillin group is stable; the subgroup of quinolones is steadily increasing, while the subgroup of tetracyclines is decreasing (Table 7, Figure 3).

TABLE 7. Human usage of antibacterial agents in Norway 1998-2004 by ATC groups. The usage is presented as DDD (Defined Daily Doses)/inhabitants/day and in % change 1998-2004. Collection of data on human usage of antimicrobial agents is presented in Appendix 2..

ATC	Groups of substances	1998	1999	2000	2001	2002	2003	2004	Change (%) 1998-2004
J01A	Tetracyclines	3.37	3.19	3.17	3.11	3.13	3.03	2.97	- 12
J01B	Amphenicols	0.004	0.005	0.004	0.003	0.002	0.002	0.001	
J01CA	Penicillins with extended spectrum	1.90	1.96	2.01	2.1	2.23	2.29	2.37	+ 25
J01CE	β-lactamase sensitive penicillins	5.12	5.01	4.66	4.68	4.48	4.38	4.23	- 17
J01CF	β-lactamase resistant penicillins	0.27	0.32	0.35	0.41	0.50	0.59	0.63	+ 133
J01CR	Combination of penicillins	0.01	0.01	0.01	0.01	0.01	0.01	0.01	
J01D	Cephalosporins, monobactams, carbapenems	0.44	0.47	0.52	0.55	0.58	0.62	0.61	+39
J01E	Sulfonamides and trimethoprim	1.34	1.26	1.17	1.16	1.15	1.08	1.09	- 19
J01F	Macrolides, lincosamides and streptogramins	1.61	1.59	1.59	1.8	1.98	1.92	1.89	+ 17
J01G	Aminoglycosides	0.05	0.05	0.04	0.06	0.06	0.07	0.06	
J01M	Quinolones	0.30	0.33	0.35	0.40	0.44	0.48	0.52	+ 60
J01X	Other antibacterials	2.2	2.34	2.39	2.55	2.57	2.63	2.83	+ 20
	Total	**16.6**	**16.6**	**16.3**	**16.8**	**17.1**	**17.1**	**17.2**	**+ 4**
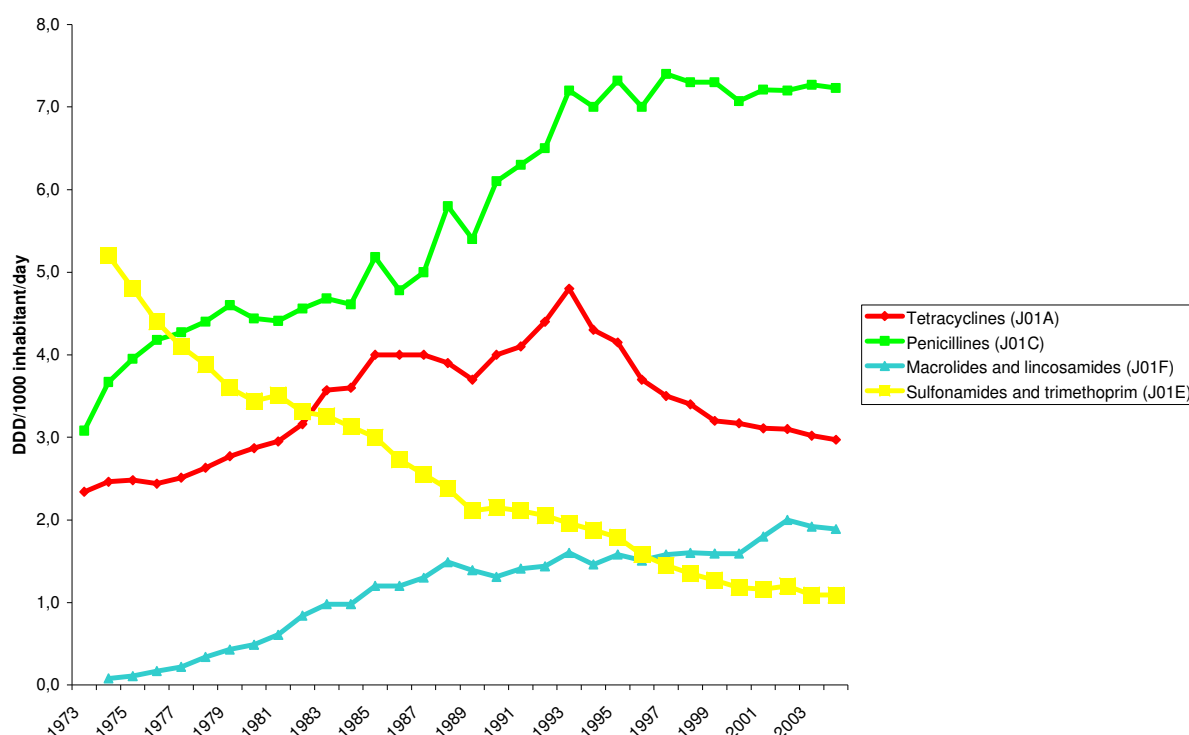
FIGURE 3. Sales of penicillins (J01C), tetracyclines (J01A), macrolides, lincosamides and streptogramines (J01F) and sulfonamides and trimethoprim (J01E) in Norway 1973-2004.

Norwegian information campaign on appropriate antibiotic use

In September 2004 the Norwegian Institute of Public Health launched the campaign "Appropriate antibiotic use – for the child's best interest".

Many children with viral infections and self-limiting bacterial infections are treated with antibiotics. This is done despite evidence-based guidelines. In Norway more than 90 % of all antibiotics are prescribed by general practitioners. Antibiotics are commonly prescribed in the treatment of upper respiratory infections in preschool children. Antibiotics are often prescribed to children as a result of parental expectations or doctors' misperceptions of these expectations. There is a need to improve the parent-doctor interaction in the consultation setting and to improve parents' knowledge on the use of antibiotics.

The objective of the campaign was to decrease the redundant use of antibiotics in treatment of upper respiratory infections in preschool children in order to prevent antibiotic resistance. The campaign consisted of information to healthcare professionals, parents and the media.

Information leaflets as an alternative to prescriptions for sore throats, otitis media, bronchitis and fever
The leaflets can be used as a tool in conjunction with the doctors' consultation. The contents emphasise the information given by the doctor during the consultation. Furthermore, the leaflets provide facts about the nature of the illnesses and why antibiotics are not effective in curing the child. Alternative methods to help the child are proposed to parents.

Information leaflet for use in maternal and child health clinics
This leaflet provides short information on different upper respiratory infections, use of antibiotics, what parents can do to help their children and information on when it is important to contact the doctor. It emphasises that antibiotics are important and efficient drugs when used correctly, but for most upper respiratory infections they have little or no effect and may cause discomfort to the child (i.e. diarrhoea, vomiting). The leaflet is made for the particular use in maternal and child health clinics, where healthcare professionals meet all children and parents shortly after birth. This approach helps parents to be better prepared in the event of their child becoming ill.

Information about the campaign was given to healthcare professionals by various routes including direct mail, The Norwegian Epidemiological Bulletin, medical journals and seminars. The public was approached through the mass media. In addition, the Norwegian Institute of Public Health has launched a new web page on antibiotic resistance www.fhi.no/smittevern. The web page provides information to the public and healthcare professionals on antibiotic use, statistics and national and international strategies to prevent antibiotic resistance. The information leaflets can be downloaded from this web page.

The response to the campaign has been positive, and healthcare professionals report that the leaflets are useful tools in improving the parent-doctor interaction. New media campaigns will be launched every autumn to endorse appropriate use of antibiotics in the treatment of upper respiratory infections in preschool children.

Hilde Kløvstad

In 2004, the penicillins (ATC group J01C) represented 43% of the total antimicrobial use in Norway (Figure 4). The sales of penicillins have been stable over years. It has, however, looking at ATC 4th levels, been a shift towards more broadspectered penicillins (Figure 5). Beta-lactamase sensitive penicillins (J01CE) are continuously decreasing - by 3% in 2004. The sales of benzylpenicillin, mainly used in hospitals, were stable hence the decrease was due to phenoxymethylpenicillin, mainly used in ambulatory care. The subgroups of penicillins with extended specter and beta-lactamase resistant penicillins are both increasing, by 3% and 7%, respectively.

The tetracyclines (J01A) represent 17% of total use. The sales have been decreasing since 1993, when the highest sale ever was registered.

The macrolides (J01FA) represents 11% of total use in 2004. The sales were fairly stable in the nineties. However, an increase of 24% was found in 2001 and 2002 (compared to 2000). The highest use was in 2002. This could be due to epidemics within the population e.g. a pertussis epidemic that reached its peak in 2001. The internal macrolide pattern has remained unchanged. Erythromycin is most frequently used, representing 54% of the subgroup. The lincosamides, in Norway represented by clindamycin, is increasing - by 82% since 1998 (Table 8).

Sales of cephalosporins, monobactams and carbapenems, although limited, have also been increasing, now representing 3.6% of the total sales of antibacterials. The internal subgroup pattern has changed since 1996 (Figure 6). 1st generation cephalosporins i.e. cefalexin and cefalotin, represents 56% of ATC group J01D. Over the years, there has been a shift towards using more 3rd generation cephalosporins and more carbapenems.

There has been a small, but stable increase in quinolone use. Still it represents only a minor fraction (3%) of total antibacterial sales, but the increase has been 60% since 1998.

The increase of ATC group J01X is mainly due to the urinary prophylactic agent methenamine, representing 14% of total antibacterial use. The sales have increased by 35% since 1998.

The usage of antibacterials varies between the 19 Norwegian counties, and the usage trends has been stable over the years, i.e. a trend for the same high-use counties and low-use counties (Figure 7)

The therapy pattern of antibacterials in hospitals differs from ambulatory care (Figure 8). The antibacterial sales in DDDs to hospitals represented, in 2004, eight percent of total sales in the country.

Penicillins (J01C) represent around 47% and 40% of the use in hospitals and in ambulatory care, respectively. The most important other groups in ambulatory care are tetracyclins, J01A (18%) and macrolides and lincosamides, J01F (11%). In hospitals cephalosporins, J01D (16%) is the most used group after the penicillins, followed by metronidazole - oral and parenteral (6%) and the quinolones, J01M (6%).

The use of antibacterials outside hospital represents 92% of the total human sale of antimicrobials. Therapy traditions in ambulatory care therefore have a great impact on the total burden of antimicrobials and consequently also on the development of bacterial resistance. Furthermore, changes towards more broad-spectered antibacterials seem to be more distinct in ambulatory care. Hence, more focus should be given to surveillance of antimicrobial use and guidance of appropriate prescription practices.

In Norway, antibiotics are prescription-only drugs. From 1 January 2004, a national prescription database has been established. Data from this register will be available in 2005. Furthermore, for overall use, the slow, but steady shift towards use of more "broad-spectered" antibacterials in Norway is of concern and deserves close surveillance.
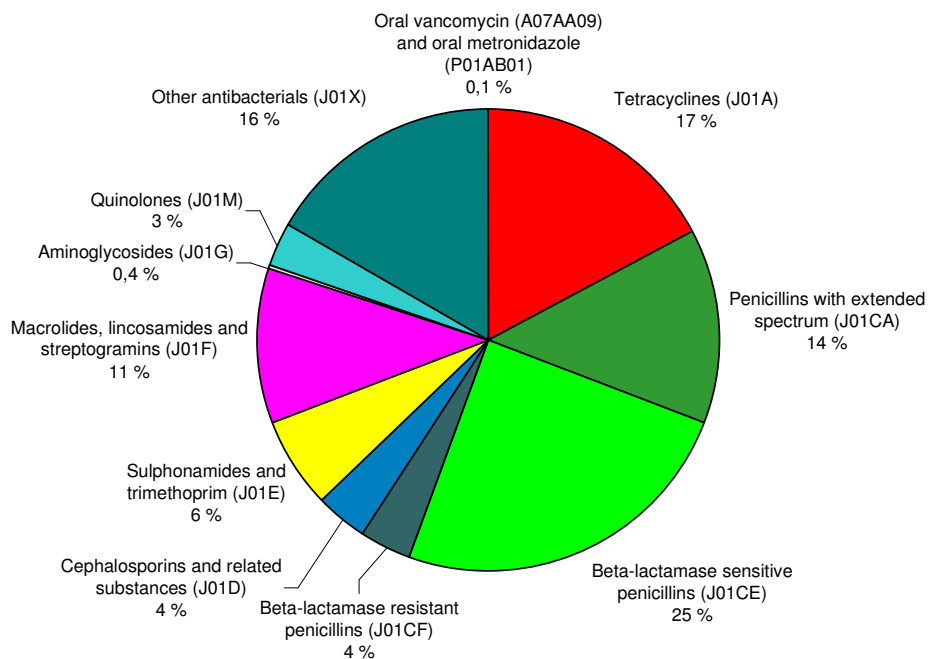
FIGURE 4. Relative amount of antimicrobial agents for systemic use in 2004 in Defined Daily Doses (DDD) (total sale in the country). Groups of antimicrobials are represented by ATC numbers.

TABLE 8. Human usage of single antimicrobial agents for systemic use in Norway (ATC group J01) 1998-2004. Sales given in DDD/1000 inhabitants/day. Collection of data on human consumption of antimicrobial agents is presented in Appendix 2.

ATC	Substance	1998	1999	2000	2001	2002	2003	2004
A07AA09	Vancomycin	0.001	0.001	0.001	0.001	0.001	0.001	0.001
J01A A02	Doxycycline	2.34	2.20	2.10	2.1	2.03	1.93	1.80
J01A A04	Lymecycline	0.09	0.09	0.14	0.19	0.26	0.30	0.34
J01A A06	Oxytetracycline	0.27	0.25	0.24	0.22	0.21	0.19	0.20
J01A A07	Tetracycline	0.67	0.65	0.69	0.64	0.62	0.60	0.62
J01B A01	Chloramphenicol	0.004	0.005	0.004	0.003	0.002	0.002	0.001
J01C A01	Ampicillin	0,09	0.09	0.09	0.08	0.09	0.1	0.1
J01C A02	Pivampicillin	0.15	0.14	0.13	0.11	0.11	0.09	0.08
J01C A04	Amoxicillin	0.85	0.87	0.83	0.89	0.94	0.95	0.94
J01C A08	Pivmecillinam	0.81	0.86	0.96	1	1.09	1.14	1.25
J01C A11	Mecillinam	0.003	0.004	0.004	0.005	0.005	0.005	0.005
J01C E01	Benzylpenicillin	0.21	0.23	0.21	0.23	0.24	0.25	0.24
J01C E02	Phenoxymethylpenicillin	4.91	4.78	4.45	4.45	4.24	4.13	3.99
J01CE08*	Benzathine benzylpenicillin	<0.0001	<0.0001	0.0001	<0.0001	0.0001	0.0001	0.0002
J01C F01	Dicloxacillin	0.19	0.22	0.25	0.31	0.39	0.48	0.51
J01C F02	Cloxacillin	0.08	0.10	0.10	0.09	0.11	0.11	0.11
J01C F05*	Flucloxacillin					0.0001	0.0002	0.0002
J01C R02	Amoxicillin and enzyme inhibitor	0.01	0.01	0.01	0.01	0.01	0.01	0.0003
J01C R05	Piperacillin and enzyme inhibitor			0.0001	0.0006	0.0014	0.0024	0.005
J01D B01	Cefalexin	0.22	0.22	0.26	0.27	0.29	0.3	0.29
J01D B03	Cefalotin	0.04	0.05	0.05	0.05	0.05	0.06	0.06
J01D C01	Cefoxitin	0.0004	0.0004	0.0004	0.0003	0.0002	0.0001	
J01D C02	Cefuroxim	0.12	0.13	0.13	0.14	0.15	0.15	0.14
J01D D01	Cefotaxim	0.03	0.04	0.04	0.05	0.05	0.07	0.07
J01D D02	Ceftazidim	0.01	0.01	0.01	0.01	0.01	0.01	0.01
J01D D04	Ceftriaxone	0.007	0.008	0.011	0.01	0.01	0.01	0.02
J01D D54	Ceftriaxone, combinations	0.0001	0.0001					
J01D F01	Aztreonam	0.0005	0.0008	0.001	0.001	0.001	0.001	0.001
J01D H02	Meropenem	0.004	0.008	0.012	0.014	0.017	0.02	0.02
J01D H51	Imipenem and enzyme inhibitor	0.007	0.006	0.006	0.005	0.005	0.006	0.005
J01E A01	Trimethoprim	0.87	0.84	0.79	0.8	0.8	0.74	0.76
J01E B02	Sulfamethizole	0.0002	0.001	0.002	0.002	0.0001		
J01E C20	Sulfonamides, combinations	0.003	0.0004					
J01E E01	Sulfamethoxazol and trimethoprim	0.47	0.42	0.38	0.36	0.36	0.34	0.34
J01F A01	Erythromycin	1.06	1.01	1.00	1.13	1.2	1.09	1.03
J01F A02	Spiramycin	0.04	0.03	0.02	0.02	0.02	0.02	0.01
J01F A09	Clarithromycin	0.24	0.26	0.26	0.3	0.36	0.37	0.37
J01F A10	Azithromycin	0.17	0.18	0.19	0.21	0.24	0.26	0.28
J01FA15	Telithromycin					0.0001	0.0003	0.0003
J01F F01	Clindamycin	0.11	0.11	0.12	0.14	0.16	0.19	0.20
J01GA01*	Streptomycin					0.0015	0.0004	0.0004
J01G B01	Tobramycin	0.03	0.03	0.02	0.03	0.04	0.04	0.03
J01G B03	Gentamicin	0.006	0.006	0.006	0.008	0.02	0.03	0.03
J01G B06*	Amikacin					0.0009	0.0008	0.0003
J01G B07	Netilmicin	0.02	0.02	0.02	0.02	0.007		
J01M A01	Ofloxacin	0.06	0.06	0.05	0.05	0.05	0.05	0.05
J01M A02	Ciprofloxacin	0.23	0.26	0.29	0.34	0.38	0.42	0.47

ATC	Substance	1998	1999	2000	2001	2002	2003	2004
J01MA12*	Levofloxacin					0.001	0.0003	
J01M B02	Nalidixic acid	0.01	0.01	0.01	0.01			
J01X A01	Vancomycin	0.005	0.004	0.005	0.005	0.006	0.006	0.007
J01X A02	Teicoplanin	0.001	0.0007	0.0012	0.0013	0.0013	0.0009	0.0007
J01X B01	Colistin	0.003	0.003	0.003	0.003	0.003	0.002	0.002
J01X C01	Fusidic acid	0.003	0.003	0.003	0.01	0.01	0.007	0.008
J01X D01	Metronidazole	0.06	0.06	0.06	0.07	0.07	0.07	0.08
J01X E01	Nitrofurantoin	0.38	0.37	0.37	0.36	0.35	0.35	0.36
J01X X05	Methenamin	1.75	1.91	1.95	2.08	2.13	2.18	2.37
J01XX08	Linezolid					0.002	0.004	0.006
P01AB01	Metronidazole	0.18	0.18	0.18	0.18	0.19	0.19	0.20
J04AB**	Rifampicin	-	0.052	0.046	0.054	0.043	0.049	0,068

* Drugs not licensed at the Norwegian marked
** Given as the amount of Rifampicin in plain and combination products. Data for 1998 is not available.

FIGURE 5. Sales of penicillins (J01C) in Norway 1996-2004 and changes within groups of penicillins.
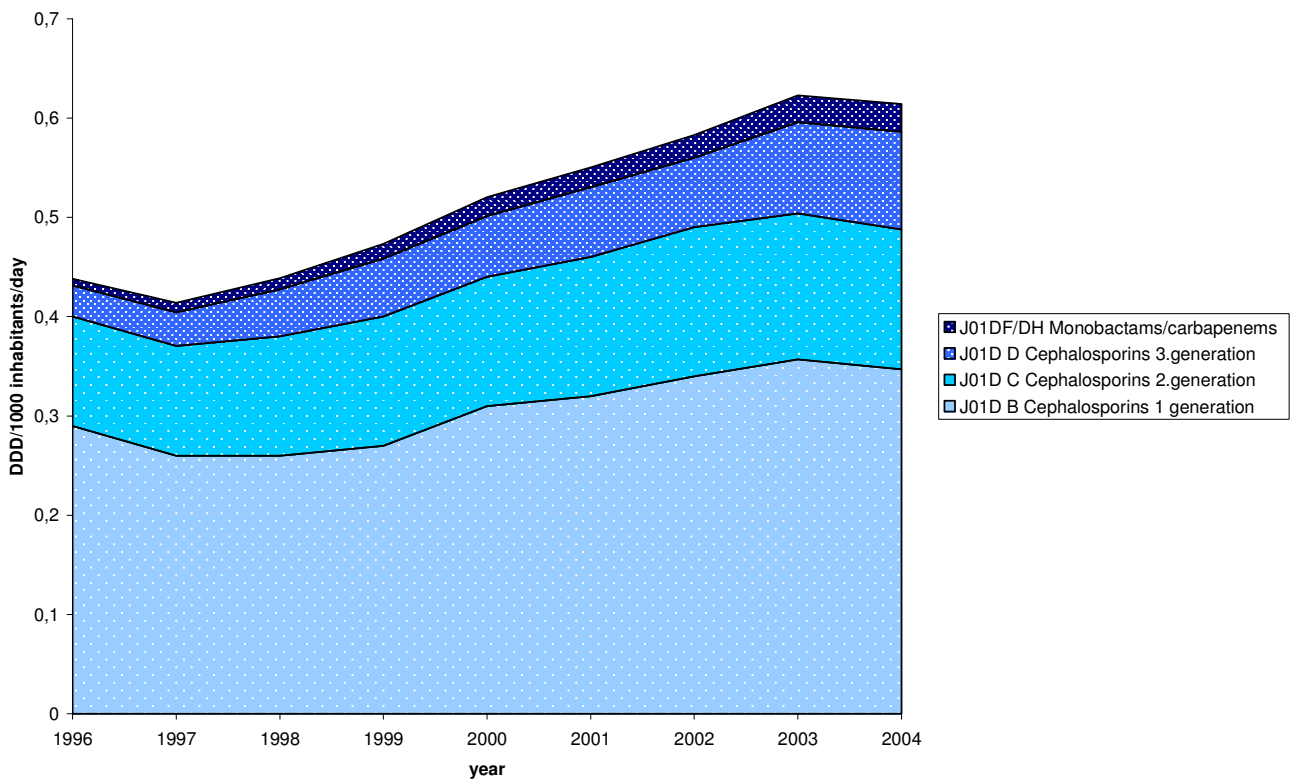
FIGURE 6. Sales of cephalosporins, monobactams and carbapenems (J01D) in Norway 1996-2004 and changes between generations of cephalosporins and monobactams/carbapenems.
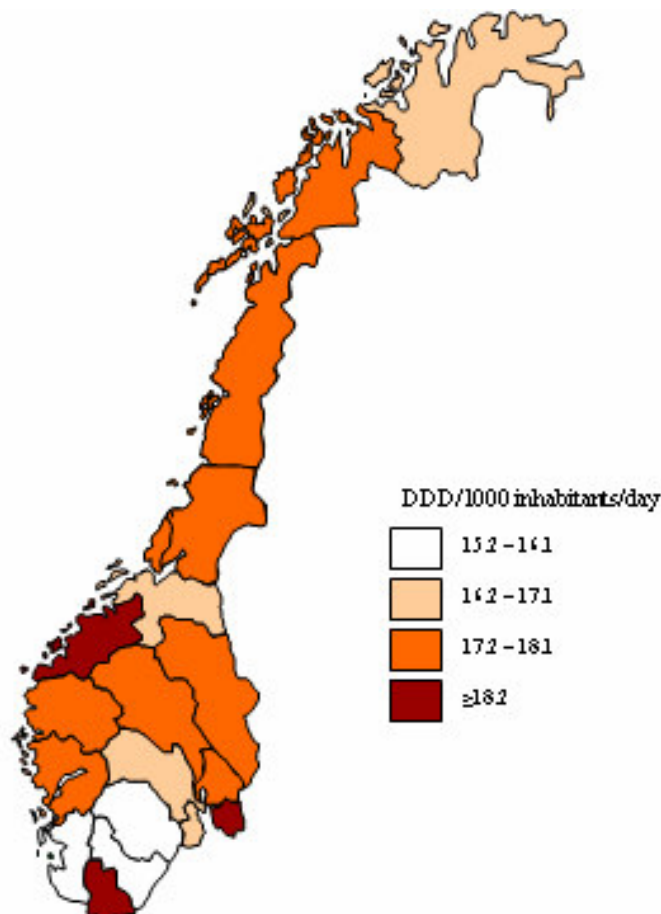
FIGURE 7. Sales of antibacterial agents for systemic use in the different counties of Norway in 2004.
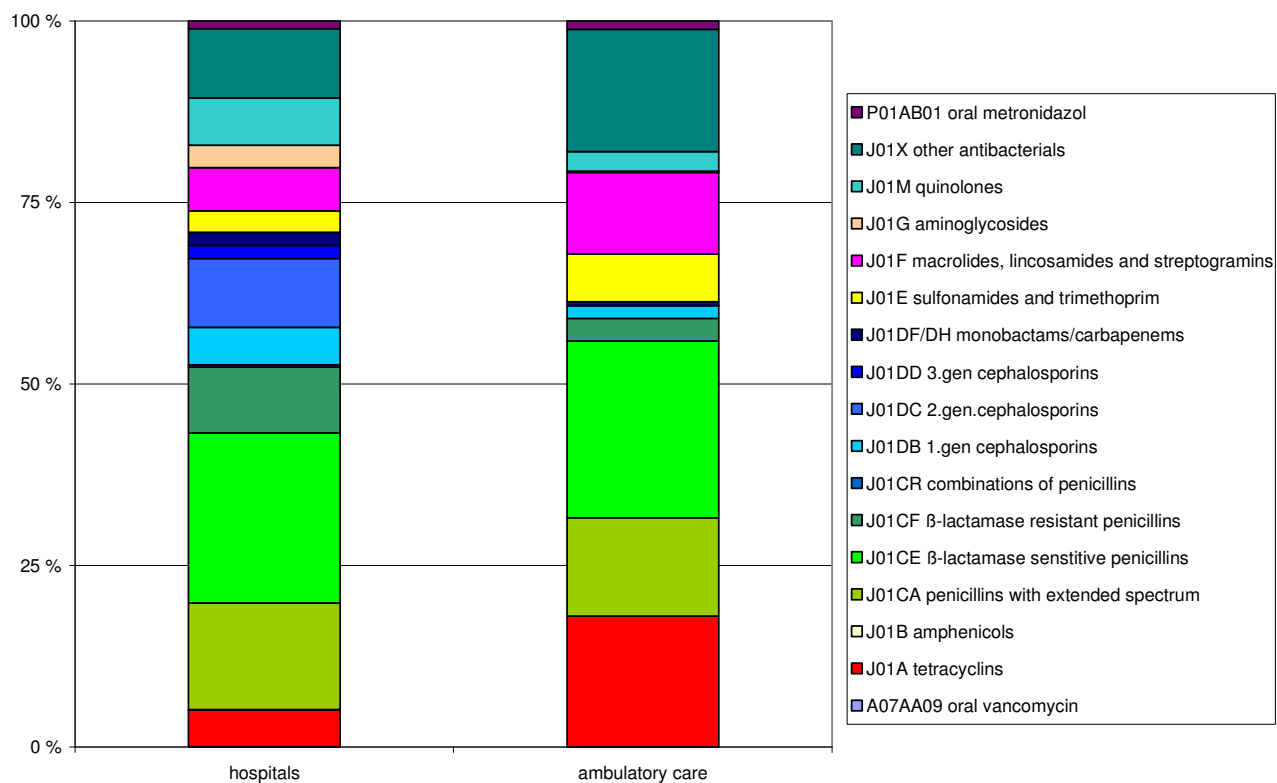
FIGURE 8. Proportions of antimicrobial agents for systemic use in Norway 2004 measured in DDD. Shown as total use, in general practice and in hospitals.

Usage of antibacterials in Norwegian hospitals

Surveillance of antibacterial usage has been performed locally in Norwegian hospitals for many years. However, comparison between hospitals has not been done. The aim of this study was to compare usage of antibacterial agents in hospitals and to explore whether hospital size measured in number of bed-days has an impact on use. Hospital use of antibacterial agents in thirteen hospitals in 1998 and 1999 was investigated. These hospitals are representative for Norwegian hospitals by including emergency, referral and university hospitals from all five health regions in the country.

Total annual use varied significantly, from 38.2 to 63.5 DDD/100 bed-days, see Figure 1. Differences in use probably reflect local therapy traditions rather than differences in patient population, indications and resistance patterns. A small increase in total use, only significant for the larger hospitals, was observed from 1998 to 1999. Therapy profile is approximately similar all over the country. Beta-lactamase sensitive penicillins (ATC group J01CE) were most frequently used. Small hospitals used more penicillins with extended specter and the magnitude of quinolone use was positively correlated to size of hospital. Large hospitals used significantly more 3rd generation cephalosporins.

Size of hospital does not influence the level of antibacterial use in Norwegian hospitals. However, therapeutic choice of antibacterial agents differs and seems to be influenced by hospital size.
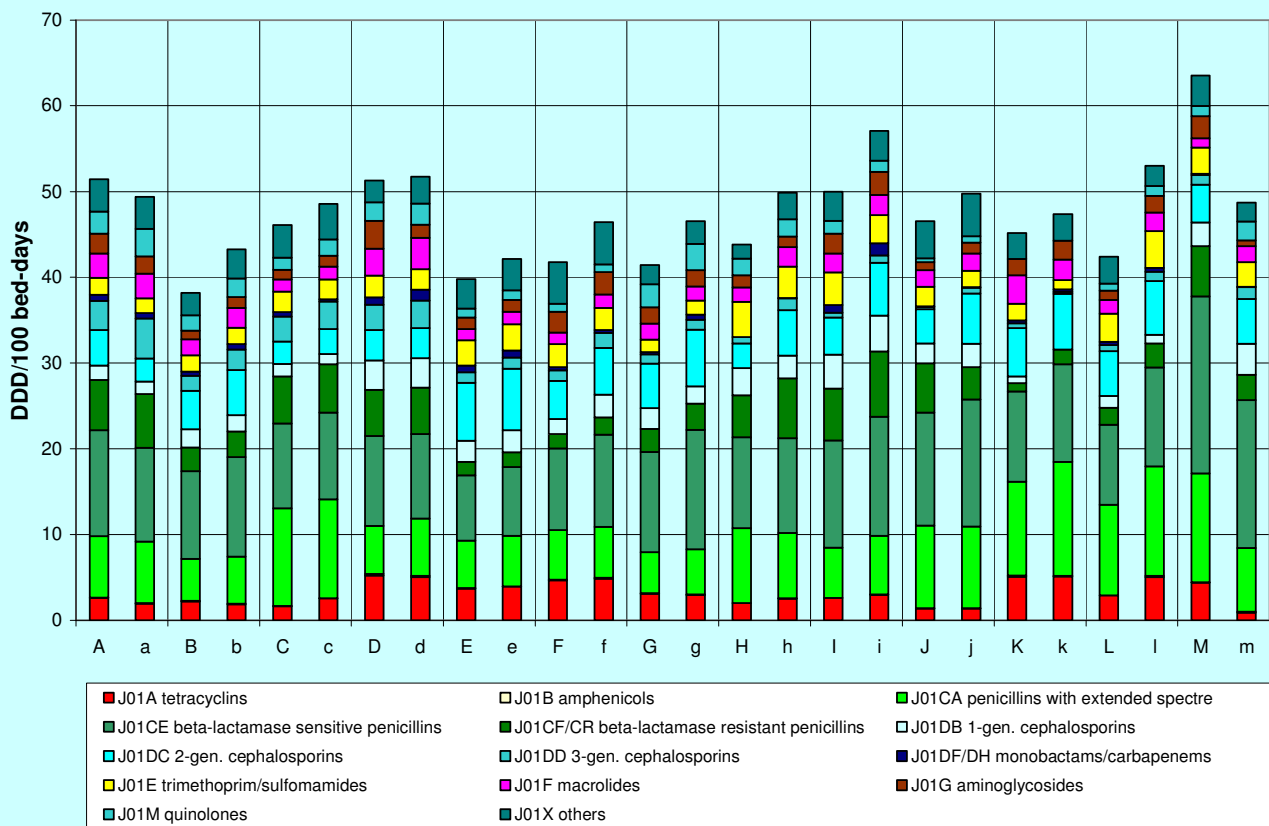
FIGURE 9. Usage of antibacterials in 13 Norwegian hospitals in 1998 (capital letters) and 1999 (small letters). Given in DDD/100 bed-days. ATC/DDD-version 2005 is used. Hospitals are ranged after size from large (A) to small (M).

References:

1. Blix HS and Harthug S. Hospital Usage of Antibacterial Agents in Relation to Size and Type of Hospital and Geographical Situation. Pharmacoepidemiol.Drug Saf 7-2-2005.

Hege Salvesen Blix

VI. OCCURRENCE OF ANTIMICROBIAL RESISTANCE

A. ANIMAL CLINICAL ISOLATES

According to the NORM-VET plan, the clinical isolates included in 2004 were *Staphylococcus intermedius* from skin or ear infections in dogs and pathogenic *E. coli* from septicaemia in poultry and from enteritis and oedema disease in swine. Sampling, laboratory methods and data processing are described in Appendix 3.

Staphylococcus intermedius from dogs

A total of 60 isolates of *Staphylococcus intermedius* from "first time" skin or ear infections in dogs were susceptibility tested. To obtain a representative sample of the Norwegian dog population, only dogs being treated for a skin infection by a veterinarian for the first time were included. See textbox "NORM-VET dog project 2004" below for more information. The results are presented in Table 9, Figure 10 and in the text.

NORM-VET dog project 2004

Previous studies regarding antimicrobial resistance in *S. intermedius* from skin and ear infections in dogs in Norway have indicated an increasing occurrence of resistance. In 2002, only 8% of the 99 isolates tested showed susceptibility to all of the 17 antimicrobials included (NORM/NORM-VET 2002). Although this survey was biased towards problematic cases (dogs with recurrent skin or ear infections), the results indicated that the resistance prevalences in *S. intermedius* from skin and ear infections in dogs were increasing as compared to earlier Norwegian data. To evaluate if the resistance in *S. intermedius* in the dog population in Norway is increasing in general, it was decided to include in NORM-VET 2004 dogs with a "first time" skin infection. The indicator bacteria *Escherichia coli* and *Enterococcus* spp. (*E. faecalis* and *E. faecium)* from the same dogs were also included in the project. To obtain a representative sample, five small animal practises spread throughout Norway were included in the study. Each clinic was asked to submit swabs from both skin and faeces from dogs with a non-treated "first time" pyoderma. Only dogs with no antimicrobial treatment during the previous six months and only one dog per owner were included. A total of 40 skin swabs, 56 ear swabs and 79 faecal swabs were taken.

TABLE 9. Antimicrobial resistance in *Staphylococcus intermedius* from dogs (n=60).

Substance	Resistance (%)	[95% CI*]	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	42	[29.1-55.1]					58.3				1.7	1.7	28.3	10.0			
Chloramphenicol	0	[0.0-6.0]						1.7	65.0	33.3							
Penicillin G**	70	[56.8-81.2]	30.0				6.7	6.7	5.0	3.3	48.3						
Oxacillin	0	[0.0-6.0]				18.3	51.7	30.0									
Cephalothin	0	[0.0-6.0]		3.3	70.0	25.0	1.7										
Trimethoprim#	-	-							3.3	25.0	50.0	16.7	3.3	1.7			
Erythromycin	8	[2.8-18.4]				1.7	78.3	11.7						8.3			
Clindamycin##	8	[2.8-18.4]				53.3	40.0	1.7	1.7	1.7				1.7			
Streptomycin	10	[3.8-20.5]							18.3	65.0	6.7					5.0	5.0
Gentamicin	0	[0.0-6.0]					91.7	8.3									
Neomycin	10	[3.8-20.5]					90.0			6.7	3.3						
Enrofloxacin	0	[0.0-6.0]			20.0	78.3	1.7										
Vancomycin	0	[0.0-6.0]						86.7	13.3								
Fusidic acid	50	[36.8-63.2]			21.7	26.7	1.7		3.3	6.7	28.3	11.7					
Avilamycin	0	[0.0-6.0]						10.0	76.7	13.3							
Virginiamycin	0	[0.0-6.0]					71.7	28.3									

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.

* CI= Confidence interval.

** Resistance to penicillin G was based on beta-lactamase production. All isolates with a positive beta-lactamase test had a MIC-value > 0.125 mg/L, and all beta-lactamase negative isolates had a MIC-value ≤ 0.125 mg/L.

#The microbiological cut-off value has not yet been decided.

##All isolates resistant to erythromycin were tested for inducible clindamycinresistance. Of the six isolates resistant to clindamycin, three were classified as resistant based on inducible clindamycin resistance.
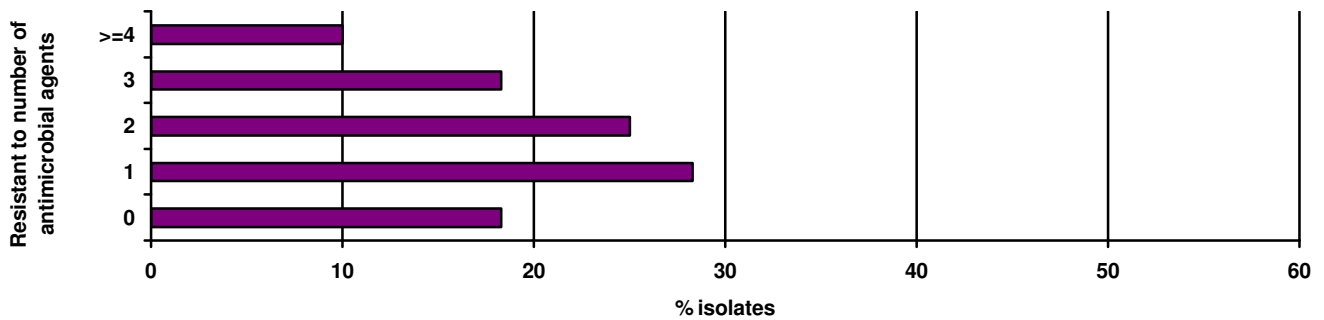
FIGURE 10. Antimicrobial resistance profile for *S. intermedius* isolates from "first time" skin infections (incl. otitis externa) in dogs (n=60). Proportion of isolates susceptible to all antimicrobial agents included and resistant to one, two, three, and four or more antimicrobial agents.

COMMENTS

The occurrence of resistance among *S. intermedius* isolates from dogs with a "first time" skin infection was high. Only 18.3% of the isolates were susceptible to all antimicrobial agents included. Altogether, 28.3% were resistant to one (mainly penicillin), 25% to two (mainly penicillin and oxytetracycline/fusidic acid), 18.3% to three (penicillin, oxytetracycline and fusidic acid) and 10% to four or more antimicrobial agents, as shown in Figure 10. The occurrence of resistance in the present data may seem slightly lower than observed in the previous NORM/NORM-VET reports. However, earlier data were sampled from diagnostic submissions and were thereby probably biased towards problematic and recurrent infections and thus more likely to be resistant. Fusidic acid is the most used preparation for local treatment of skin infections including otitis externa in dogs. Resistance to fucidic acid in *S. intermedius* has until recently not been a problem. However, this favourable situation has changed

in the last years and an increased occurrence of resistance to fucidic acid is now recognized, as confirmed in this study. It would be desirable to study the genetic mechanisms responsible for fusidic acid resistance in staphylococci from animals and humans more thoroughly than has been done previously in order to elucidate common reservoirs of resistance genes and their zoonotic potential.

For systemic treatment of skin infections in dogs a trimethoprim/sulfamethoxazole combination followed by erythromycin, lincomycin or clindamycin are probably the most commonly used antimicrobial agents. The present data indicate that these drugs will still be effective for treatment of most cases of "first time" *S. intermedius* skin infections. In case of recurrent or chronic pyoderma/otitis externa in dogs, however, it is recommendable to perform susceptibility testing.

Resistance to trimethoprim versus trimethoprim/sulfamethoxazole

The VetMIC® plates used for susceptibility testing only includes trimethoprim although the combination trimethoprim/sulfamethoxazole is normally used for therapeutic purposes. However, for monitoring purposes it is preferable to test susceptibility to each substance separately. The susceptibility data for trimethoprim are sparse and therefore a microbiological cut-off value has not yet been decided upon. The 60 dog isolates were also tested for susceptibility to trimethoprim/sulfamethoxazole applying a disk diffusion technique (Rosco) in the routine diagnostic. All the isolates were susceptible to the combination trimethoprim/sulfamethoxazol.

Escherichia coli from swine and poultry

A total of 45 isolates of *E. coli* from swine with the diagnosis enteritis or oedema disease (serotypes O8, O9, O139, O141 and O149) and 19 *E. coli* isolates from septicaemia in poultry (18 broilers and one turkey) were susceptibility tested. The results are presented in Table 10 and Figure 11.

TABLE 10. Antimicrobial resistance in *Escherichia coli* from diagnostic submissions from swine (n=45) and poultry (n=19).

Substance	Sample	Resistance (%) [95% CI*]	Distribution (%) of MIC values (mg/L)														
			0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥512
Oxytetracycline	Swine	24 [12.9-39.5]						73.3	2.2					2.2	22.2		
	Poultry	5 [0.1-26.6]						94.7							5.3		
Chloramphenicol	Swine	4 [0.5-15.2]						20.0	71.1	4.4		4.4					
	Poultry	0 [0.0-17.7]							94.7	5.3							
Florfenicol	Swine	0 [0.0-7.9]							84.4	15.6							
	Poultry	0 [0.0-17.7]							89.5	10.5							
Ampicillin	Swine	7 [1.4-18.3]					8.9	57.8	26.7					6.7			
	Poultry	0 [0.0-17.7]						78.9	21.1								
Ceftiofur	Swine	0 [0.0-7.9]			4.4	37.8	55.6	2.2									
	Poultry	0 [0.0-17.7]				78.9	21.1										
Trimethoprim	Swine	7 [1.4-18.3]				42.2	46.7	4.4						6.7			
	Poultry	5 [0.1-26.6]				5.3	73.7	15.8						5.3			
Sulfamethoxazole	Swine	7 [1.4-18.3]										71.1	20.0	2.2			6.7
	Poultry	5 [0.1-26.6]										78.9	10.5	5.3			5.3
Streptomycin	Swine	47 [31.7-62.1]							2.2	33.3	17.8	2.2	2.2	22.2	11.1		8.9
	Poultry	5 [0.1-26.0]								21.1	73.7				5.3		
Gentamicin	Swine	0 [0.0-7.9]					64.4	35.6									
	Poultry	0 [0.0-17.7]					10.5	78.9	10.5								
Neomycin	Swine	2 [0.1-11.8]							97.8				2.2				
	Poultry	0 [0.0-17.7]							100.0								
Enrofloxacin	Swine	0 [0.0-7.9]	55.6	42.2		2.2											
	Poultry	26 [9.2-51.2]	42.1	31.6			26.3										
Nalidixic acid	Swine	2 [0.1-11.8]					26.7	57.8	13.3					2.2			
	Poultry	26 [9.2-51.2]						68.4	5.3					5.3	15.8	5.3	

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.
* CI= Confidence interval.
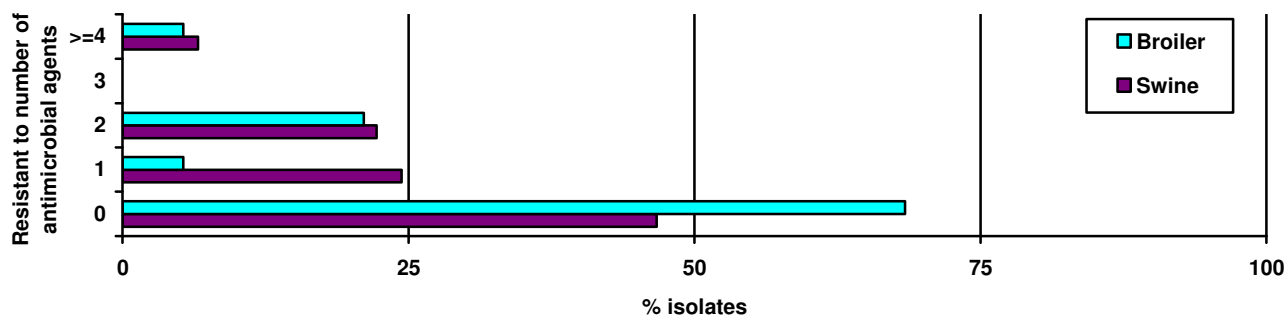
FIGURE 11. Antimicrobial resistance profile for *E. coli* from diagnostic samples from enteritis and oedema disease in swine (n=45) and septicaemia in poultry (n=19). Proportion of isolates susceptible to all antimicrobial agents included and resistant to one, two, three, and four or more antimicrobial agents.

COMMENTS

SWINE

The data indicate a moderate occurrence of resistance among *E. coli* from diseased swine. In total, 46.7% of the isolates were susceptible to all antimicrobial agents included. As presented in Figure 11, 24.4% were resistant to one (predominantly streptomycin), 22.2% to two (mainly streptomycin and oxytetracycline) and 6.6% to four or more antimicrobial agents. Tetracycline and streptomycin are among the most commonly used antimicrobial agents for therapy in swine production. No fluoroquinolone resistance was observed.

POULTRY

The data indicate a moderate occurrence of resistance among *E. coli* from diseased poultry. In total, 68.4% of the isolates were susceptible to all antimicrobial agents included. As presented in Figure 11, 5.3% were resistant to one, 21.1% to two (mainly nalidixid acid and enrofloxacin), and 5.3% to four or more antimicrobial

agents. The occurrence of resistance to quinolones is of concern. All five (26%) isolates resistant to nalidixic acid also had MIC-values for enrofloxacin just above the cut-off value for enrofloxacin. This may indicate that fluoroquinolone resistance in poultry is emerging. In 2002, 2% of the clinical poultry *E. coli* isolates were fluoroquionole resistant, which indicates, although the number of isolates tested was limited, that there has been an increase in fluoroquinolone resistance over the last two years. No quinolone preparations are licensed for use in poultry in Norway. However, veterinarians in Norway may apply for authorisation to use drugs for which no marketing authorisation has been granted. Sulfaclozin (1996-2002) and minor amounts of enrofloxacin (1992-2004) in preparations intended for flock treatment of poultry has been sold in Norway with such exemption. In addition, flumequine (cross-resistance with nalidixic acid) was used for clinical purposes to a very limited extent in the 1980s and the early 1990s.

B. INDICATOR BACTERIA FROM ANIMALS AND FOOD

The prevalence of acquired antimicrobial resistance among certain bacteria of the normal enteric microflora can serve as an indicator of the selective antimicrobial pressure in the various populations. These bacteria may form a reservoir of transferable resistance genes from which antimicrobial resistance can be spread to other bacteria, including those responsible for infections in animals or humans. Thus, monitoring of resistance among so-called indicator bacteria of the normal enteric microflora from healthy animals as well as indicator bacteria from feed and food is important in order to get a better understanding of the resistance situation, to detect trends, and to evaluate the effects of interventions.

In NORM-VET, *E. coli* and *Enterococcus* spp. serve as indicator bacteria. In 2004, indicator bacteria from dogs as well as from swine and broilers at slaughter were included in the monitoring. The faecal samples from dogs were from dogs with a "first time" skin infection but otherwise considered healthy. See textbox "NORM-VET dog project 2004", page 24, for more information. Sampling, laboratory methods and data processing are described in Appendix 3.

Escherichia coli from dogs

A total of 79 faecal samples from dogs were collected and *E. coli* was isolated from 68 (86%) of the samples. One isolate per sample positive for *E. coli* was susceptibility tested. The results are presented in Table 11 and Figure 12.

TABLE 11. Antimicrobial resistance in *Escherichia coli* from faecal samples from dogs (n=68).

Substance	Resistance (%) [95% CI*]	Distribution (%) of MIC values (mg/L)														
		0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥512
Oxytetracycline	3 [0.0-10.2]					2.9	69.1	25.0						2.9		
Chloramphenicol	0 [0.0-5.3]						2.9	1.5	51.5	44.1						
Florfenicol	0 [0.0-5.3]								25.0	72.1	2.9					
Ampicillin	9 [3.3-18.2]					1.5	1.5	41.2	45.6	1.5			8.8			
Ceftiofur	0 [0.0-5.3]			2.9	25.0	69.1	2.9									
Trimethoprim	6 [1.6-14.4]				13.2	63.2	17.6						5.9			
Sulfamethoxazole	9 [3.3-18.2]										64.7	22.1	2.9	1.5		8.8
Streptomycin	13 [6.2-23.6]								20.6	66.2	2.9	2.9		4.4	2.9	
Gentamicin	0 [0.0-5.3]					4.4	89.7	4.4	1.5							
Neomycin	0 [0.0-5.3]								97.1	2.9						
Enrofloxacin	2 [0.0-7.9]	22.1	72.1	4.4		1.5										
Nalidixic acid	2 [0.0-7.9]						2.9	45.6	50.0					1.5		

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.
*CI= Confidence interval

COMMENTS

The occurrence of resistance among faecal *E. coli* isolates from dogs was low. In total, 85.3% of the isolates were susceptible to all antimicrobial agents included. Altogether, 2.9% were resistant to one (predominantly streptomycin), 2.9% to two (mainly streptomycin and sulfamethoxazoles) and 8.8% to three or more antimicrobial agents. Streptomycin is not used in dogs as no preparation is available in Norway, but dihydrostreptomycin might be used for enteric infections. Sulfa is often used, in combination with trimethoprim, for treatment of different types of infections. One isolate (1.5%) was resistant to both enrofloxacin and nalidixid acid. Although this is the first time *E. coli* isolates (as indicator bacteria) from dogs has been included in NORM-VET, the occurrence of fluoroquinolone resistance is noteworthy and monitoring for such resistance should be continued. For the period 2000 - 2004, there was a minor increase in the usage of fluoroquinoles in preparations approved for use in dogs and cats (from 7 to 11 kg), but the usage is still very limited.

Escherichia coli from swine and broilers

A total of 130 faecal and 131 meat samples from swine and 91 faecal and 100 meat samples from broilers were collected. From swine, *E. coli* was identified in 125 (96.1%) of the faecal samples and 97 (74.6 %) of the meat samples. From broilers, *E. coli* was identified in 86 (94.5%) of the faecal samples and 87 (87 %) of the meat samples. One isolate per sample positive for *E. coli* was susceptibility tested. The results are presented in Table 12 and Figure 12.

TABLE 12. Antimicrobial resistance in *Escherichia coli* from faecal (n=125) and (n=97) meat samples from swine and from faecal (n=86) and meat samples (n=87) from broilers.

Substance	Sample*	Resistance (%)	[95% CI**]	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥512
Oxytetracycline	Swine[F]	10	[5.1-16.2]					5.6	70.4	14.4					0.8	8.8		
	Swine[M]	8	[3.6-15.1]					3.1	72.9	15.6						8.3		
	Broiler[F]	7	[1.6-12.4]					1.2	67.4	24.4						7.0		
	Broiler[M]	7	[2.6-14.4]					3.4	78.2	11.5						6.9		
Chloramphenicol	Swine[F]	<1	[0.0-4.4]						0.8	7.2	54.4	36.8					0.8	
	Swine[M]	1	[0.0-5.6]							15.5	57.7	25.8					1.0	
	Broiler[F]	0	[0.0-4.2]							8.1	69.8	22.1						
	Broiler[M]	0	[0.0-4.2]							3.4	73.6	23.0						
Florfenicol	Swine[F]	0	[0.0-2.9]								40.0	56.8	3.2					
	Swine[M]	0	[0.0-3.7]								46.4	52.6	1.0					
	Broiler[F]	0	[0.0-4.2]								51.2	47.7	1.2					
	Broiler[M]	0	[0.0-4.2]								49.4	48.3	2.3					
Ampicillin	Swine[F]	8	[3.9-14.2]						5.6	56.0	28.0	2.4	1.6		6.4			
	Swine[M]	9	[4.3-16.9]							7.2	47.4	36.1		1.0	8.2			
	Broiler[F]	17	[10.1-27.1]					1.2	14.0	43.0	24.4				17.4			
	Broiler[M]	23	[14.6-33.3]						5.8	34.9	36.0				23.3			
Ceftiofur	Swine[F]	0	[0.0-2.9]				32.8	64.0	3.2									
	Swine[M]	0	[0.0-3.7]			1.0	38.1	58.8	2.1									
	Broiler[F]	0	[0.0-4.2]			3.5	39.5	52.3	4.7									
	Broiler[M]	0	[0.0-4.2]				24.1	58.6	16.1	1.1								
Trimethoprim	Swine[F]	4	[1.3-9.1]				27.2	57.6	10.4	0.8					4.0			
	Swine[M]	7	[3.0-14.3]				24.7	60.8	7.2						7.2			
	Broiler[F]	2	[0.3-8.2]				33.7	55.8	7.0	1.2				1.2	1.2			
	Broiler[M]	2	[0.3-8.1]				20.7	66.7	10.3						2.3			
Sulfamethoxazole	Swine[F]	12	[6.9-19.0]										76.0	12.0				12.0
	Swine[M]	11	[5.8-19.4]										81.4	5.2	2.1			11.3
	Broiler[F]	14	[7.4-23.1]										75.6	10.5				14.0
	Broiler[M]	13	[6.5-21.5]										79.3	8.0				12.6
Streptomycin	Swine[F]	34	[25.4-42.6]								21.6	44.8	6.4	1.6	8.8	9.6	1.6	5.6
	Swine[M]	20	[12.2-28.9]								29.2	51.0	3.1	1.0	4.2	4.2	4.2	3.1
	Broiler[F]	9	[4.1-17.5]								33.7	57.0	7.0	1.2		1.2		
	Broiler[M]	16	[9.1-25.5]								19.5	64.4	10.3	3.4		2.3		
Gentamicin	Swine[F]	0	[0.0-2.9]					16.8	75.2	8.0								
	Swine[M]	0	[0.0-3.7]					13.4	81.4	4.1	1.0							
	Broiler[F]	0	[0.0-4.2]					16.3	76.7	7.0								
	Broiler[M]	0	[0.0-4.2]					6.9	82.8	9.2	1.1							
Neomycin	Swine[F]	<1	[0.0-4.4]								96.8	2.4		0.8				
	Swine[M]	1	[0.0-5.6]								99.0			1.0				
	Broiler[F]	0	[0.0-4.2]								96.5	3.5						
	Broiler[M]	0	[0.0-4.2]								95.4	4.6						
Enrofloxacin	Swine[F]	0	[0.0-2.9]	16.8	78.4	4.8												
	Swine[M]	0	[0.0-3.7]	27.8	70.1	2.1												
	Broiler[F]	0	[0.0-4.2]	20.9	70.9	7.0	1.2											
	Broiler[M]	3	[0.7-9.8]	13.8	79.3	3.4		1.1	2.3									
Nalidixic acid	Swine[F]	0	[0.0-2.9]						1.6	50.4	46.4	1.6						
	Swine[M]	0	[0.0-3.7]						7.2	44.3	47.4		1.0					
	Broiler[F]	0	[0.0-4.2]							3.5	62.8	33.7						
	Broiler[M]	3	[0.7-9.8]							2.3	57.5	36.8						3.4

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.
*F=faeces, M=meat.
** CI= Confidence interval.
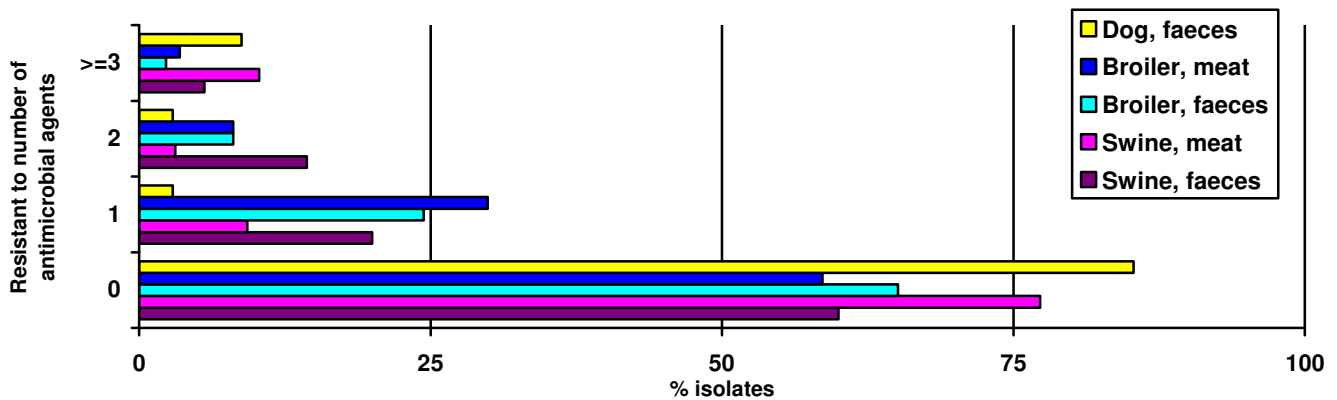
FIGURE 12. Antimicrobial resistance profile for *E. coli* from faecal and meat samples from swine (125 faecal and 97 meat isolates), broiler (86 faecal and 87 meat isolates), and dog (68 faecal isolates). Proportion of isolates susceptible to all antimicrobials included and resistant to one, two, and three or more antimicrobial agents.

COMMENTS

SWINE

The data indicate a moderate occurrence of resistance among *E. coli* from faecal and meat samples from a representative group of Norwegian swine.

In total, 60.0% and 77.3% of the isolates, respectively, were susceptible to all antimicrobial agents included. Altogether, 20% and 9.3% of the faecal and meat isolates, respectively, were resistant to one (predominantly streptomycin), 14.4% and 3.1%, respectively, to two (mainly streptomycin and sulfamethoxazoles) and 5.6% and 10.3%, respectively, to three or more antimicrobial agents (Figure 12). Resistance to streptomycin was most frequent in both categories (faeces and meat), followed by resistance to sulfamethoxazole, oxytetracycline and ampicillin. All these antimicrobial agents are commonly used for clinical purposes in swine.

One faecal isolate was resistant to neomycin and one faecal isolate was resistant to chloramphenicol. Veterinary drugs containing chloramphenicol were withdrawn from the Norwegian market in 1992. No resistance to the fluoroquinolone enrofloxacin or to the quinolone nalidixic acid was observed. The usage of fluoroquinolones in food producing animals in Norway is very limited. No resistance to ceftiofur or gentamicin was observed. No preparations containing cephalosporins or the aminoglycoside gentamicin have been approved for veterinary use in Norway. However, veterinarians are allowed to prescribe antimicrobial agents registered for use in humans, and such prescribing does occur among especially small animal practitioners.

BROILERS

The occurrence of resistance among *E. coli* from faecal and meat samples from broilers was moderate.

In total, 65.1% and 58.6% of the isolates, respectively, were susceptible to all antimicrobial agents included. Altogether, 24.4% and 29.9% of the faecal and meat isolates, respectively, were resistant to one (predominantly ampicillin), 8.1% and 8.1%, respectively, to two (mainly ampicillin and sulfamethoxazoles) and 2.3% and 3.5%, respectively, to three or more antimicrobial agents (Figure 12). Resistance to ampicillin was most commonly observed, followed by resistance to sulfamethoxazole and oxytetracycline. Compared to data from NORM-VET 2002, there is a significant (p<0.05) increase in resistance to ampicillin in *E. coli* from broiler meat samples. The same is the case for streptomycin, however, this is mainly explained by a change of the microbiological cut off value. See textbox entitled "The genetic background for streptomycin resistance in *Escherichia coli* influences the distribution of MICs", page 31 for further information. There is some use of tetracycline and amoxicillin (cross-resistance with ampicillin) for clinical purposes in broilers, whereas streptomycin and trimetoprim are not used in Norwegian broiler production.

The genetic background for streptomycin resistance in
Escherichia coli influences the distribution of MICs

The expression of the various genes involved in conferring resistance to antimicrobial agents is not always at the same level. Some resistance genes are involved in conferring high-level resistance, whereas expression of low-level resistance may be the case for others.

The genetic background for resistance to streptomycin in a selection of 136 *E. coli* isolates with MICs to streptomycin of ≥16mg/L was studied. The isolates originated from meat and meat products and were collected within the frame of NORM-VET. PCR was carried out for detection of the streptomycin resistance genes *strA-strB* and the integron associated *aadA* gene cassettes. The *strA-strB* genes and/or an *aadA* gene cassette were detected in 110 (80.9%) of the isolates. The *strA-strB* genes were the most prevalent, detected in 90 isolates. The *aadA* gene cassettes were detected in 29 isolates, whereas nine isolates harboured both the *strA-strB* genes and an *aadA* gene cassette.

The distribution of MIC values differed considerably between isolates harbouring the *strA-strB* genes (solely) (MIC_{50} =128 mg/L) and isolates harbouring an *aadA* gene cassette (solely) (MIC_{50}=16 mg/L) (Figure 13). The *strA-strB* genes are probably involved in conferring high-level resistance to streptomycin, whereas conferring low-level resistance seems to be the case for the *aadA* gene cassettes. Isolates harbouring both the *strA-strB* genes and an *aadA* gene cassette had higher streptomycin MICs than those harbouring either one alone (Figure 13). This study shows that the distribution of streptomycin MIC values in *E. coli* can be greatly influenced by the genes encoding resistance to streptomycin. Low-level streptomycin resistance, caused by the presence of *aadA* gene cassettes in integrons, represents an obstacle in classifying *E. coli* as susceptible or resistant to streptomycin. Furthermore, the determination of an epidemiological cut-off value for surveillance purposes is also complicated by dissemination of integrons containing the *aadA* cassettes. Based on these findings, for *E. coli*, a microbiological cut-off value of > 8 mg/L is proposed for streptomycin, and is used in this report.
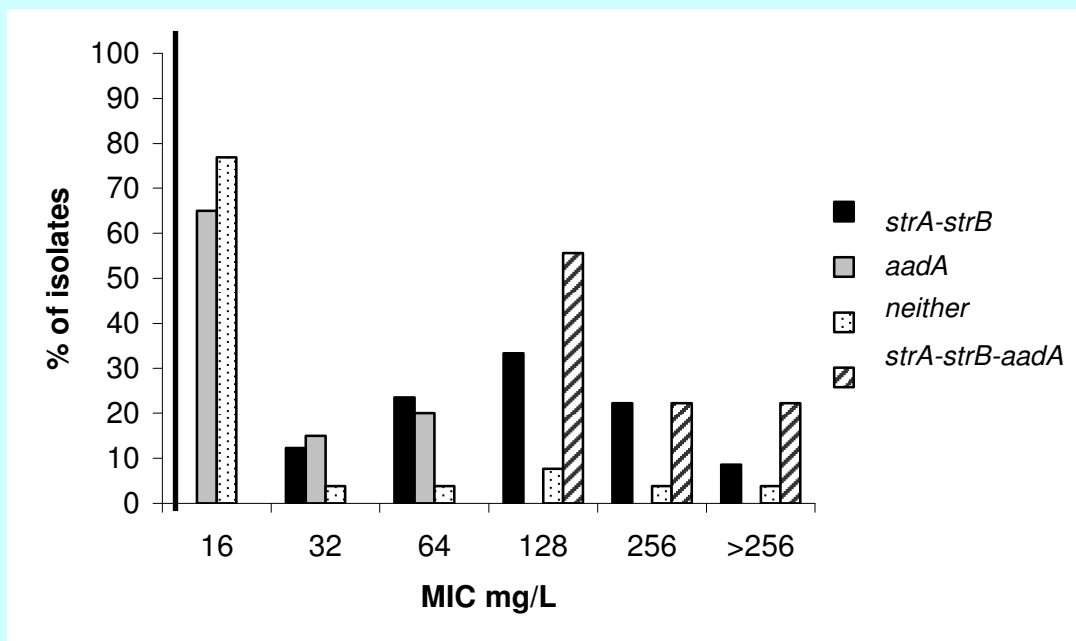
FIGURE 13. Distribution (%) of MICs of streptomycin in *E. coli* isolates with a MIC ≥ 16 and harbouring the *strA-strB* gene (solely) (n=81) (black), *aadA1* gene cassette (n=20) (grey), both the *strA-strB* genes and an *aadA* gene cassette (n=9) (black and white stripes) and either the *strA-strB* genes nor an *aadA* gene cassette (n=26) (black points). The vertical black thin line indicates a possible choice of a microbiological cut-off vale for resistance to streptomycin at >8mg/L.

References:
1. Sunde M., Norström M. The genetic background for streptomycin resistance in *Escherichia coli* influences the distribution of MICs. J. Antimicrob. Chemother. Jul 2005; 56: 87-90.

Marianne Sunde and Madelaine Norström

Enterococcus spp. from dogs

A total of 79 faecal samples from dogs were collected. *E. faecium* or *E. faecalis* was isolated from 48 (60.8%) of the samples. One isolate per positive sample was susceptibility tested. The results are presented in Tables 13, 14, Figure 14 and in the text.

TABLE 13. Antimicrobial resistance in *Enterococcus faecalis* from faecal samples from dogs (n=36)

Substance	Resistance (%)	[95% CI*]	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	≥ 2048
Oxytetracycline	28	[14.2-45.2]			8.3	61.1	2.8				16.7	11.1					
Chloramphenicol	3	[0.1-14.5]				8.3		25.0	63.9		2.8						
Ampicillin	0	[0.0-9.7]			11.1	72.2	16.7										
Erythromycin	6	[0.7-18.7]			8.3	16.7	47.2	22.2	2.8					2.8			
Streptomycin	11	[3.1-26.1]													88.9		11.1
Gentamicin	3	[0.1-14.5]													94.4	2.8	2.8
Neomycin	6	[0.7-18.7]							2.8	11.1	47.2	22.2	8.3	2.8	5.6		
Vancomycin	3	[0.1-14.5]				5.6	69.4	22.2	2.8								
Bacitracin**	3	[0.1-14.5]							8.3	55.6	33.3			2.8			
Avilamycin	0	[0.0-9.7]				30.6	63.9	5.6									
Virginiamycin#	NR#	NR#					2.8	2.8	11.1	58.3	25.0						
Flavomycin	8	[1.8-22.5]					22.2	19.4	44.4	5.6				8.3			
Narasin	3	[0.1-14.5]	44.4	38.9	11.1	2.8		2.8									

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.
* CI= Confidence interval.
** Measured in U/ml.
# Not relevant, as *E. faecalis* is inherently resistant to virginiamycin.

TABLE 14. Antimicrobial resistance in *Enterococcus faecium* from faecal samples from dogs (n=12).

Substance	Resistance (%)	[95% CI*]	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	≥ 2048
Oxytetracycline	42	[15.2-72.3]				58.3				8.3	8.3	8.3	16.7				
Chloramphenicol	0	[0.0-26.5]						8.3	91.7								
Ampicillin	8	[0.2-38.5]			16.7	41.7	16.7	16.7				8.3					
Erythromycin	33	[9.1-65.1]			16.7		25.0	25	16.7	16.7							
Streptomycin	0	[0.0-26.5]												100			
Gentamicin	0	[0.0-26.5]												100			
Neomycin	0	[0.0-26.5]								83.3	16.7						
Vancomycin	0	[0.0-26.5]				66.7	33.3										
Bacitracin**	17	[2.1-48.4]							8.3	8.3	16.7	50.0	16.7				
Avilamycin	0	[0.0-26.5]				16.7	25.0	50.0	8.3								
Virginiamycin	0	[0.0-26.5]				33.3	16.7	50.0									
Flavomycin#	NR#	NR#												100			
Narasin	0	[0.0-26.5]			8.3	75.0	16.7										

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest Measured in
* CI= Confidence interval.
** Measured in U/ml.
# Not relevant, as *E. faecium* is inherently resistant to flavomycin.

COMMENTS

The occurrence of resistance among faecal *E. faecalis* and *E. faecium* from healthy dogs was moderate. In total, 66.7% of the *E. faecalis* and 25.0% of the *E. faecium* isolates were susceptible to all antimicrobial agents included, while 19.4% and 50%, respectively, were resistant to one, and 13.9% and 25%, respectively, resistant to two or more antimicrobial agents (Figure 14). Resistance to oxytetracycline was most commonly observed followed by resistance to erythromycin, ampicillin and bacitracin. Tetracycline, erythromycin and ampicillin are used for clinical purposes in dogs.

One isolate of *E. faecalis* was resistant to vancomycin (MIC=8), but was not *vanA* positive. Another *E. faecalis* isolate showed high-level gentamicin resistance. No preparations containing vancomycin or the aminoglycoside gentamicin have been approved for veterinary use in Norway. However, veterinarians are allowed to prescribe antimicrobial agents licenced for use in humans, and it is assumed that some prescribing of gentamicin occur especially among small animal practitioners

Enterococcus spp. from swine and broilers

A total of 130 faecal and 131 meat samples from swine and 91 faecal and 100 meat samples from broilers were collected. *E. faecium* or *E. faecalis* was identified in 67 (51.5%) of the faecal samples and 58 (44.3%) of the meat samples from swine, and from 84 (92.3 %) of the faecal samples and 79 (79.0%) of the meat samples from broilers. One isolate per positive sample was susceptibility tested. The results are presented in Tables 15 and 16, Figure 14 and in the text.

TABLE 15. Antimicrobial resistance in *Enterococcus faecalis* from faecal (n=45) and meat (n=47) samples from swine and from faecal (n=22) and meat (n=29) samples from broilers.

Substance	Sample*	Resistance (%)	[95% CI**]	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	≥2048
Oxytetracycline	Swine[F]	69	[53.4-81.8]				28.9	2.2		2.2	13.3	13.3	37.8	2.2				
	Swine[M]	9	[2.4-20.4]				80.9	10.6				6.4	2.1					
	Broiler[F]	23	[7.8-45.4]			4.5	72.7				4.5	4.5	9.1	4.5				
	Broiler[M]	35	[17.9-54.3]				65.5				3.4	13.8	10.3	6.9				
Chloramphenicol	Swine[F]	0	[0.0-7.9]					13.3	86.7									
	Swine[M]	0	[0.0-7.6]					19.1	80.9									
	Broiler[F]	0	[0.0-15.4]					27.3	72.7									
	Broiler[M]	0	[0.0-11.9]					27.6	72.4									
Ampicillin	Swine[F]	0	[0.0-7.9]			8.9	80.0	11.1										
	Swine[M]	0	[0.0-7.6]			14.9	51.1	34.0										
	Broiler[F]	0	[0.0-15.4]			9.1	90.9											
	Broiler[M]	0	[0.0-11.9]			31.0	58.6	10.3										
Erythromycin	Swine[F]	4	[0.5-15.2]			6.7	6.7	62.2	20.0	2.2				2.2				
	Swine[M]	14	[6.2-28.3]			10.6	4.3	19.1	51.1	12.8				2.1				
	Broiler[F]	9	[1.1-29.2]			18.2	9.1	27.3	36.4	4.5				4.5				
	Broiler[M]	17	[5.9-35.8]			17.2	10.3	20.7	34.5	3.4	3.4	6.9		3.4				
Streptomycin	Swine[F]	13	[5.1-26.8]												84.4	2.2		13.3
	Swine[M]	2	[0.1-11.3]												97.9			2.1
	Broiler[F]	0	[0.0-15.4]												100			
	Broiler[M]	3	[0.1-17.8]												96.6			3.4
Gentamicin	Swine[F]	2	[0.1-11.8]												97.8		2.2	
	Swine[M]	0	[0.0-7.6]												100			
	Broiler[F]	0	[0.0-15.4]												100			
	Broiler[M]	0	[0.0-11.9]												100			
Neomycin	Swine[F]	7	[1.4-18.3]							2.2	2.2	31.1	55.6	2.2			6.7	
	Swine[M]	2	[0.1-11.3]								4.3	12.8	61.7	17.0	2.1		2.1	
	Broiler[F]	0	[0.0-15.4]									9.1	72.7	18.2				
	Broiler[M]	0	[0.0-11.9]									13.8	62.1	20.7	3.4			
Vancomycin	Swine[F]	0	[0.0-7.9]				4.4	84.4	11.1									
	Swine[M]	0	[0.0-7.6]				2.1	68.1	29.8									
	Broiler[F]	0	[0.0-15.4]				4.5	72.7	22.7									
	Broiler[M]	0	[0.0-11.9]					79.3	20.7									
Bacitracin[#]	Swine[F]	2	[0.1-11.8]					2.2		2.2	8.9	73.3	11.1	2.2				
	Swine[M]	6	[1.3-17.5]							2.1	10.6	46.8	34.0	4.3	2.1			
	Broiler[F]	9	[1.1-29.2]								9.1	45.5	36.4	9.1				
	Broiler[M]	31	[15.3-50.8]								10.3	31.0	27.6	3.4	6.9	20.7		
Avilamycin	Swine[F]	0	[0.0-7.9]				4.4	82.2	13.3									
	Swine[M]	0	[0.0-7.6]				10.6	72.3	17.0									
	Broiler[F]	0	[0.0-15.4]				9.1	90.9										
	Broiler[M]	0	[0.0-11.9]				17.2	65.5	13.8	3.4								
Virginiamycin[##]	Swine[F]	NR[##]	NR[##]								51.1	48.9						
	Swine[M]	NR[##]	NR[##]							2.1	70.2	27.7						
	Broiler[F]	NR[##]	NR[##]						4.5	4.5	63.6	27.3						
	Broiler[M]	NR[##]	NR[##]							3.4	69.0	27.6						
Flavomycin	Swine[F]	2	[0.1-11.8]					17.8	26.7	48.9	4.4			2.2				
	Swine[M]	11	[3.6-23.1]					17.0	8.5	57.4	6.4			10.6				
	Broiler[F]	0	[0.0-15.4]					9.1	27.3	63.6								
	Broiler[M]	3	[0.1-17.8]					13.8	17.2	44.8	20.7		3.4					
Narasin	Swine[F]	0	[0.0-7.9]	13.3	46.7	40.0												
	Swine[M]	0	[0.0-7.6]	21.3	72.3	6.4												
	Broiler[F]	23	[7.8-45.4]	18.2	36.4	18.2	4.5	22.7										
	Broiler[M]	7	[0.9-22.8]	27.6	31.0	24.1	10.3	6.9										

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.

*F=faeces, M=meat

** CI = Confidence interval.

[#] Measured in U/ml.

[##] Not relevant, as *E. faecalis* is inherently resistant to virginiamycin.

TABLE 16. Antimicrobial resistance in *Enterococcus faecium* from faecal (n=22) and meat (n=11) samples from swine and from faecal (n=62) and meat (n=50) samples from broilers.

Substance	Sample*	Resistance (%)		Distribution (%) of MIC values (mg/L)														
			[95% CI**]	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	≥ 2048
Oxytetracycline	Swine[F]	18	[5.2-40.3]			9.1	68.2	4.5					9.1	9.1				
	Swine[M]	0	[0.0-28.5]				90.9	9.1										
	Broiler[F]	18	[8.2-27.2]			19.4	61.3	1.6					8.1	6.5	3.2			
	Broiler[M]	6	[1.3-16.6]			8.0	86.0				2.0	4.0						
Chloramphenicol	Swine[F]	0	[0.0-15.4]						36.4	63.6								
	Swine[M]	0	[0.0-28.5]						54.5	45.5								
	Broiler[F]	0	[0.0-5.8]					1.6	1.6	12.9	46.8	37.1						
	Broiler[M]	0	[0.0-7.0]						10.0	30.0	60.0							
Ampicillin	Swine[F]	0	[0.0-15.4]		4.5	4.5	18.2	36.4	31.8	4.5								
	Swine[M]	0	[0.0-28.5]		18.2	9.1	36.4	27.3	9.1									
	Broiler[F]	0	[0.0-5.8]		8.1	30.6	17.7	24.2	17.7	1.6								
	Broiler[M]	0	[0.0-7.0]			10.0	40.0	40.0	10.0									
Erythromycin	Swine[F]	18	[5.2-40.3]			22.7	4.5	18.2	36.4	13.6	4.5						
	Swine[M]	27	[6.0-61.0]				9.1	18.2	45.5	27.3							
	Broiler[F]	6.4	[0.0-12.6]			17.7	8.1	56.5	11.3	1.6	1.6	3.2					
	Broiler[M]	24	[13.1-38.2]			10.0	16.0	40.0	10.0	12.0	10.0			2.0			
Streptomycin	Swine[F]	14	[2.9-34.9]												77.3		9.1	13.6
	Swine[M]	0	[0.0-28.5]												100			
	Broiler[F]	0	[0.0-5.8]												100			
	Broiler[M]	2	[0.1-10.7]												98.0			2.0
Gentamicin	Swine[F]	0	[0.0-15.4]												100			
	Swine[M]	0	[0.0-28.5]												100			
	Broiler[F]	0	[0.0-5.8]												100			
	Broiler[M]	0	[0.0-7.0]												100			
Neomycin	Swine[F]	0	[0.0-15.4]								63.6	22.7	13.6					
	Swine[M]	0	[0.0-28.5]								45.5	36.4	18.2					
	Broiler[F]	0	[0.0-5.8]								80.6	14.5	3.2	1.6				
	Broiler[M]	0	[0.0-7.0]								78.0	16.0	6.0					
Vancomycin	Swine[F]	0	[0.0-15.4]				63.6	27.3	9.1									
	Swine[M]	0	[0.0-28.5]				81.8	9.1	9.1									
	Broiler[F]	5	[1.0-13.5]				75.8	17.7	1.6						4.8			
	Broiler[M]	0	[0.0-7.0]				58.0	30.0	12.0									
Bacitracin[#]	Swine[F]	32	[13.9-54.9]				4.5		4.5	4.5	9.1	45.5	13.6	4.5	13.6			
	Swine[M]	9	[0.2-41.3]							9.1	18.2	63.6	9.1					
	Broiler[F]	18	[9.2-29.5]				8.1	29.0	3.2	4.8	16.1	21	6.5	6.5	4.8			
	Broiler[M]	54	[39.3-68.2]				2.0	4.0	6.0	6.0	2.0	26.0	16.0	22.0	16.0			
Avilamycin	Swine[F]	0	[0.0-15.4]					4.5	40.9	45.5	9.1							
	Swine[M]	0	[0.0-28.5]						72.7	27.3								
	Broiler[F]	0	[0.0-5.8]				6.5	29.0	51.6	12.9								
	Broiler[M]	0	[0.0-7.0]					10.0	64.0	22.0	4.0							
Virginiamycin	Swine[F]	0	[0.0-15.4]				13.6	22.7	59.1	4.5								
	Swine[M]	9	[0.2-41.3]				45.5	9.1	18.2	18.2	9.1							
	Broiler[F]	2	[0.0-8.7]			6.5	29.0	21.0	40.3	1.6	1.6							
	Broiler[M]	4	[0.5-13.7]			2.0	42.0	26.0	16.0	10.0	4.0							
Flavomycin[##]	Swine[F]	NR[##]	NR[##]												100			
	Swine[M]	NR[##]	NR[##]							9.1					90.9			
	Broiler[F]	NR[##]	NR[##]									3.2	4.8	1.6	90.3			
	Broiler[M]	NR[##]	NR[##]				2.0								98.0			
Narasin	Swine[F]	0	[0.0-15.4]	4.5	18.2	63.6	13.6											
	Swine[M]	0	[0.0-28.5]		36.4	54.5		9.1										
	Broiler[F]	79	[68.8-88.3]	4.8	6.5	6.5	3.2		29.0	48.4		1.6						
	Broiler[M]	84	[70.9-92.8]		2.0	8.0	4.0	2.0	30.0	54.0								

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.

*F=faeces, M=meat

** CI = Confidence interval.

[#] Measured in U/ml.

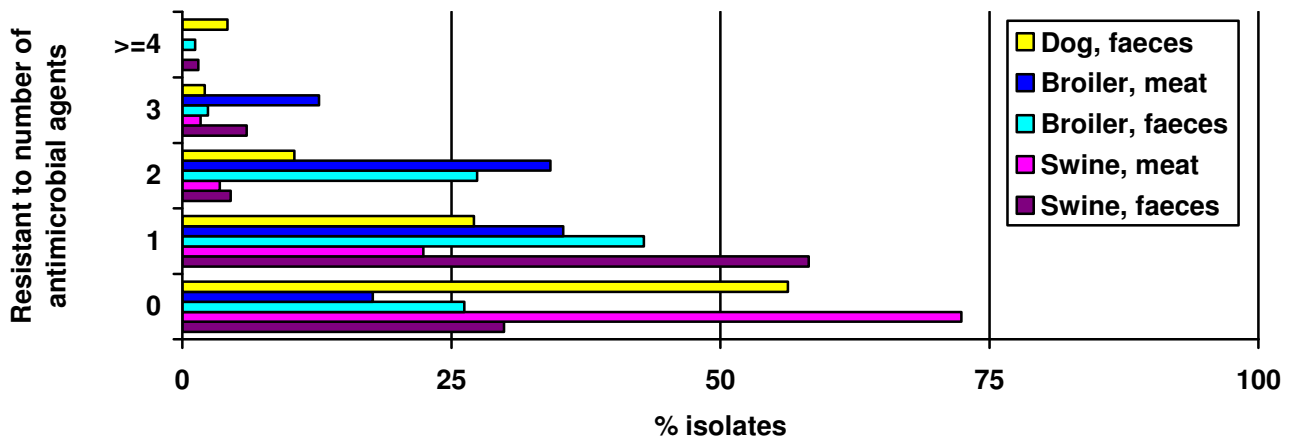[##] Not relevant, as *E. faecium* is inherently resistant to flavomycin.

FIGURE 14. Antimicrobial resistance profile for enterococci (*E. faecalis* and *E. faecium*) from porcine faecal (n=67) and meat (n=58) samples, broiler faecal (n=84) and meat (n=79) samples, and dog faecal samples (n=48). Proportion of isolates susceptible to all antimicrobial agents included and resistant to one, two, three and four or more antimicrobial agents.

COMMENTS

E. faecalis is known to be inherently resistant to the streptogramin virginiamycin, while *E. faecium* is known to be susceptible to this antimicrobial agent. The situation is reversed for flavomycin. The use of virginiamycin in animal production in Norway has been negligible, and the substance was banned in 1998. Flavomycin has never been approved in Norway. Resistance to virginiamycin and flavomycin is not included in the following comments.

SWINE

The occurrence of resistance among *E. faecalis* and *E. faecium* from healthy swine was moderate. The resistance frequencies observed for the faecal isolates were higher than for the meat isolates. The resistance frequencies for the meat isolates were more or less at the same level as observed for pork isolates in NORM-VET 2002 and 2000 and in a Norwegian survey from 1998 (Kruse, H. SNT-report no. 1, 1999).

Regarding *E. faecalis*, a total of 22.2% of the faecal and 72.3% of the meat isolates were susceptible to all antimicrobial agents included. Altogether, 66.7% of the faecal and 23.4% of the meat isolates were resistant to one antimicrobial agent. Altogether, 6.7% of the faecal and 2.1% of the meat isolates were resistant to two antimicrobial agents, whereas 4.4% of the faecal and 2.1% of the meat isolates were resistant to to three or more antimicrobial agents.

Regarding *E. faecium*, a total of 45.5% of the faecal and 72.7% of the meat isolates were susceptible to all antimicrobial agents included. Altogether, 40.9% of the faecal and 18.2% of the meat isolates were resistant to one antimicrobial agent. Altogether, 0% of the faecal and 9.1% of the meat isolates were resistant to two antimicrobial agents, whereas 13.6% of the faecal and 0% of the meat isolates were resistant to to three or more antimicrobial agents.

Resistance to oxytetracycline, streptomycin and erythromycin was most common. Tetracycline and streptomycin are common therapeutics in Norwegian pig production, the latter one in combination with penicillin. The occurrence of resistance among the faecal *E. faecalis* isolates to erythromycin has increased significantly

(p<0.05) compared to NORM-VET 2002. No macrolides or lincosamides are currently licensed for use in pigs in Norway. However, a veterinary spiramycin formulation was available in the 1990s. Moreover, veterinarians may apply for authorisation to use drugs for which no marketing authorisation has been granted, and since 1998 the macrolide tylosin has been used with such exemption for therapeutic use in pigs (3.7 kg active substance sold in 2004).

Neomycin is used in minor amounts as an oral therapeutic agent against enteritis in pigs. All veterinary preparations containing chloramphenicol were withdrawn from the Norwegian market in 1992.

No antimicrobial growth promoters are used in Norwegian pig production.

BROILER

Coccidiostats are routinely used in Norwegian broiler production and since 1996 such use has been dominated by the ionophore narasin. The selection pressure exerted by the common use of narasin in broiler production is probably the reason why narasin resistance is commonly observed among enterococci from broilers, *E. faecium* in particular, as opposed to enterococci from swine and dogs. The prevalence of resistance to the other antimicrobial agents among *E. faecalis* and *E. faecium* from healthy broilers was moderate. In general, a lower prevalence of resistance was observed among the *E. faecium* isolates as compared to the *E. faecalis*.

Regarding *E. faecalis*, a total of 54.6% of the faecal and 37.9% of the meat isolates were susceptible to all antimicrobial agents included. Altogether, 36.4% of the faecal and 34.5% of the meat isolates were resistant to one antimicrobial agent. Altogether 9.1% of the faecal and 27.6% of the meat isolates were resistant to two or more antimicrobial agents.

Regarding *E. faecium*, a total of 16.1% of the faecal and 6.0% of the meat isolates were susceptible to all antimicrobial agents included. Altogether, 45.2% of the faecal and 36.0%, of the meat isolates were resistant to one antimicrobial agent. Altogether 38.7% of the faecal

and 58.0% of the meat isolates were resistant to two or more antimicrobial agents.

Resistance was most commonly observed to oxytetracycline, erythromycin and bacitracin. There is minor use of tetracycline for clinical purposes in Norwegian broiler production. No spiramycin preparation is currently licensed for therapeutic use in poultry (withdrawn 1998 due to limited sales). Cross-resistance between erythromycin and spiramycin is common. Bacitracin was used as a growth promoter in earlier years. However, during the 1990s the usage of bacitracin as a growth promoter was negligible, and since 1997 no such use has been recorded in animal production in Norway.

Three (5%) of the 62 faecal isolates of *E. faecium* were *vanA* positive, all with MIC-values >128, whereas none of the 50 meat *E. faecium* isolates were vancomycin resistant. This means that 3% of the *E. faecium* isolates from poultry obtained by a random selection were vancomycin resistant.

Avoparcin, which confers cross-resistance to vancomycin, was routinely used as a growth promoter in Norwegian broiler and turkey production from 1986 until it was banned in 1995. Studies have shown that this use has selected for an extensive occurrence of vancomycin resistant enterococci (VRE) in Norwegian broiler production, and that VRE have persisted and remained at a high prevalence level for at least eight years after the ban was implemented.

Due to technical problems, results from the selective isolation of VRE from broiler were obtained for only 35

and 60 of the faecal and meat samples, respectively. VRE were detected in 57.2% (20/35) of the faecal samples and in 10% (6/60) of the meat samples. All meat isolates and 90% of the faecal isolates were identified as *E. faecium*, and the remaining 10% of the faecal isolates were identified as *Enterococcus* spp. None of the isolates were identified as *E. faecalis*. The prevalence of VRE in the fecal samples was considerably lower in 2004 than in 2002. This is in accordance with the results obtained in a long-term cohort study, which showed that the flock prevalence of VRE continued to be at a high level after the avoparcin ban, but seemed to have been declining significantly during the last years. In 2004, the prevalence of VRE in the cohort study was approximately 50%, which is in accordance with the present NORM-VET results.

The prevalence of VRE in the meat samples were considerably lower than in a study conducted in 1998, where VRE were detected in 81% of the meat samples by an enrichment method identical to the one used in NORM-VET 2004. However, the type of meat sample in the two studies was quite different, which might at least partly explain the difference. By the conventional NORM-VET isolation method, the fraction of enterococci from poultry being resistant to vancomycin has been approximately the same in 2002 and 2004. This indicates that the concentration of VRE in the samples is at a fairly constant level, something which is also in accordance with the results from the abovementioned long-term cohort study.

C. ZOONOTIC AND NON-ZOONOTIC ENTEROPATHOGENIC BACTERIA

Zoonotic and non-zoonotic enteropathogenic bacteria represent a considerable public health problem. Furthermore, the increasing occurrence of antimicrobial resistance in such bacteria represent a major public health concern. Therefore, it is of great importance to monitor the occurrence of antimicrobial resistance in zoonotic and other enteropathogenic bacteria at relevant stages in the farm-to-fork continuum.

In Norway, *Salmonella* isolates from feed, animals and food, as well as a representative number of *Campylobacter* isolates from broiler and broiler meat are monitored for antimicrobial resistance. Regarding human isolates, all *Salmonella*, *Shigella* and *Yersinia enterocolitica* isolates are monitored, as well as a representative number of *Campylobacter* isolates. Sampling, laboratory methods and data processing are described in Appendix 4.

SALMONELLA SPP.

Salmonella from animals

The situation regarding *Salmonella* spp. in food producing animals in Norway is very good. Such animals are virtually free from *Salmonella* spp. except for the endemic occurrence of *S. enterica* subsp. *diarizonae* in sheep. To document this favourable situation, Norway runs an extensive surveillance programme that covers both live animals (cattle, pigs and poultry) and meat (cattle, pigs,

sheep and poultry). The *Salmonella* isolates examined in NORM-VET include those that are detected in this programme, in addition to isolates from other relevant projects as well as clinical submissions to the National Veterinary Institute. The data are presented in Table 17 and in the text.

TABLE 17. Antimicrobial resistance in *Salmonella* spp. (n=47); *S.* Typhimurium (n=22) and other *Salmonella* spp. (n=25) from animals.

Substance	Resistance (%)	[95%CI*]	Distribution of MIC values (mg/L)														
			0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	0	[0.0-7.6]						70.2	29.8								
Chloramphenicol	0	[0.0-7.6]							19.1	78.7	2.1						
Florfenicol	0	[0.0-7.6]								91.5	8.5						
Ampicillin	0	[0.0-7.6]					6.4	74.5	17.0	2.1							
Ceftiofur	0	[0.0-7.6]					27.7	70.2	2.1								
Trimethoprim	0	[0.0-7.6]				36.2	55.3	8.5									
Sulfamethoxazole	0	[0.0-7.6]										19.1	12.8	57.4	8.5	2.1	
Streptomycin	0	[0.0-7.6]							2.1	23.4	53.2	21.3					
Gentamicin	0	[0.0-7.6]					61.7	38.3									
Neomycin	0	[0.0-7.6]						100									
Enrofloxacin	2	[0.5 -14.5]		34.0	59.6	4.3	2.1										
Nalidixic acid	2	[0.5 -14.5]						6.4	89.4	2.1				2.1			

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.

*CI = Confidence interval.

COMMENTS

In 2004, only five isolates of *S.* Typhimurium from two cattle and three pigs, and one isolate of *S.* Senftenberg from poultry were detected in the national surveillance programme. The other *Salmonella* isolates tested were from diagnostic submissions. The isolates of *S.* Typhimurium were from one dog, one pigeon, ten wild animals and five wild birds. Twelve isolates of *S. enterica* ss. *diarizonae* (occurs endemically in the Norwegian sheep

population) were from sheep, goat and geese and the other *Salmonella* spp. were from wild animals and reptiles. Only one isolate, *S.* Newport from a snake, was resistant to enrofloxacin and nalidixid acid. All other isolates were susceptible to all animicrobials included.

The data, although very limited, indicate that antimicrobial resistance is not very widespread among those *Salmonella* that sometimes are isolated from Norwegian animals.

Salmonella from human clinical specimens

In 2004, 1567 human cases of salmonellosis, excluding typhoid and paratyphoid fever, were reported in Norway (incidence rate 34.2 per 100,000). In 72% of the cases, the infection was reported as having been acquired abroad, whereas for 22% the infection was classified as domestically acquired. For the remaining cases, the place of acquisition was unknown.

For *S*. Enteritidis (51% of the cases), the proportion of cases reported as imported was particularly high (86%). *S*. Enteritidis has never been detected in Norwegian poultry. For *S*. Typhimurium (13% of the cases), approximately 42% of the infections were acquired in Norway, which is partly explained by the endemic occurrence of specific clones of this serovar in Norwegian wildlife. Data from surveillance programmes show that domestically produced food is not an important source of human salmonellosis acquired in Norway. The most likely sources of infection in these cases are wildlife, imported food products or other patients infected abroad. Thus, the isolates categorized as "infected in Norway" also partly reflect the resistance situation outside Norway.

The incidence of multiresistant *S*. Typhimurium DT104 infection, especially from domestically aquired cases, decreased markedly in 2003 as compared to 2002, and has remained at the same low level in 2004. For the *S*. Typhimurium infections acquired domestically, none of the isolates were caused by multiresistant DT104, as opposed to 11 (11%) for those *S*. Typhimurium infections acquired abroad.

In 2004, 192 isolates of *S*. Typhimurium, 750 isolates of *S*. Enteritidis, 15 isolates of *S*. Typhi, 16 isolates of *S*. Paratyhi A, 4 isolates of *S*. Paratyphi B, and 449 isolates of other *Salmonella* spp. were susceptibility tested. The results are presented in Figure 15, Tables 18-25, and in the text.

TABLE 18. *Salmonella* Typhimurium isolates (n=78) from patients infected in Norway. Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	79.5	0	20.5	6	-	28
Chloramphenicol	≥ 20	≤ 19	92.3	-	7.7	6	-	≥ 36
Ampicillin	≥ 32	≤ 12	0	82.1	17.9	6	-	30
TMS**	≥ 20	≤ 12	93.6	0	6.4	6	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	98.7	1.3	0	25	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	97.4	-	2.6	6	-	28

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.

TABLE 19. *Salmonella* Typhimurium isolates (n=78) from patients infected in Norway. Distribution (%) of zone diameters (mm).

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	15.4	1.3		2.6							1.3					
Chloramph.	7.7															
Ampicillin	17.9															1.3
TMS*	6.4															
Ciprofloxacin																
Nalidixic acid	2.6														2.6	2.6

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline		5.1	20.5	15.4	29.5	6.4	2.6								
Chloramph.			11.5	16.7	28.2	19.2	14.1		1.3						1.3
Ampicillin		2.6	6.4	7.7	14.1	16.7	17.9	9.0	6.4						
TMS*			1.3	1.3	1.3		2.6		1.3	2.6	6.4	11.5	21.8	11.5	32.1
Ciprofloxacin			1.3				1.3		1.3		5.1	2.6	1.3	5.1	82.1
Nalidixic acid		3.8	12.8	26.9	29.5	11.5	7.7								

Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
**TMS=Trimethoprim/sulfamethoxazole.

TABLE 20. *Salmonella* Typhimurium isolates (n=100) including DT104 (n=11) from patients infected outside Norway. Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	55.0	0.0	45.0	6	-	32
Chloramphenicol	≥ 20	≤ 19	71.0	-	29.0	6	-	≥ 36
Ampicillin	≥ 32	≤ 12	0.0	63.0	37.0	6	-	30
TMS**	≥ 20	≤ 12	90.0	0.0	10.0	6	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	98.0	2.0	0.0	24	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	76.0	-	24.0	6	-	30

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.

TABLE 21. *Salmonella* Typhimurium isolates (n=100) including DT104 (n=11) from patients infected outside Norway. Distribution (%) of zone diameters (mm).

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	23.0	4.0	3.0	4.0	6.0	2.0	1.0		1.0	1.0					1.0	
Chloramph.	26.0		2.0							1.0						
Ampicillin	37.0									1.0						
TMS*	9.0				1.0											
Ciprofloxacin																
Nalidixic acid	21.0						1.0			1.0	1.0					1.0

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	2.0	1.0	11.0	11.0	11.0	9.0	7.0	1.0			1.0				
Chloramph.		1.0	6.0	12.0	21.0	15.0	15.0								1.0
Ampicillin	2.0		5.0	4.0	10.0	10.0	17.0	11.0	3.0						
TMS*				1.0	1.0	4.0	1.0	5.0	8.0	10.0	7.0	26.0	8.0	19.0	
Ciprofloxacin			1.0	1.0		5.0	5.0	4.0	8.0			2.0	6.0	5.0	63.0
Nalidixic acid	2.0	1.0	14.0	18.0	15.0	15.0	8.0	1.0	1.0						

Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
*TMS=Trimethoprim/sulfamethoxazole.

TABLE 22. *Salmonella* Enteritidis isolates from patients (n=750[#]). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	97.1	0.1	2.8	6	-	34
Chloramphenicol	≥ 20	≤ 19	99.7	-	0.3	19	-	≥ 36
Ampicillin	≥ 32	≤ 12	0.8	93.6	5.6	6	-	≥ 36
TMS**	≥ 20	≤ 12	97.9	0.1	2	6	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	98.3	1.6	0.1	18	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	73.6	-	26.4	6	-	30

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.
Place of infection; Norway (n=66), abroad (n=654), unknown (n=30).

TABLE 23. *Salmonella* Enteritidis isolates from patients (n=750[#]). Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	1.2	1.1	0.4	0.1										0.1	0.5	0.4
Chloramph.														0.3	0.3	
Ampicillin	5.6							0.1			0.1		0.1		0.1	0.5
TMS*	2.0											0.1				
Ciprofloxacin													0.1			
Nalidixic acid	26.3								0.1				0.3		0.1	0.4

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	1.9	1.7	8.0	8.9	20.4	21.6	23.3	4.3	5.2	0.5	0.1		0.1		
Chloramph.	0.3	0.8	5.3	11.1	29.6	26.9	18.5	2.9	1.9	0.3	0.9	0.1	0.1	0.1	0.5
Ampicillin	1.1	0.8	7.5	4.8	16.8	21.9	28.1	7.9	3.3	0.4	0.5	0.1			0.1
TMS*			0.1	0.1			0.7	0.1	2.0	1.7	10.4	8.0	25.6	18.7	30.4
Ciprofloxacin			0.4	1.2	2.1	7.1	4.0	9.3	1.5	1.1	0.9	4.8	5.3	62.1	
Nalidixic acid	1.1	2.7	12.0	19.6	19.5	10.3	6.5	0.7	0.5						

Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
*TMS=Trimethoprim/sulfamethoxazole.
Place of infection; Norway (n=66), abroad (n=654), unknown (n=30).

TABLE 24. *Salmonella* spp. (excluding *S*. Typhimurium, *S*. Enteritidis, *S*. Typhi and *S*. Paratyphi) (n=449[#]). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)	
	S	R	S	I	R		
Tetracycline	≥ 20	≤ 16	69.7	0.4	29.8	6 -	30
Chloramphenicol	≥ 20	≤ 19	91.8	-	8.2	6 -	≥ 36
Ampicillin	≥ 32	≤ 12	1.3	84.6	14	6 -	≥ 36
TMS**	≥ 20	≤ 12	88.4	0.4	11.1	6 -	≥ 36
Ciprofloxacin	≥ 27	≤ 18	91.8	6.7	1.6	12 -	≥ 36
Nalidixic acid	≥ 17	≤ 16	77.7	-	22.3	6 -	≥ 36

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.
Place of infection; Norway (n=83), abroad (n=340), unknown (n=26).

TABLE 25. *Salmonella* spp. (excluding *S*. Typhimurium, *S*. Enteritidis, *S*. Typhi and *S*. Paratyphi) (n=449[#]). Distribution (%) of zone diameters (mm).

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	13.6	9.4	4.5	0.9	0.4	0.7		0.2	0.2				0.2	0.2	1.1	1.8
Chloramph.	5.1	0.9	0.2	0.7	0.2	0.2		0.2				0.2	0.2	0.2	0.2	0.2
Ampicillin	14.0									0.4		0.7	0.2		0.2	
TMS*	10.9				0.2			0.2				0.2				0.2
Ciprofloxacin							0.7	0.7	0.2						0.4	
Nalidixic acid	20.7					0.2		0.2	0.4	0.4	0.2	0.2	0.2	0.4	0.2	0.2

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	2.2	4.0	7.8	12.5	14.0	12.5	11.6	1.6	0.7						
Chloramph.	1.3	3.6	10.5	18.7	25.8	16.0	10.5	1.8	1.1	0.7	0.4		0.4		0.4
Ampicillin	1.3	1.3	6.5	4.9	13.4	12.7	22.7	10.9	8.5	0.9	0.7	0.2			0.4
TMS*		0.2	0.2		0.4	0.4	2.0	2.0	7.6	2.9	12.7	11.4	20.0	10.0	18.3
Ciprofloxacin	0.4	0.2	1.3	2.0	2.2	2.2	6.0	2.4	2.0	1.1	1.8	0.4	8.5	6.9	60.4
Nalidixic acid	1.6	1.6	10.5	16.9	19.4	12.5	8.9	2.9	2.0						0.2

Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
*TMS=Trimethoprim/sulfamethoxazole. # Place of infection; Norway (n=83), abroad (n=340), unknown (n=26).

RESULTS AND COMMENTS

For *S*. Typhimurium, resistance to tetracycline was most common followed by resistance to ampicillin, chloramphenicol, nalidixic acid and trimethoprim/ sulfamethoxazole.

The proportion of *S*. Typhimurium isolates susceptible to all antimicrobial agents was higher for the category "infected in Norway" (79.5%) than for the "infected abroad" category (48.3%) (Figure 15). Moreover, multiresistance (resistance to more than two antimicrobial agents) was more common in the category "infected abroad" (24.7%) as compared to the category "infected in Norway" (14.1%). A significant discrepancy for the two categories was observed for quinolones; in the category "infected abroad", 24.0% of the isolates were resistant to nalidixic acid as opposed to 2.6% among those from patients "infected in Norway". None of the isolates showed reduced susceptibility to ciprofloxacin.

The vast majority of *S*. Enteritidis isolates had been acquired abroad. The proportion of *S*. Enteritidis isolates resistant to the different antimicrobial agents included except for nalidixid acid was considerably lower than for *S*. Typhimurium. In total, 26.6% of the isolates of *S*. Enteritidis were resistant to nalidixid acid. Altogether 0.1% were resistant to ciprofloxacin, whereas intermediate susceptibility to ciprofloxacin was observed in 1.6% of the isolates. All but two of the isolates intermediately susceptible to ciprofloxacin were also resistant to nalidixic acid. The resistance frequencies observed for *S*.

Typhimurium and *S*. Enteritidis in NORM/NORM-VET 2004 are quite similar to those observed in previous reports. There may be some indications that imported cases of *S*. Typhimurium may more often be multiresistant and that resistance to nalidixid acid is increasing compared to 2003.

The few isolates of *S*. Typhi (n=15), *S*. Paratyphi A (n=16) and *S*. Paratyhi B (n=4) detected and susceptibility tested in 2004 indicate that multiresistance is common in *S*. Typhi and *S*. Paratyphi A, and that these isolates are commonly resistant to nalidixid acid. The majority of these infections had been acquired outside Norway and only three cases were aquired domestically. Of the isolates of *S*. Paratyphi A and *S*. Typhi, eight and 11 isolates, respectively, were resistant to one or more of the antimicrobial agents included. Of the 31 isolates of *S*. Paratyphi A and *S*. Typhi, nine isolates were resistant to nalidixid acid, six isolates to each of the antimirobials oxytetracycline, chloramphenicol and sulfamethoxazole, respectively and five isolates were resistant to ampicillin. Among the isolates of *S*. Paratyphi B, two were resistant to chloramphenicol, both from cases aquired domestically.

With regard to isolates of *Salmonella* spp. other than *S*. Typhimurium, *S*. Enteritidis, *S*. Typhi and *S*. Paratyphi, the vast majority of infections had been acquired abroad. Resistance was quite widespread. Resistance to

tetracycline was most common, followed by resistance to nalidixic acid, ampicillin, trimethoprim-sulfamethoxazole, and chloramphenicol. The prevalence of resistance to nalidixic acid was relatively high (22.3%). Similar to what was observed for *S.* Enteritidis isolates, ciprofloxacin

resistance was observed in 1.6%, while 6.7% showed reduced susceptibility to ciprofloxacin indicating that fluoroquinolone resistance is developing.

It is emphasized that the use of fluoroquinolones in Norway is limited in both human and veterinary medicine.
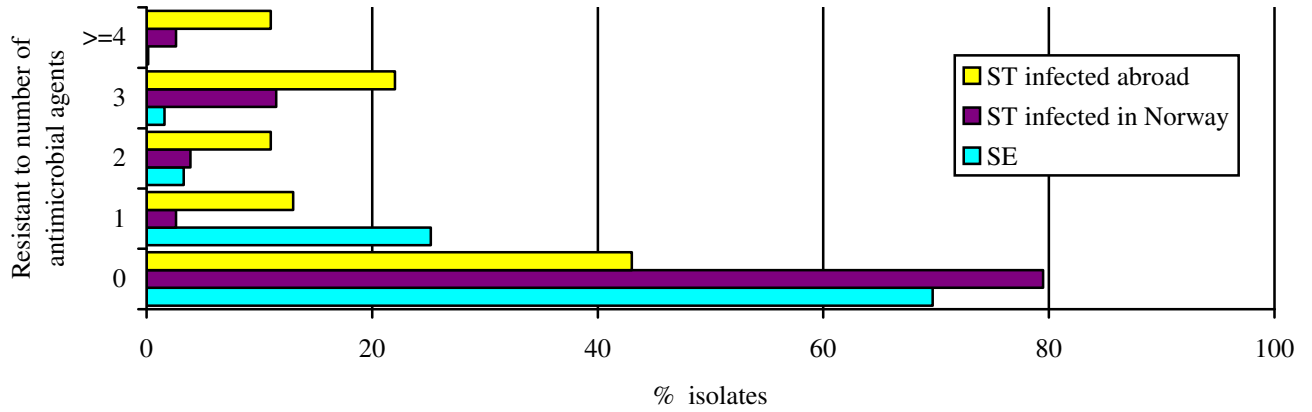

FIGURE 15. Antimicrobial resistance profiles for all *Salmonella* Enteritidis (SE) from humans (n=750) and for *Salmonella* Typhimurium (ST) from humans infected in Norway (n=78) and infected abroad (n=100). Proportion of isolates susceptible to all antimicrobial agents included and resistant to one, two, three, or four or more antimicrobial agents.

CAMPYLOBACTER SPP.

Campylobacter jejuni from broilers

The isolates of *Campylobacter jejuni* in broilers originate from the Norwegian action plan against *Campylobacter* spp. in broilers. All broiler flocks slaughtered before 50 days of age are tested for the presence of *Campylobacter* spp. In addition, 100 samples of broiler meat products from retail level are tested monthly. In 2004, one isolate per positive farm as well as one isolate from each batch of positive broiler meat products were submitted for susceptibility testing.

A total of 108 isolates, 75 from broiler flocks (faecal or cloacal samples) and 33 from broiler meat, were susceptibility tested. The results are presented in Table 26, Figure 16 and in the text.

TABLE 26. Antimicrobial resistance in *Campylobacter jejuni* (n=108); from broiler flocks (n=75) and from broiler meat products (n=33).

Substance	Resistance (%)	[95% CI*]	Distribution (%) of MIC values (mg/L)														
			0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	<1	[0.0-5.1]				97.2	1.9				0.9						
Ampicillin	6	[2.1-11.7]						13.0	31.5	38.9	8.3	2.8	1.9	1.9	1.9		
Erythromycin	0	[0.0-3.4]				6.5	52.8	36.1	4.6								
Gentamicin	0	[0.0-3.4]				5.6	56.5	35.2	2.8								
Enrofloxacin	0	[0.0-3.4]	1.9	36.1	52.8	9.3											
Nalidixic acid	0	[0.0-3.4]							1.9	47.2	50.0	0.9					

Bold vertical lines denote microbiological cut-off values. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested.
*CI = Confidence interval..

RESULTS AND COMMENTS

The results show that the occurrence of resistance among *C. jejuni* isolates from Norwegian broilers is low. A total of 93.5% of the isolates tested were susceptible to all antimicrobial agents included. Altogether 6.5% were resistant to one antimicrobial agent (ampicillin or oxytetracycline). The results reflect the usage of antimicrobial agents in poultry production. Antimicrobial agents (except coccidiostats) are rarely used, and only for therapeutical purposes. If used, amoxicillin (cross-resistance with ampicillin) and tetracycline are the drugs of choice.

The results are similar to those presented in previous NORM/NORM-VET reports (2001, 2002 and 2003).
The level of resistance and the resistance patterns for *C. jejuni* isolated from Norwegian broilers are quite similar to what was observed for *C. jejuni* isolated from humans infected within Norway, except for a higher prevalence of resistance to quinolones (nalidixic acid and ciprofloxacin) among the human isolates. This relationship was also observed in NORM/NORM-VET 2001, 2002 and 2003 (see textbox "Antimicrobial resistance in *Campylobacter jejuni* from humans and broilers in Norway" on page 45) and Figure 16.

Campylobacter spp. from human clinical specimens

Of the 2275 cases of human campylobacteriosis recorded in Norway in 2004 (incidence rate 49.8 per 100,000), 49% were reported as acquired abroad. The vast majority of cases were sporadic. Norwegian case-control studies have revealed that consumption of broiler meat purchased fresh and drinking untreated water are important risk factors for domestically acquired campylobacteriosis.

A total of 232 isolates of *C. jejuni*, 104 from patients infected in Norway and 128 from patients infected abroad, as well as 14 isolates of *C. coli* were susceptibility tested. The results are presented in Tables 27-30, Figure 16 and in the text.

TABLE 27. *Campylobacter jejuni* isolates from patients infected in Norway (n=104). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)			MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R					
Doxycycline	≤ 2	≥ 4	94.2	0	5.8	0.047	-	256	0.19	0.5
Erythromycin	≤ 0.5	≥ 8	8.7	87.5	3.8	0.047	-	256	1	2
Gentamicin	≤ 4	≥ 8	97.1	0	2.9	0.023	-	24	0.5	2
Ciprofloxacin	≤ 1	≥ 4	91.3	0	8.7	0.064	-	32	0.19	0.5
Nalidixic acid	≤ 16	≥ 32	90.4	0	9.6	1	-	256	4	16

*S=Susceptible, I=Intermediately susceptible, R=Resistant.

TABLE 28. *Campylobacter jejuni* isolates from patients infected in Norway (n=104). Distribution (%) of MICs (mg/L).

Substance	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Doxycycline		2.9	27.9	44.2	17.3	1.0	1.0		1.0		1.0	1.0		2.9
Erythromycin		1.0		1.0	6.8	50.9	32.7	3.8		1.0				2.9
Gentamicin	1.0	1.0	5.7	14.4	33.7	27.0	12.5	1.9	1.0		1.9			
Ciprofloxacin		1.0	26.0	51.0	12.5	1.0					8.7			
Nalidixic acid						1.0	14.4	51.0	19.3	4.8	1.0			8.7

Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.

TABLE 29. *Campylobacter jejuni* isolates from patients infected outside Norway (n=128). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)			MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R					
Doxycycline	≤ 2	≥ 4	40.6	0	59.4	0.032	-	256	6	64
Erythromycin	≤ 0.5	≥ 8	21.1	75.0	3.9	0.064	-	256	1	2
Gentamicin	≤ 4	≥ 8	96.9	0	3.1	0.064	-	12	0.25	1.5
Ciprofloxacin	≤ 1	≥ 4	30.5	0.8	68.8	0.064	-	32	32	32
Nalidixic acid	≤ 16	≥ 32	31.2	0	68.8	1	-	256	256	256

*S=Susceptible, I=Intermediately susceptible, R=Resistant.

TABLE 30. *Campylobacter jejuni* isolates from patients infected outside Norway (n=128). Distribution (%) of MICs (mg/L).

Substance	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Doxycycline	0.8	1.6	10.0	14.8	8.6	1.6	3.1	7.8	5.4	14.0	18.0	7.0	3.9	3.1
Erythromycin		0.8	0.8	5.5	14.1	42.2	27.4	5.5	2.4	0.8				0.8
Gentamicin		5.5	23.5	23.5	28.9	7.8	4.7	3.2	2.4	0.8				
Ciprofloxacin		2.3	10.1	13.3	3.9	0.8	0.8			1.6	67.2			
Nalidixic acid						1.6	14.1	9.4	5.5	0.8	0.8	0.8		67.2

Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.

RESULTS AND COMMENTS

The data show that resistance was significantly more widespread among *C. jejuni* isolates derived from patients infected abroad as opposed to patients infected in Norway. Only 19.5% of the isolates in the first category were susceptible to all antimicrobial agents included as opposed to 88.5% of the isolates from patients infected in Norway (Figure 16). These discrepancies are explained by the widespread occurrence among isolates acquired abroad as opposed to patients infected in Norway of resistance to ciprofloxacin/nalidixic acid (68.8%/68.8% versus 8.7%/9.6%) and to tetracycline (59.4% versus 5.8%) (Tables 27 and 29 and Figure 16).

The level of resistance and the resistance patterns for *C. jejuni* isolated from humans infected within Norway correspond quite well with what was observed for *C. jejuni* isolated from Norwegian broilers, except for a higher prevalence of resistance to quinolones (nalidixic acid and ciprofloxacin) among the human isolates. This relationship was also observed in NORM/NORM-VET 2001, 2002 and 2003. See textbox "Antimicrobial resistance in *Campylobacter jejuni* from humans and broilers in Norway" on page 45 and Figure 16.

Only two of the 14 isolates of *C. coli* were acquired in Norway. In total, 12 of the *C. coli* isolates were resistant to at least one of the antimicrobial agents included (mainly quinolones and doxycycline). Two isolates were resistant only to doxycycline. Ten isolates were resistant to both nalidixic acid and ciprofloxacin and six of these were also resistant to doxycycline. Of the latter category, two isolates were in addition resistant to erythromycin. *C. coli* is typically associated with pigs and pork.
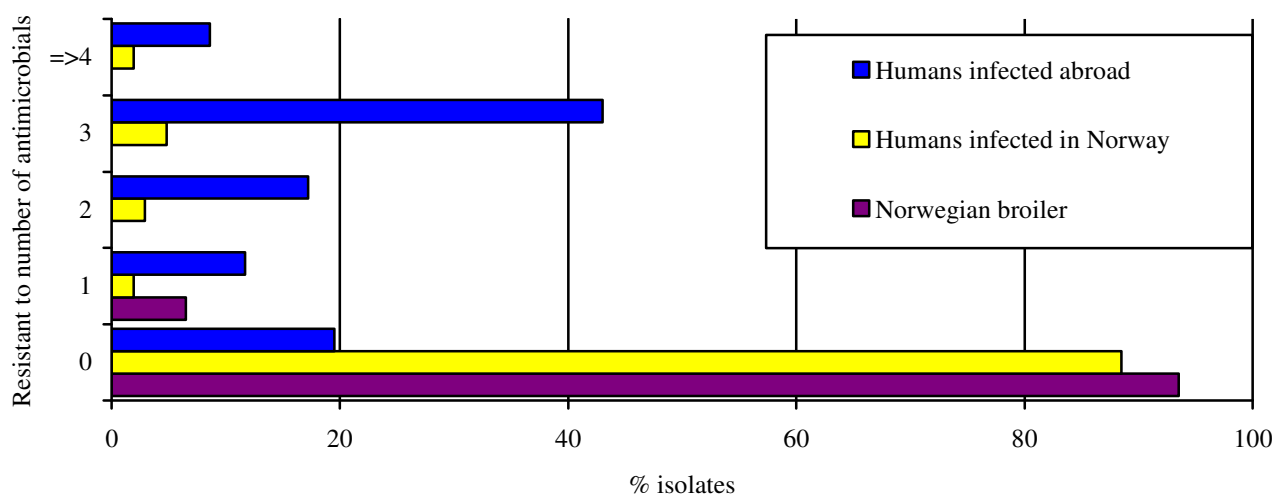
FIGURE 16. Antimicrobial resistance profiles for *Campylobacter jejuni* from Norwegian broiler (n=108) (cloacal/faecal samples (n=75) and meat products (n=33)), humans infected in Norway (n=104) and humans infected abroad (n=128). Proportion of isolates susceptible to all antimicrobial agents included or resistant to one, two, three, or four or more antimicrobial agents. The isolates from humans were tested for susceptibility to doxycycline, erythromycin, gentamicin, ciprofloxacin and nalidixic acid, whereas the broiler and broiler meat isolates in addition were tested for susceptibility to ampicillin (and to oxytetracycline rather than doxycycline).

Antimicrobial resistance in *Campylobacter jejuni* from humans and broilers in Norway

Fluoroquinolone-resistant *Campylobacter* species is recognised as an emerging public health problem [1]. Fluoroquinolone resistance in *Campylobacter* spp. is increasing in many countries throughout the world [2-5]. In several countries, an association between the usage of fluoroquinolones in food animals and the occurrence of fluoroquinolone resistant *Campylobacter* from humans has been documented [2]. Fluoroquinolones are commonly used in poultry medicine both for therapeutic and prophylactic purposes. Poultry meat is generally considered as the most important risk factor for campylobacteriosis in humans [4, 6, 7]. Thus, an extended use of fluoroquinolones in poultry production thereby increases the risk of transmission of fluoroquinolone resistant *Campylobacter* spp. to humans. Antimicrobial agents are normally not indicated for treatment of campylobacteriosis in humans. However, severe cases may require treatment. An increased resistance to fluoroquinolones reduces the possibilities to treat severe infections in humans, which can have fatal consequences [1, 8]. Norway has a long tradition for a restrictive antimicrobial policy in animal husbandry, and fluoroquinolones were never licensed for use in poultry.

Campylobacteriosis is currently the most frequently reported cause of bacterial gastroenteritis in Norway. Most cases are sporadic and close to half of the reported cases acquire the infection in Norway. A case-control study conducted in Norway during 1999-2000 identified consumption of untreated drinking water, consumption of poultry meat purchased raw, consumption of barbecued meat, and professional contact with animals as significant risk factors with regard to campylobacteriosis [9]. A recent study study assessed and compared the prevalences of resistance in *Campylobacter jejuni* isolates from imported and indigenous human cases and from Norwegian broilers using data from NORM/NORM-VET for the period 2001-2003 [10].

TABLE 31. Percentage of isolates of *Campylobacter jejuni* from various sources and years classified as resistant to various antimicrobial agents.

Substance	Cut-off Value* mg/L	Norwegian broilers			Humans infected in Norway			Humans infected abroad		
		2001 n=113	2002 n=161	2003 n=139	2001 n=84	2002 n=37	2003 n=63	2001 n=129	2002 n=110	2003 n=144
Doxycycline**	>2	0.9	0.0	2.2	9.5	2.7	9.5	43.4	54.5	62.5
Erythromycin	>2	0.0	1.0	0.0	4.8	2.7	4.8	3.1	6.4	5.6
Gentamicin	>4	0.0	0.0	0.0	0.0	0.0	0.0	0.8	1.8	3.5
Nalidixic acid	>16	2.7	1.9	1.4	8.3	8.1	11.1	59.7	71.8	72.9
Ciprofloxacin***	>2	2.7	0.6	1.4	7.1	2.7	7.9	59.7	71.8	70.8

*Values above the cut-off value indicate resistance
** Oxytetracycline for broilers
***Enrofloxacin for broilers in 2002 and 2003

Resistance was considerably more widespread among *Campylobacter jejuni* from humans infected abroad than from humans infected within Norway. The discrepancy was particularly notable for fluoroquinolone resistance (67.4% versus 6.5%). This is likely a reflection of a low resistance prevalence in Norwegian broiler isolates (in total 1.2% fluoroquinolone resistant).

The study documents that a limited use of fluoroquinolones in food animal production within a country is associated with a low prevalence of fluoroquinolone resistance in indigenous human *C. jejuni* isolates. Thus, the restricted use of fluoroquinolones in food producing animals is recommended in order to keep such antimicrobial agents efficient for the treatment of severe cases of campylobacteriosis in humans.

References:
1. OIE/WHO/FAO, Joint FAO/OIE/WHO Expert Workshop on Non-Human Antimicrobial Usage and Antimicrobial Resistance: Scientific assessment, Geneve, 1-5 December, 2003. World Health Organization, 2004. http://www.who.int/foodsafety/publications/micro/nov2003/en/
2. Engberg J, Aarestrup FM, Taylor DE, Gerner-Smidt P, Nachamkin I. Quinolone and macrolide resistance in *Campylobacter jejuni* and *C. coli*: resistance mechanisms and trends in human isolates. Emerg Infect Dis 2001; **7**: 24-34.
3. Piddock LJV. Quinolone resistance and *Campylobacter* spp. J Antimicrob Chemother 1995; **36**: 891-898.
4. Smith KE, Besser JM, Hedberg CW, Leano FT, Bender JB, Wicklund JH. Quinolone-resistant *Campylobacter jejuni* infections in Minnesota. 1992-1998. N Engl J Med 1999; **340**: 1525-1532.
5. Velázquez JB, Jiménez A, Chomón B, Villa G. Incidence and transmission of antibiotic resistance in *Campylobacter jejuni* and *Campylobacter coli*. J Antimicrob Agents Chemother 1995; **32**: 1170-1173.
6. Friedman CR, Neimann J, Wegener HC, Tauxe RV. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. In: Nachamkin I, Blaser MJ. *Campylobacter* 2nd edition. ASM press. Washington DC. 2001; 121-138.
7. WHO (World Health Organization). The increasing incidence of human campylobacteriosis. Report and proceedings of a WHO consultation of experts. Copenhagen, Denmark, 21-25 November 2000. World Health Organization, 2001. http://www.who.int/emc-documents/zoonoses/whocdscsraph20017c.html
8. Helms M, Vastrup P, Gerner-Smidt P, Molbak K. Excess mortality associated with antimicrobial drug-resistant *Salmonella Typhimurium*. Emerg Infect Dis 2002; **8**: 490-495.
9. Kapperud G, Espeland G, Wahl E et al. Factors associated with increased and decreased risk of *Campylobacter* infection: a prospective case-control study in Norway. Am J Epidemiol 2003; **158**: 234-242.
10. Norström M, Hofshagen M, Stavnes T, Schau J, Lassen J and Kruse H. Antimicrobial resistance in *Campylobacter jejuni* from humans and broilers in Norway. Epid and Infection (in press)

Madelaine Norström, Jørgen Lassen and Hilde Kruse

Yersinia enterocolitica from human clinical specimens

Most cases of *Yersinia enterocolitica* infection in Norway are normally domestically acquired. However, in 2004, 36.5% of the 96 reported cases were classified as imported, whereas this proportion was 24% and 19% in 2002 and 2003, respectively.

In 2004, a total of 81 isolates of *Y. enterocolitica* O:3 were susceptibility tested. The results are presented in Tables 32 and 33.

TABLE 32. *Yersinia enterocolitica* serogroup 0:3 isolates from human clinical cases (n=81[#]). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	93.8	0	6.2	6	-	≥ 36
Chloramphenicol	≥ 20	≤ 19	86.4	-	13.6	6	-	≥ 36
Ampicillin	≥ 32	≤ 12	0	0	100	6	-	11
TMS**	≥ 20	≤ 12	86.4	12.3	1.2	9	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	96.3	3.7	0	23	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	96.3	-	3.7	6	-	≥ 36

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.
[#] Place of infection; Norway (n=41), Abroad (n=33), Unknown (n=7).

TABLE 33. *Yersinia enterocolitica* serogroup 0:3 isolates (n=81[#]) from human clinical cases. Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	6.2														1.2	1.2
Chloramph.	13.6															
Ampicillin	61.7	11.1	19.8	6.2		1.2										
TMS**				1.2						1.2	2.5	4.9	2.5	1.2	1.2	
Ciprofloxacin																
Nalidixic acid	3.7															1.2

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	2.5	1.2		2.5	11.1	3.7	17.3	7.4	23.5	6.2	11.1		2.5	1.2	1.2
Chloramph.	1.2		4.9	7.4	9.9	3.7	21	3.7	14.8	2.5	4.9	2.5	4.9	1.2	3.7
Ampicillin															
TMS**	1.2		1.2		7.4		3.7	4.9	12.3	6.2	14.8	12.3	6.2	2.5	12.3
Ciprofloxacin		1.2			2.5		4.9	2.5	4.9	1.2	7.4	6.2	7.4	8.6	53.1
Nalidixic acid	3.7	1.2	3.7	4.9	11.1	11.1	23.5	11.1	11.1	3.7	6.2	1.2	1.2		1.2

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). **TMS=Trimethoprim/sulfamethoxazole.
[#] Place of infection; Norway (n=41), Abroad (n=33), Unknown (n=7).

RESULTS AND COMMENTS

The occurrences of resistance differed from what has been presented in the previous NORM/NORM-VET reports (2000-2003). In total, 6.2% and 13.6% of the isolates in 2004 were resistant to tetracycline and chloramphenicol, respectively, as compared to 0% and 5.2% in 2003. These increases were statistically significant (p<0.05). The occurrence of resistance in the isolates from infections aquired abroad were higher, but not significantly higher, than those aquired domestically. There were no differences in the prevalence of resistance to tetracyclines between isolates from infections aquired abroad and domestically. The higher prevalences of resistance to chloramphenicol and to nalidixid acid observed in 2004 as compared to previous years (2000-2003) are a result of a larger proportion of cases aquired abroad in 2004. In total, 21.2% and 9.1% of the isolates from the infections aquired abroad were resistant to chloramphenicol and nalidixid acid, respectively, compared to 9.8% and 0% of the isolates from domestic cases.

All isolates expressed reduced susceptibility to ampicillin, an intrinsic resistance trait in strains of serogroup O:3.

Shigella spp. from human clinical specimens

It is emphasized that almost all the reported *Shigella* infections in Norway were acquired abroad. In 2004, 14.7% of the reported cases were classified as domestically acquired. Thus, the resistance prevalences reported here predominantly relate to isolates originating in other countries. The distribution of the *Shigella* species was as follows: *S. sonnei* 67 (49%), *S. flexneri* 52 (38%), *S. boydii* 12 (9%), and *S. dysenteriae* 5 (3%). The results for *S. sonnei* and *S. flexneri* are presented in Tables 34-37 and in the text.

TABLE 34. *Shigella sonnei* isolates from human clinical cases (n=67). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	19.4	0	80.6	6	-	33
Chloramphenicol	≥ 20	≤ 19	97.0	-	3.0	6	-	32
Ampicillin	≥ 32	≤ 12	1.5	85.1	13.4	6	-	≥ 36
TMS**	≥ 20	≤ 12	9.0	6.0	85.1	6	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	100	0	0	27	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	88.1	-	11.9	6	-	34

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.

TABLE 35. *Shigella sonnei* isolates from human clinical cases (n=67). Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	76.1	3.0		1.5												
Chloramph.	1.5											1.5			1.5	
Ampicillin	13.4								1.5			1.5	10.4	11.9	28.4	19.4
TMS**	82.1	1.5				1.5			1.5		1.5		3.0		1.5	
Ciprofloxacin																
Nalidixic acid	7.5	3.0	1.5										1.5			

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline		3.0	1.5	4.5	6.0	1.5	1.5					1.5			
Chloramph.			4.5	11.9	28.4	19.4	10.4	3.0	13.4	3.0	1.5				
Ampicillin	10.4			1.5											1.5
TMS**						1.5					1.5		3.0		1.5
Ciprofloxacin						3.0	4.5			3.0	1.5		1.5		86.6
Nalidixic acid				1.5	3.0	4.5	19.4	20.9	22.4	6.0	6.0	1.5	1.5		

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
**TMS=Trimethoprim/sulfamethoxazole.

TABLE 36. *Shigella flexneri* isolates from human clinical cases (n=52). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	7.7	0	92.3	6	-	32
Chloramphenicol	≥ 20	≤ 19	19.2	-	80.8	6	-	≥ 36
Ampicillin	≥ 32	≤ 12	0	19.2	80.8	6	-	30
TMS**	≥ 20	≤ 12	21.2	0	78.8	6	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	96.2	3.8	0	25	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	86.5	-	13.5	6	-	34

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.

TABLE 37. *Shigella flexneri* isolates from human clinical cases (n=52). Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	90.4	1.9														
Chloramph.	42.3	9.6	7.7	1.9	7.7	3.8	5.8	1.9								
Ampicillin	75.0	1.9	1.9	1.9											1.9	
TMS**	76.9	1.9														
Ciprofloxacin																
Nalidixic acid	13.5															

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline				1.9			1.9		1.9		1.9				
Chloramph.				1.9								3.8	1.9		11.5
Ampicillin						5.8	3.8	3.8	3.8						
TMS**						3.8	1.9		1.9	1.9	1.9				9.6
Ciprofloxacin				1.9	1.9	3.8	1.9				1.9				88.5
Nalidixic acid			1.9		1.9	1.9	11.5	19.2	21.2	11.5	7.7	7.7	1.9		

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
**TMS=Trimethoprim/sulfamethoxazole.

RESULTS AND COMMENTS

As is the case in reports from other countries, resistance was widespread among *Shigella* isolates, regardless of the species. The resistance frequencies were particularly high for trimethoprim/sulfamethoxazole and tetracycline, followed by ampicillin and chloramphenicol. These drugs are commonly used for various clinical purposes within human medicine in many parts of the world.

For ampicillin and chloramphenicol there were species differences, as resistance was highly prevalent among *S. flexneri* and less prevalent among *S. sonnei*. Resistance to nalidixic acid was relatively common among both *S. flexneri* and *S. sonnei*. Resistance to fluoroquinolones was rarely observed, but the detection of *Shigella* isolates intermediately susceptible to ciprofloxacin and resistant to nalidixic acid may indicate that fluoroquinolone resistance is developing.

D. BACTERIA FROM HUMAN CLINICAL SPECIMENS

Distribution of bacterial species in blood cultures

Surveillance of antimicrobial resistance is usually based on prevalences of reduced susceptibility and resistance to certain combinations of antimicrobials and bacterial species in clinical samples. The NORM surveillance programme generally follows this approach. However, there is a serious limitation to the model because transition from susceptible bacterial species to inherently more resistant ones will represent de facto emergence of resistance which is not detected. In order to complement the surveillance of individual species, NORM 2004 therefore included registration of all positive blood cultures in the laboratory information systems of the participants. A patient with a given microbial isolate was excluded from registration with a new isolate of the same identity within a month from the first entry. This rule was applied irrespective of changes in resistance pattern. There

were no restrictions concerning registration with a different pathogen. It proved difficult to systematically evaluate the clinical significance of isolates from the skin flora. In Table 38, proportions are therefore estimated from all isolates and from all isolates excluding common skin contaminants such as coagulase negative staphylococci, *Micrococcus* spp, *Corynebacterium* spp., *Bacillus* spp. and *Propionibacterium* spp. This does not imply that such isolates should be disregarded in all instances, but only that it was not possible to enforce a standardised protocol for inclusion of the clinically significant isolates. Similarly, all isolates were registered as individual finding although polymicrobial bloodstream infections are regularly detected in the laboratories. Again, limitations of the data extraction procedure prohobited in-depth analysis of these parameters.

TABLE 38. Number of blood culture isolates by species, and proportion of all isolates and proportion of isolates excluding possible skin contaminants (Coagulase negative staphylococci, *Micrococcus* spp., *Bacillus* spp., *Corynebacterium* spp. and *Propionibacterium* spp.). Gram positive bacteria.

Species	No. of isolates	% of all isolates	% excl. contaminants
Staphylococcus aureus	1053	12.3	14.0
Coagulase negative staphylococci	967	11.3	-
Micrococcus spp.	14	0.2	-
Streptococcus pneumoniae	994	11.6	13.2
Streptococcus pyogenes	198	2.3	2.6
Streptococcus agalactiae	171	2.0	2.3
Streptokokker. betahaemolytic group C and G	64	0.7	0.9
Streptococcus spp.. viridans- and non-haemolytic	397	4.6	5.3
Enterococcus faecalis	390	4.6	5.2
Enterococcus faecium	93	1.1	1.2
Enterococcus spp.	27	0.3	0.4
Lactococcus lactis	3	< 0.1	< 0.1
Aerococcus spp.	7	0.1	0.1
Abiotrophia spp.	6	0.1	0.1
Leuconostoc spp.	1	< 0.1	< 0.1
Gemella haemolysans	1	< 0.1	< 0.1
Bacillus spp.	12	0.1	-
Paenibacillus glucanolyticus	3	< 0.1	< 0.1
Listeria monocytogenes	19	0.2	0.3
Erysipelothrix rhusiopathiae	1	< 0.1	< 0.1
Corynebacterium spp.	47	0.5	-
Rothia mucilaginosus	1	< 0.1	< 0.1
Arcanobacterium spp.	1	< 0.1	< 0.1
Gardnerella vaginalis	1	< 0.1	< 0.1
Unidentified gram positive coccus	7	< 0.1	0.1
Total aerobic Gram positive bacteria	**4478**	**52.3**	**45.8**

TABLE 38 continued. Number of blood culture isolates by species, and proportion of all isolates and proportion of isolates excluding possible skin contaminants (Coagulase negative staphylococci, *Micrococcus* spp., *Bacillus* spp., *Corynebacterium* spp. and *Propionibacterium* spp.). Gram negative bacteria.
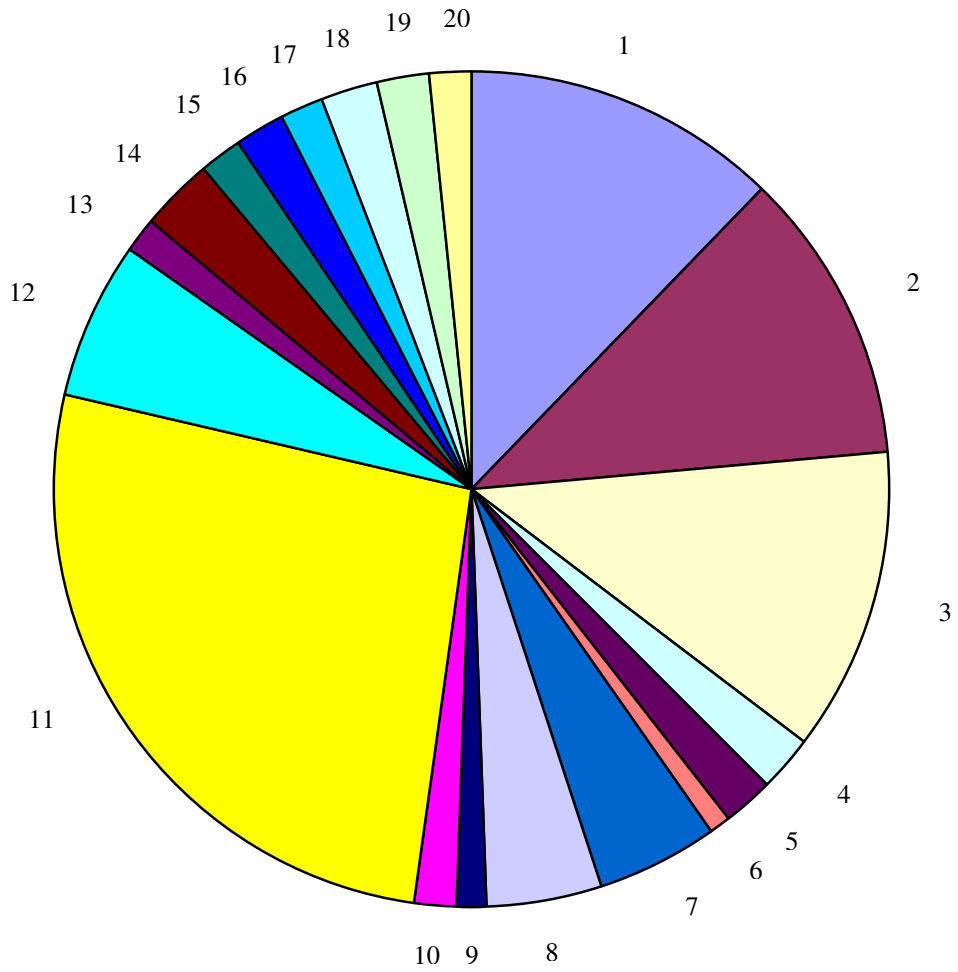
Species	No. of isolates	% of all isolates	% excl. contaminants
Neisseria meningitidis	11	0.1	0.1
Neisseria spp.	2	< 0.1	< 0.1
Moraxella spp.	6	0.1	0.1
Actinobacillus spp.	3	< 0.1	< 0.1
Capnocytophaga spp.	1	< 0.1	< 0.1
Eikenella corrodens	2	< 0.1	< 0.1
Pasteurella multocida	3	< 0.1	< 0.1
Cardiobacterium hominis	2	< 0.1	< 0.1
Haemophilus influenzae	47	0.5	0.6
Haemophilus parainfluenzae	5	0.1	0.1
Escherichia coli	2243	26.2	29.9
Klebsiella pneumoniae	387	4.5	5.2
Klebsiella oxytoca	118	1.4	1.6
Klebsiella spp.	27	0.3	0.4
Enterobacter cloacae	93	1.1	1.2
Enterobacter spp.	32	0.4	0.4
Proteus mirabilis	206	2.4	2.7
Proteus spp.	21	0.2	0.3
Morganella morganii	22	0.2	0.3
Citrobacter spp.	20	0.2	0.3
Hafnia alvei	1	< 0.1	< 0.1
Providencia spp.	2	< 0.1	< 0.1
Serratia spp.	25	0.3	0.3
Salmonella enterica. salmonellosis group	47	0.5	0.6
Salmonella enterica. ssp. Typhi	15	0.2	0.2
Salmonella enterica. ssp. Paratyphi	17	0.2	0.2
Shigella flexneri	2	< 0.1	< 0.1
Yersinia pseudotuberculosis	2	< 0.1	< 0.1
Pseudomonas aeruginosa	141	1.6	1.9
Pseudomonas spp.	21	0.2	0.3
Stenotrophomonas maltophilia	1	< 0.1	< 0.1
Comamonas spp.	2	< 0.1	< 0.1
Delftia acidovorans	1	< 0.1	< 0.1
Chryseomonas luteola	1	< 0.1	< 0.1
Acinetobacter baumannii	26	0.3	0.3
Acinetobacter spp.	16	0.2	0.2
Alcaligenes spp.	3	< 0.1	< 0.1
Sphingomonas spp.	1	< 0.1	< 0.1
Agrobacterium spp.	1	< 0.1	< 0.1
Campylobacter jejuni	4	< 0.1	0.1
Campylobacter spp.	4	< 0.1	0.1
Unidentified Gram negative rod	7	0.1	0.1
Total aerobic Gram negative bacteria	**3591**	**42.0**	**47.8**

TABLE 38 continued. Number of blood culture isolates by species, and proportion of all isolates and proportion of isolates excluding possible skin contaminants (Coagulase negative staphylococci, *Micrococcus* spp., *Bacillus* spp., *Corynebacterium* spp. and *Propionibacterium* spp.). Anaerobes and yeasts.

Species	No. of isolates	% of all isolates	% excl. contaminants
Bacteroides fragilis group	154	1.8	2.1
Bacteroides spp.	24	0.2	0.3
Porphyromonas spp.	6	0.1	0.1
Prevotella spp.	7	0.1	0.1
Fusobacterium spp.	10	0.1	0.1
Veillonella spp.	6	0.1	0.1
Sutterella wadsworthensis	1	< 0.1	< 0.1
Unidentified anaerobic Gram negative rod	2	< 0.1	< 0.1
Clostridium spp.	57	0.7	0.8
Peptostreptococcus spp.	33	0.4	0.4
Peptococcus spp.	8	0.1	0.1
Propionebacterium spp.	10	0.1	-
Lactobacillus	8	0.1	0.1
Actinomyces spp.	2	< 0.1	< 0.1
Eggerthella lenta	4	< 0.1	0.1
Actinobaculum schalii	1	< 0.1	< 0.1
Eubacterium limosum	1	< 0.1	< 0.1
Unidentified anaerobic Gram positive rod	4	< 0.1	0.1
Unidentified anaerobic bacterium	1	< 0.1	< 0.1
Total anaerobic bacteria	**339**	**4.0**	**4.4**
Candida albicans	102	1.2	1.4
Candida dubliniensis	2	< 0.1	< 0.1
Candida glabrata	19	0.2	0.3
Candida kruzei	2	< 0.1	< 0.1
Candida tropicalis	13	0.1	0.2
Candida parapsilosis	5	0.1	0.1
Candida lusitaniae	1	< 0.1	< 0.1
Cryptococcus neoformans	4	< 0.1	0.1
Unidentified yeast	2	< 0.1	< 0.1
Total yeasts	**150**	**1.8**	**2.0**
Total all bacteria and yeasts	**8558**	**100**	**100**

As seen in Table 38 and Figure 17, Gram positive and Gram negative bacteria represented 52.3% and 42.0% of all isolates, respectively. Anaerobic bacteria and yeasts were less common with 4.0% anaerobes and 1.8% yeasts. The difference between Gram positives and Gram negatives was eliminated when all isolates of the predominantly Gram positive skin flora were excluded. *E. coli* was by far the most common pathogen (26.2% of all, 29.9 % excluding skin contaminants) followed by *S. aureus* (12.3% / 14.0%) and *S. pneumoniae* (11.6% / 13.2%). Coagulase negative staphylococci represented 11.3% of the total number of isolates.

FIGURE 17. Distribution of blood culture isolates in Norway 2004 (n=8,558) based on data from the information systems of the participating laboratories.

Legend:
1. Staphylococcus aureus
2. Coagulase negative staphylococci
3. Streptococcus pneumoniae
4. Streptococcus pyogenes
5. Streptococcus agalactiae
6. Streptococcus, betahaemolytic group C and G
7. Streptococcus spp. viridans- and non-haemolytic
8. Enterococcus faecalis
9. Enterococcus faecium
10. Other Gram positive bacteria
11. Escherichia coli
12. Klebsiella spp.
13. Enterobacter spp.
14. Proteus spp.
15. Other Enterobacteriaceae
16. Pseudomonas spp.
17. Other Gram negative bacteria
18. Bacteroides spp.
19. Other anaerobic bacteria
20. Yeasts

Escherichia coli in blood cultures

TABLE 39. *Escherichia coli* blood culture isolates (n=982). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)		MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R				
Ampicillin	≤ 0.5	≥ 16	0.4	67.6	32.0	0.5 -	≥ 256	4	≥ 256
Amoxi/Clav**	≤ 0.5	≥ 16	0.1	89.4	10.5	0.5 -	≥ 256	4	16
Cefotaxime	≤ 1	≥ 8	99.0	0.3	0.7	0.012 -	≥ 256	0.064	0.125
Ceftazidime	≤ 1	≥ 16	98.9	0.5	0.6	0.016 -	≥ 256	0.25	0.5
Cefpirome	≤ 1	≥ 16	99.3	0.3	0.4	0.016 -	≥ 256	0.064	0.125
Ciprofloxacin	≤ 0.5	≥ 2	96.7	0.1	3.2	0.001 -	≥ 32	0.016	0.032
Gentamicin	≤ 2	≥ 8	98.4	0.3	1.3	0.012 -	≥ 256	0.25	0.5
Meropenem	≤ 0.5	≥ 4	100.0	0.0	0.0	0.002 -	0.5	0.032	0.032
Pip/Tazo***	≤ 8	≥ 32	98.2	0.7	1.1	0.012 -	≥ 256	2	4
TMS****	≤ 2	≥ 16	80.9	0.5	18.6	0.008 -	≥ 32	0.125	≥ 32
ESBL			99.3	-	0.7				

*S=Susceptible, I=Intermediately susceptible, R=Resistant.
**Amoxi/Clav=Amoxicillin/clavulanic acid.
***Pip/Tazo=Piperacillin/tazobactam
****TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

TABLE 40. *Escherichia coli* blood culture isolates (n=982). Distribution (%) of MICs (mg/L).*

	≤0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Ampicillin							0.4	3.5	11.5	41.2	11.4	1.5	0.5	0.3	0.6	29.0
Amoxi/Clav**							0.1	2.5	7.9	44.4	34.4	8.4	1.3	0.1		0.7
Cefotaxime		2.0	8.8	41.6	39.8	5.0	1.3	0.4		0.3	0.2		0.2			0.3
Ceftazidime		0.2	0.6	5.4	31.8	49.5	9.8	1.6	0.3		0.1	0.2	0.2	0.1		0.1
Cefpirome		2.4	23.2	52.6	17.8	2.4	0.5	0.2	0.2		0.1		0.1			0.3
Ciprofloxacin	17.6	57.6	16.3	1.4	1.0	1.9	0.7	0.1			0.1	0.1	3.0			
Gentamicin		0.3	0.6	0.7	6.7	45.0	37.0	6.4	1.6	0.2	0.1	0.2	0.4	0.4		0.2
Meropenem	2.1	20.9	73.1	2.6	0.4	0.6	0.2									
Pip/Tazo***		0.2	0.1	0.1	0.1	0.4	2.7	14.5	61.2	16.8	1.9	0.7	0.3	0.3	0.1	0.4
TMS****	0.3	1.4	11.2	30.9	24.9	6.5	4.0	1.0	0.6	0.3	0.2	0.2	18.4			

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.
**Amoxi/Clav=Amoxicillin/clavulanic acid.
***Pip/Tazo=Piperacillin/tazobactam
****TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

The breakpoints for resistance to ampicillin and amoxicillin/clavulanic acid were reduced from ≥ 32 mg/L to ≥ 16 mg/L in 2004. The breakpoint for susceptibility to ampicillin was concomitantly reduced from ≤ 1 mg/L to ≤ 0.5 mg/L, which is the same as for amoxicillin/clavulanic acid. These changes account for the minor changes in SIR distribution seen from 2003 to 2004 with the prevalence of ampicillin susceptibility being reduced from 3.5% to 0.4% and the prevalences of intermediate susceptibility and resistance to amoxicillin/calvulanic acid being reduced from 97.6% to 89.4% and increased from 2.1% to 10.5%, respectively (Table 39 and Figure 18). As can be seen in Table 40, the normal distribution of amoxicillin/clavulanic acid MICs was bisected by the lowered breakpoint of 16 mg/L, and the data consequently suggest a re-evaluation of this adjustment.

The breakpoints for ciprofloxacin were also modified in 2004 as part of the European effort to harmonize breakpoints under the umbrella of EUCAST. The category of intermediate susceptibility has been narrowed by an increase of the lower breakpoint from 0.125 mg/L to 0.5 mg/L and a decrease of the upper breakpoint from 4 mg/L to 2 mg/L. As noted in the NORM/NORM-VET 2003 report there has been an increase in quinolone non-susceptibility over the last 4 years to a level of 5.1% in 2003. The present figure of 3.3% non-susceptibility in 2004 should not be interpreted as a new downward trend as this is only an artefact of changing breakpoints. When the figure from 2003 is recalculated using the new breakpoints it appears that the prevalence of non-susceptibility has actually increased over the last year from 2.4% (0.2% I and 2.2% R) to 3.3% (0.1% I and 3.2% R). Conversely, the data from 2004 would have given a prevalence of non-susceptibility of 5.9% if the old breakpoints had been used, see Figure 19. The trend can also be seen by the increase of the MIC$_{50}$ from 0.002 mg/L

to 0.016 mg/L. The MIC$_{90}$ was maintained at 0.032 mg/L. From a methodological point of view, the strength of collecting continous resistance data as opposed to categorized routine results was demonstrated in this setting of changing breakpoints.

There were no significant changes in the SIR distribution of trimethoprim/sulfamethoxazole, and the vast majority of isolates was still susceptible to broad-spectrum antimicrobials such as cefotaxime, ceftazidime, cefpirome, meropenem, gentamicin and piperacillin/tazobactam. However, it should be noted that the prevalence of non-susceptibility to gentamicin increased from 0.7% (0.1% I and 0.6% R) in 2003 to 1.4% (0.3% I and 1.1% R) in 2004. This trend will be closely watched as it may threaten the traditional aminoglycoside-based empirical treatment regimens often used for septicaemia in Norway. Cefuroxime was removed from the programme as it has not proved useful for surveillance purposes.

All isolates were specifically examined for production of extended spectrum beta-lactamases (ESBL) by a disk approximation test, and isolates with a positive ESBL test and/or reduced susceptibility to ceftazidime (MIC $\geq$ 1 mg/L) and/or cefotaxime (MIC $\geq$ 1 mg/L) and/or cefpirome (MIC $\geq$ 1 mg/L) were further characterized by combination Etests and/or molecular examinations. In addition, zone diameters were measured in the disk approximation assays in order to evaluate the utility of single substrates as well as combinations of substrates for the detection of ESBL. The data from this methodological exercise are presented under a separate heading below.

A total of seven ESBL producing isolates were detected giving a 0.7% prevalence of ESBL producers among all isolates. Two hospitals were represented by two isolates each, whereas the remaining three isolates originated from separate institutions. Most of the isolates had MICs of cefotaxime significantly above the MICs of ceftazidime which is accordance with the recent finding of CTX-M as the predominant class of ESBL enzymes in Norway. The finding of 0.7% ESBL producers is an increase from 2001 (0%), 2002 (0.3%) and 2003 (0.3%).
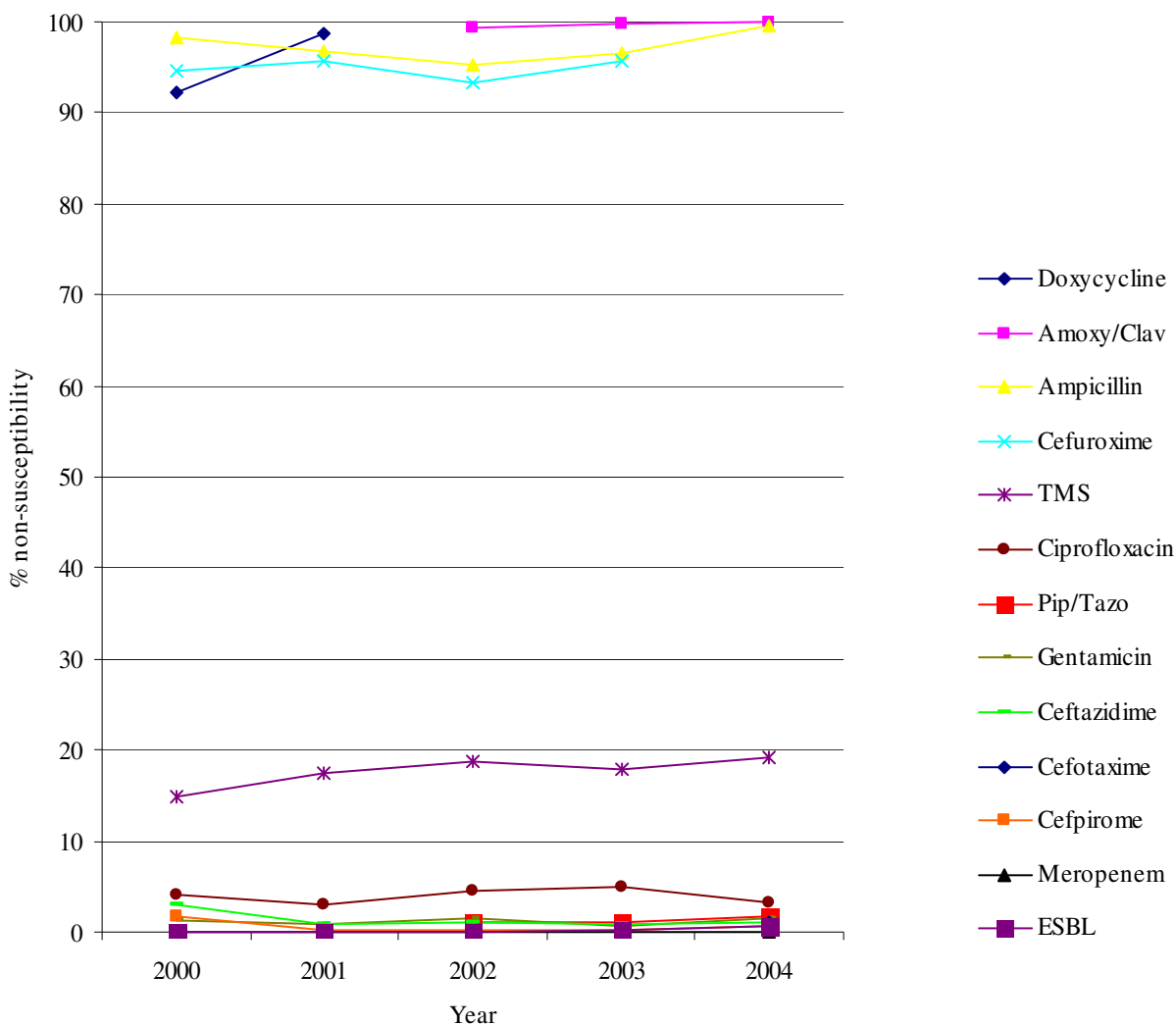

FIGURE 18. Prevalence of non-susceptibility to various antimicrobials in *Escherichia coli* blood culture isolates 2000-2004. The breakpoints for resistance and susceptibility were adjusted for beta-lactams and ciprofloxacin in 2004, see text.
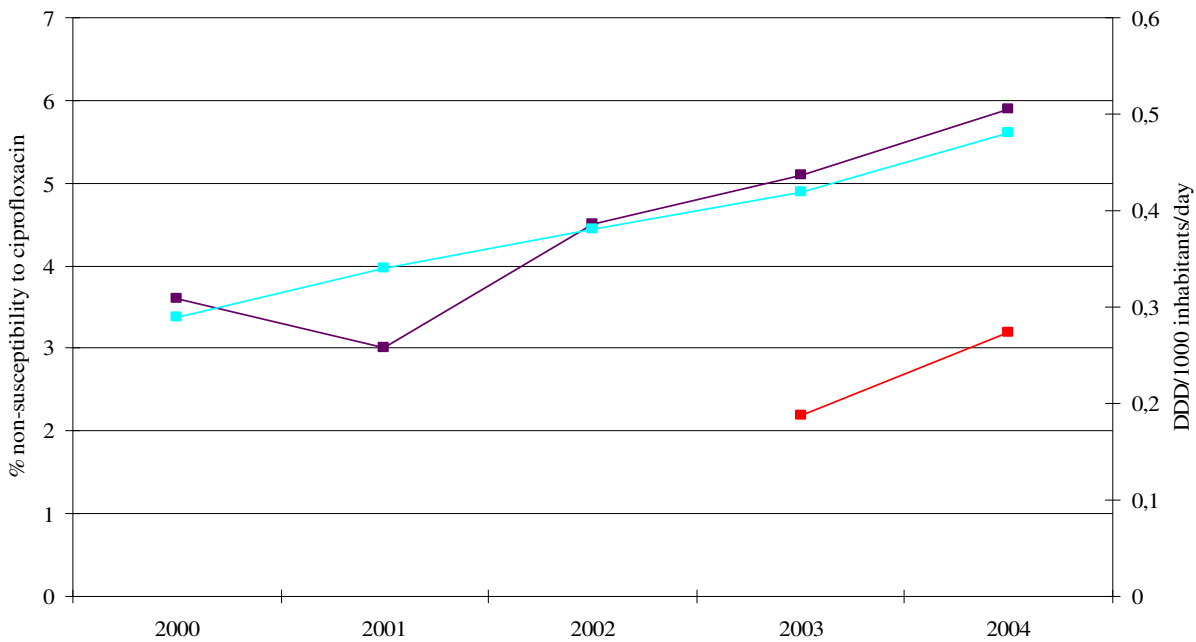
FIGURE 19. Prevalence of ciprofloxacin non-susceptibility in *Escherichia coli* blood culture isolates as defined by the former (magenta) and present (red) breakpoint protocol versus usage of ciprofloxacin (turquoise) 2000 – 2004.

European initiatives for surveillance of antimicrobial use and resistance – ESASC and EARSS

Antibacterial usage data from European countries have been collected by the European Surveillance of Antimicrobial Consumption ESAC. The aim of the ESAC project is to collect publicly available, comparable and reliable antibiotic use data in all European countries. The project has been granted by the DG/SANCO of the European Commission. The project was initiated in 2001. Data has been collected for the years 1997-2003 and the project has now been continued (2004-2007). Altogether 34 countries have provided data on ambulatory care, hospital care or total antibiotic usage. The data is collected as DDDs for ATC 5th levels in ATC group J01. A large variation in level of use and in usage patterns between countries is found. The data has been published (1, 2). More information may be found on the ESAC website www.uia.esac.be.

The European Antimicrobial Resistance Surveillance System was established in 1998 as an EU-financed "network og networks". EARSS is based on collection of routine laboratory data from a total of 926 laboratories / 1678 hospitals in 30 European countries within and outside the EU. The system only collects information on systemic isolates (blood and CSF) due to large variations between countries in microbiological sampling practices for less severe infections. The collected data are categorized according to SIR systems, but there is no uniform protocol of breakpoints applied. EARSS defines a standard protocol for susceptibility testing and provides an external quality assurance scheme in order to ensure some comparability of data across national borders. Norway has been a nominal participant in EARSS since 1999, but regular data entry did not commence until 2003 when collated NORM data were used. So far in 2004 and 2005, a total of 8 Norwegian laboratories have been recruited to provide routine data to EARSS including historical records from 1999 onwards. The Norwegian participation is from 2004 organized by NORM in Tromsø in collaboration with St. Olav University Hospital in Trondheim. More information is available at the EARSS website www.earss.rivm.nl and at www.antibiotikaresistens.no.

References:

1. Goossens, H., Ferech, M., Vander, Stichele R., and Elseviers, M. Outpatient Antibiotic Use in Europe and Association With Resistance: a Cross-National Database Study. Lancet 12-2-2005; 365(9459):579-87.
2. Vander Stichele, R. H., Elseviers, M. M., Ferech, M., Blot, S., and Goossens, H. European Surveillance of Antimicrobial Consumption (ESAC): Data Collection Performance and Methodological Approach. Br.J Clin.Pharmacol. 2004; 58(4):419-28.
3. Bronzwaer SL, Goettsch W, Olsson-Liljequist B, Wale MC, Vatopoulos AC, Sprenger MJ.. European Antimicrobial Resistance Surveillance System (EARSS): objectives and organisation. Euro Surveill. 1999 Apr;4(4):41-44.
4. Bruinsma N, Kristinsson KG, Bronzwaer S, Schrijnemakers P, Degener J, Tiemersma E, Hryniewicz W, Monen J, Grundmann H; EARSS. Trends of penicillin and erythromycin resistance among invasive Streptococcus pneumoniae in Europe. J Antimicrob Chemother. 2004 Dec;54(6):1045-50.

Hege Salvesen Blix and Gunnar Skov Simonsen

Klebsiella spp. in blood cultures

TABLE 41. *Klebsiella* spp. blood culture isolates (n=359). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)			MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R					
Ampicillin	≤ 0.5	≥ 16	0.0	8.1	91.9	1	-	≥ 256	32	≥ 256
Amoxi/Clav**	≤ 0.5	≥ 16	0.0	96.1	3.9	1	-	≥ 32	2	4
Cefotaxime	≤ 1	≥ 8	98.3	1.1	0.6	0.016	-	8	0.064	0.25
Ceftazidime	≤ 1	≥ 16	98.3	1.4	0.3	0.032	-	32	0.125	0.5
Cefpirome	≤ 1	≥ 16	98.3	1.7	0.0	0.016	-	8	0.064	0.25
Ciprofloxacin	≤ 0.5	≥ 2	97.5	0.8	1.7	0.002	-	≥ 32	0.032	0.125
Gentamicin	≤ 2	≥ 8	99.4	0.0	0.6	0.016	-	≥ 256	0.25	0.5
Meropenem	≤ 0.5	≥ 4	100.0	0.0	0.0	0.032	-	0.5	0.032	0.064
Pip/Tazo***	≤ 8	≥ 32	94.1	3.4	2.5	0.032	-	≥ 256	2	8
TMS****	≤ 2	≥ 16	95.3	0.6	4.2	0.016	-	≥ 32	0.125	0.5
ESBL			99.4	-	0.6					

*S=Susceptible, I=Intermediately susceptible, R=Resistant.
**Amoxi/Clav=Amoxicillin/clavulanic acid.
***Pip/Tazo=Piperacillin/tazobactam
****TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

TABLE 42. *Klebsiella* spp. blood culture isolates (n=359). Distribution (%) of MICs (mg/L).*

	≤ 0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Ampicillin									0.3	1.7	1.4	4.7	21.4	31.5	18.4	6.4	14.2
Amoxi/Clav**									8.4	55.4	27.9	4.5	3.1	0.8			
Cefotaxime			6.7	24.2	44.0	15.0	4.7	2.5	1.1	0.8	0.3	0.6					
Ceftazidime				1.1	15.6	38.4	28.7	8.1	6.1	1.4			0.3				
Cefpirome			1.9	11.7	62.1	13.9	5.3	1.9	1.4	0.8	0.3	0.6					
Ciprofloxacin	0.8	2.5	20.1	37.0	26.7	3.3	3.6	3.3	0.8	0.8	0.3	0.3	0.3				
Gentamicin			0.3	0.6	0.3	5.6	50.1	38.4	3.9	0.3			0.3				0.3
Meropenem				71.3	26.2	1.7	0.3	0.6									
Pip/Tazo***				0.3	0.3	0.5	0.5	1.4	9.2	45.7	28.4	7.2	3.3	0.3	0.3		1.9
TMS****			0.3	0.8	13.6	38.4	30.9	7.0	3.1	0.8	0.3	0.3		4.2			

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.
**Amoxi/Clav=Amoxicillin/clavulanic acid.
***Pip/Tazo=Piperacillin/tazobactam
****TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

Klebsiella spp. blood culture isolates included 217 *K. pneumoniae*, 64 *K. oxytoca* and 78 isolates not identified to the species level giving a total of 359 *Klebsiella* spp. isolates (Tables 41 and 42).

Except for amoxicillin/clavulanic acid, there were no more than one dilution step differences in beta-lactam MIC$_{50}$ and MIC$_{90}$ values between *K. pneumoniae* and *K. oxytoca*, and the two species were therefore analysed together. The changes in prevalences of resistance corresponded to the adjustments of breakpoints for Enterobacteriaceae. The reduction of the ampicillin breakpoint to ≤ 0.5 mg/L has completely eliminated the S category as was the intention, whereas the reduction of the breakpoint for resistance from ≥ 32 mg/L to ≥ 16 mg/L has reduced the intermediate category from 38.9% to 8.1% and increased the R category from 59.1% to 91.9%. None of these changes reflect changes in the underlying microbiology of

Klebsiella spp. in Norway. The 3.9% resistance to amoxicillin/clavulanic acid were constituted by a total 14 isolates of which eight were *K. oxytoca*, five were *K. pneumoniae* and one was not speciated. The relative predominance of *K. oxytoca* in this group may reflect a greater propensity for hyperproduction of the chromosomal K1 enzyme in *K. oxytoca* than the analoguous SHV enzyme in *K. pneumoniae* thus overcoming the inhibitory effect of clavulanic acid.

As for *E. coli*, blood culture isolates of *Klebsiella* spp. isolates were examined specifically for production of extended spectrum beta-lactamases by a disk approximation test, and isolates with a positive ESBL test and/or reduced susceptibility to cefotaxime (MIC ≥ 1 mg/L) and/or ceftazidime (MIC ≥ 1 mg/L) and/or cefpirome (MIC ≥ 1 mg/L) were further characterized by combination Etests and/or molecular examinations. Two

ESBL producing isolates were detected; one *K. pneumoniae* and the other unspeciated. The overall prevalence of ESBL production of 0.6% corresponds nicely with the R categories for ceftazidime (0.3%) and cefotaxime (0.6%). The zone diameters in the disk approximation assays were measured in order to evaluate the utility of single substrates as well as combinations of substrates for the detection of ESBL.

There were no significant changes in the prevalences of resistance to non beta-lactam antimicrobials such as gentamicin and trimethoprim/sulfamethoxazole. The apparent reduction of ciprofloxacin resistance is due to the reduction of the breakpoint for resistance from 4 mg/L to 2 mg/L and the increase of the breakpoint for

susceptibility from 0.125 mg/L to 0.5 mg/L. The prevalence of non-susceptibility of 8.3% (8.0% I and 0.3% R) in 2003 corresponds to 0.7% I and 0.3% R using the adjusted breakpoints. When compared to the 2004 data using the same breakpoints there is a noteworthy increase in the R category from 0.3% to 1.7%. The trend of increasing non-susceptibility to quinolones seen in previous years as well as in *E. coli* is thus continued.

There was a marked difference between *K. pneumoniae* and *K. oxytoca* in terms of resistance to non beta-lactam antimicrobials with *K. pneumoniae* being significantly more resistant to ciprofloxacin (4.1% non-susceptibility) and trimethoprim/sulfamethoxazole (6.9% non-susceptibility) than *K. oxytoca* (0% and 1.6% non-susceptibility, respectively).
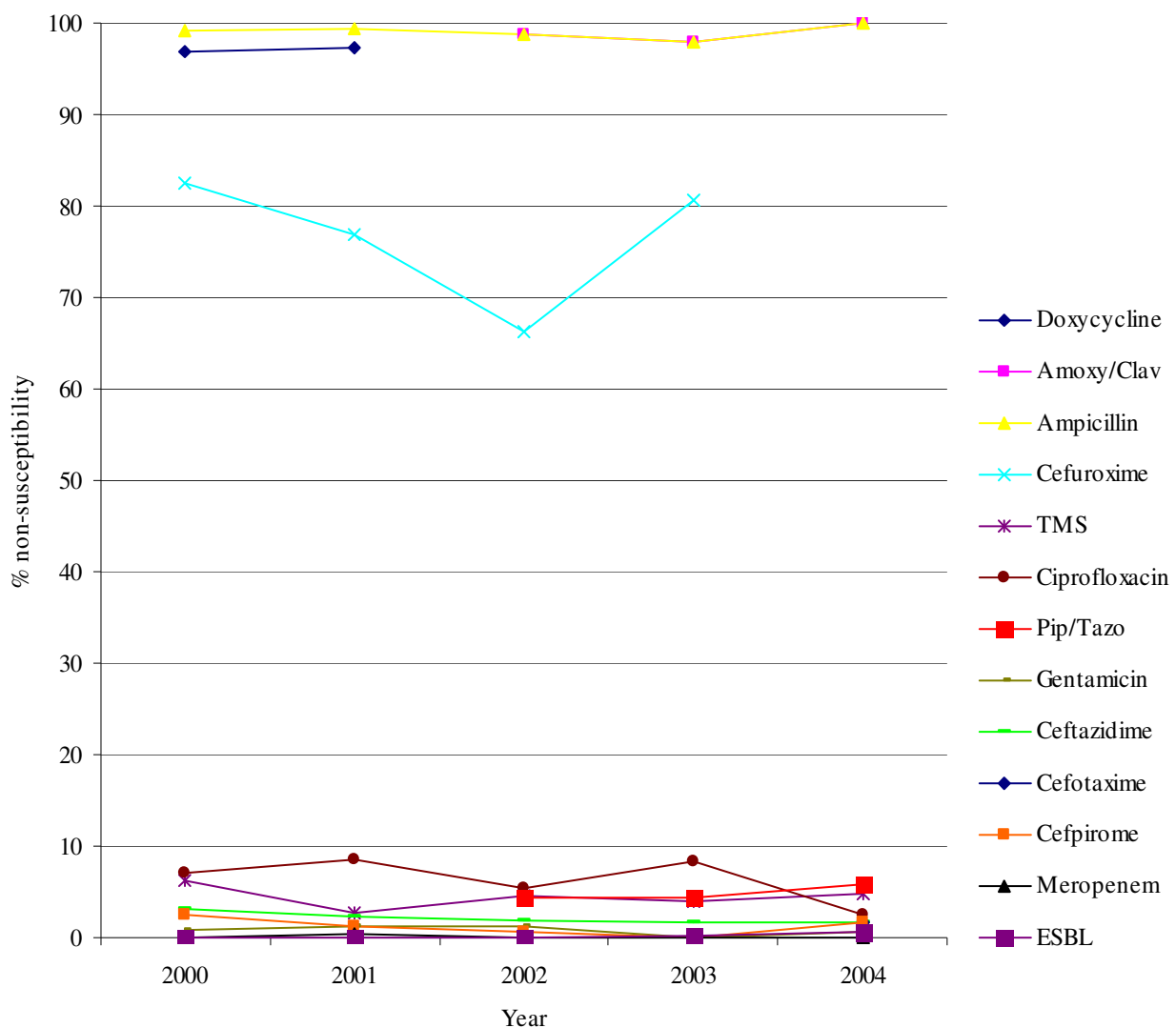

FIGURE 20. Prevalence of non-susceptibility to various antimicrobials in *Klebsiella* spp. blood culture isolates 2000-2004. The breakpoints for resistance and susceptibility were adjusted for beta-lactams and ciprofloxacin in 2004, see text.

Detection of extended-spectrum beta-lactamases (ESBL) in *Escherichia coli* and *Klebsiella* spp.

Due to the increasing focus on detection of extended-spectrum beta-lactamases in Enterobacteriaceae there is a need for rapid and simple methods for screening of ESBL production. The NORM protocol for 2004 therefore included measurement of zone diameters for five different beta-lactam substrates in addition to the combination disk containing amoxicillin and clavulanic acid.

TABLE 43. *Escherichia coli* blood and urinary tract isolates (n=1,880). Distribution (%) of zone diameters (mm). Sampling, laboratory methods, and data handling are described in Appendix 5.

	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Amoxi/Clav*	0.1			0.1	0.3	0.1	0.6	0.5	1.2	0.9	2.4	1.5	3.7	3.0	9.6
Aztreonam	0.1			0.1						0.1		0.1			0.1
Cefotaxime	0.1		0.1				0.1		0.1						0.1
Ceftazidime					0.1			0.1		0.1		0.1	0.1		0.1
Cefpodoxime	0.2	0.1		0.1	0.1			0.1			0.1	0.1		0.1	0.2
Cefpirom					0.1				0.1	0.1					

	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35
Amoxi/Clav*	3.6	11.7	5.0	15.0	5.8	13.4	2.4	5.8	0.9	3.1	0.9	1.2	0.3	1.1	0.2
Aztreonam	0.2	0.1	0.2	0.1	0.1	0.3	0.1	0.1	0.2	0.8	0.6	2.2	1.9	5.4	5.1
Cefotaxime	0.1	0.1	0.1	0.1	0.1	0.2	0.1		0.1	0.7	0.9	3.2	3.0	9.6	6.1
Ceftazidime		0.2	0.2	0.3	0.3	0.4	0.3	1.9	1.2	6.5	3.6	16.7	9.3	21.7	8.8
Cefpodoxime	0.2	0.4	0.1	0.2	0.3	0.4	0.8	2.4	2.5	9.3	5.7	15.5	10.2	20.9	8.7
Cefpirom			0.1	0.1	0.1	0.2	0.2	0.6	0.3	1.3	1.0	4.0	2.6	10.2	7.8

	36	37	38	39	40	41	42	43	44	45	46	47	48	49	≥ 50
Amoxi/Clav*	0.8	0.2	0.4		3.4	0.1	0.2	0.1	0.2					0.1	0.3
Aztreonam	13.8	7.8	17.7	6.7	19.2	3.7	7.4	1.5	3.7	0.4	0.5	0.1	0.2		
Cefotaxime	18.5	8.6	19.7	4.4	16.3	1.6	4.5	0.3	1.1	0.1	0.3		0.1		
Ceftazidime	13.7	4.2	5.8	0.8	3.0	0.2	0.1		0.2	0.1					
Cefpodoxime	10.5	3.1	4.2	1.2	1.7	0.2	0.3		0.1						
Cefpirom	19.0	11.1	19.2	4.6	11.6	1.6	2.6	0.4	1.0		0.1				0.1

*Amoxi/Clav=Amoxicillin/clavulanic acid.

There was some variation in the delineation of the normal distribution between different beta-lactam substrates, see Table 43. Cut-off values for decreased susceptibility are suggested in Table 44 together with detection rates for the six confirmed ESBL strains with registered zone diameters. Between 1.3% and 2.0% of all isolates had zone diameters below the suggested cut-off values and would thus have needed confirmatory testing in a diagnostic setting. As expected, cefpodoxime was less specific for ESBL detection than cefotaxime and cefpirom, but more sensitive than ceftazidime. Surprisingly, ceftazidime was clearly less specific and sensitive than cefotaxime and cefpodoxime. The two ESBL isolates not detected by ceftazidime had zone diameters of 28 mm and 30 mm, and adjustment of the cut-off values to accomodate these strains would consequently have compromised the specificity of the test even further. In the present epidemiological situation in Norway, aztreonam and cefotaxime appear as the two single substrates most suitable for ESBL detection. However, the distribution of ESBL fenotypes may change rapidly and the more prudent recommendation of using either cefpodoxime alone or cefotaxime and ceftazidime in combination therefore seems reasonable. Distributions of zone diameters for *E. coli* are presented graphically in Figure 21.

TABLE 44. Suggested cut-off values for detection of ESBL production in *Escherichia coli* and detection rate of confirmed ESBL strains using different beta-lactam substrates.

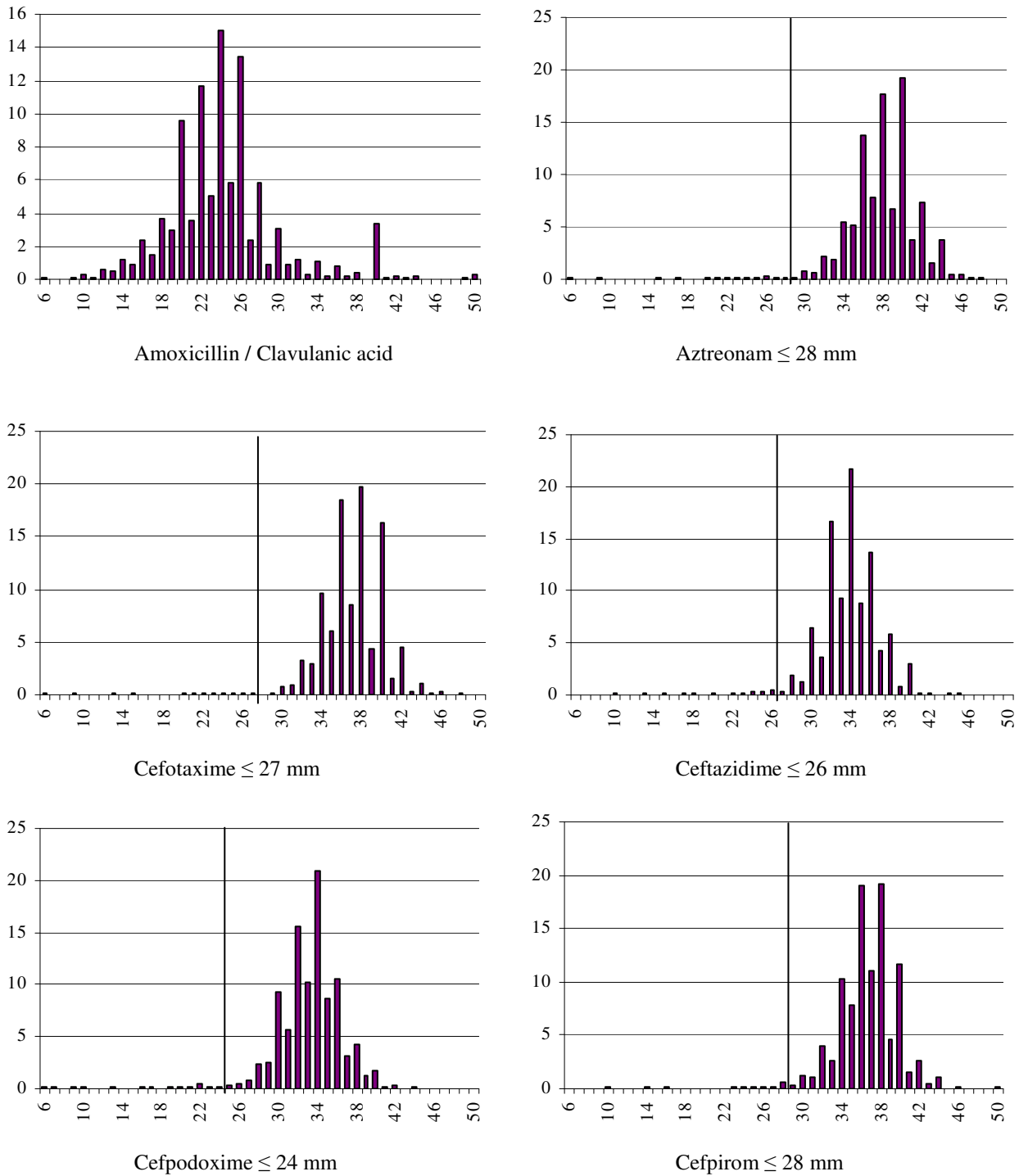| Substrate	Suggested cut-off values for ESBL detection	% of non-ESBL with reduced susceptibility	No. of ESBL with reduced susceptibility	No. of ESBL not detected by screening
Aztreonam	≤ 28 mm	1.7%	6/6	0/6
Cefotaxime	≤ 27 mm	1.3%	6/6	0/6
Ceftazidime	≤ 26 mm	2.0%	4/6	2/6
Cefpodoxime	≤ 24 mm	2.0%	6/6	0/6
Cefpirom	≤ 28 mm	1.6%	5/6	1/6

FIGURE 21. Distributions disk diffusion zone diameters (mm) of aztreonam, amoxicillin/clavulanic acid, cefotaxime, ceftazidime, cefpodoxime and cefpirom in *E. coli* from blood cultures and urine. Suggested cut-off values for ESBL detection are represented by vertical black bars.

TABLE 45. *Klebsiella* spp. blood isolates (n=328) examined by the ESBL disk approximation test. Sampling, laboratory methods, and data handling are described in Appendix 5.

	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Amoxi/Clav*			0.3			0.3	0.3	0.3	0.3	0.3	1.2		0.3	0.3	1.9
Aztreonam	0.6	0.3								0.3	0.3			0.3	
Cefotaxime															
Ceftazidime														0.3	
Cefpodoxime									0.3	0.3				0.3	
Cefpirom												0.3	0.3		

	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35
Amoxi/Clav*	1.5	4.6	2.5	15.4	7.1	21.0	9.3	14.5	0.6	8.0		1.2	0.3	3.4	1.5
Aztreonam				0.3		0.6		0.9		0.9	0.6	1.5	1.5	3.6	1.8
Cefotaxime		0.3	0.3		0.3	1.2	0.3	0.3	0.3	1.5	1.2	2.2	1.5	8.0	7.7
Ceftazidime			0.6	0.6	0.3	0.9	0.9	3.7	1.5	6.1	2.4	19.5	9.8	19.8	10.4
Cefpodoxime		0.6		0.3	0.6	0.6	0.3	0.6	1.6	1.9	0.6	4.7	6.6	16.6	10.9
Cefpirom				0.9	0.3			0.9	0.6	3.5	0.6	6.3	4.4	18.0	8.5

|	36	37	38	39	40	41	42	43	44	45	46	47	48	49	≥ 50
Amoxi/Clav*	0.3	0.6	1.2	0.3	0.3		0.3		0.3						
Aztreonam	13.7	8.5	19.1	4.9	21.3	3.6	7.3	0.9	4.3	0.9	0.3		0.3		1.2
Cefotaxime	17.6	10.8	20.4	4.6	16.4	0.3	1.5	0.9	1.5		0.3				
Ceftazidime	11.9	3.4	6.4	0.3	0.9		0.3								
Cefpodoxime	20.0	6.9	11.6	1.9	8.8	0.9	1.6		0.6	0.3	0.3	0.3			
Cefpirom	24.1	9.5	10.4	1.9	8.5		0.6								

*Amoxi/Clav=Amoxicillin/clavulanic acid.

Klebsiella spp. and *E. coli* are normally subjected to the same breakpoints and epidemiological cut-off values. Although this may be practical in the daily laboratory routine, the test properties may be compromised due to inherent differences between the two genera. The following proportions of false positives were detected when *E. coli* cut-off values were applied to *Klebsiella* spp.: aztreonam (≤ 28 mm) 3.6%; cefotaxime (≤ 27 mm) 2.4%; ceftazidime (≤ 26 mm) 2.7%; cefpodoxime (≤ 24 mm) 1.8%; and cefpirom (≤ 28 mm) 2.7% (Table 45, Figure 22). Only a single confirmed ESBL positive *Klebsiella pneumoniae* strain was registered with beta-lactam zone diameters. This strain would have been identified by all the five substrates examined. A valid evaluation of substrate sensitivities would require a higher number of true positive ESBL strains.
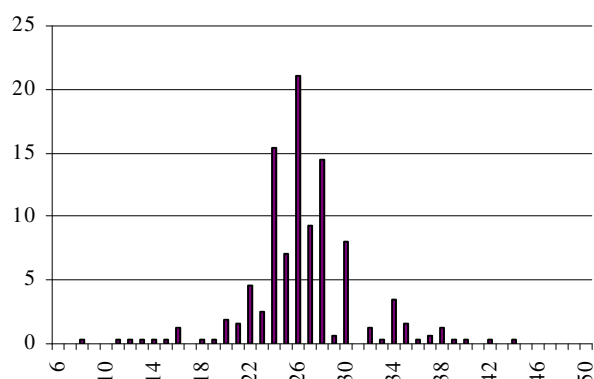
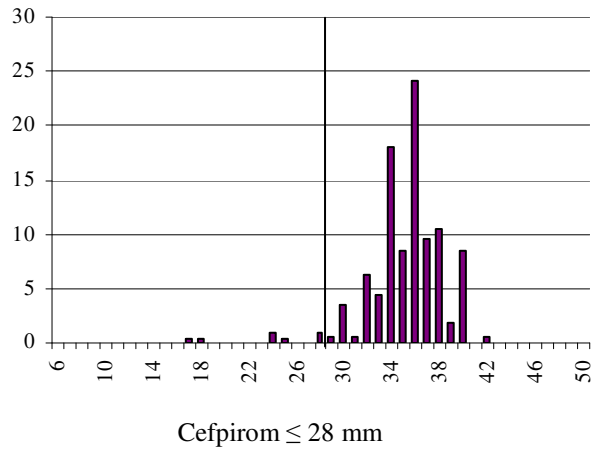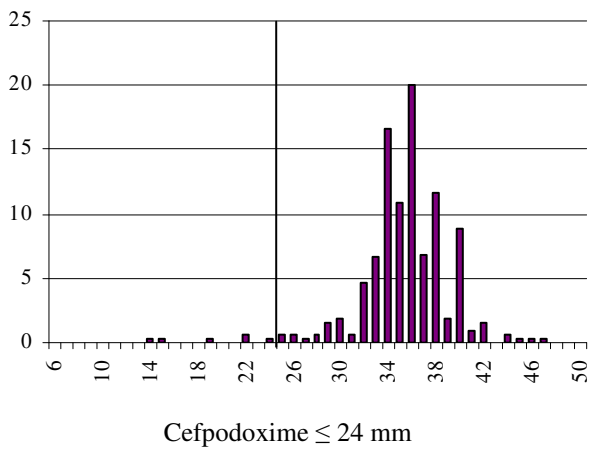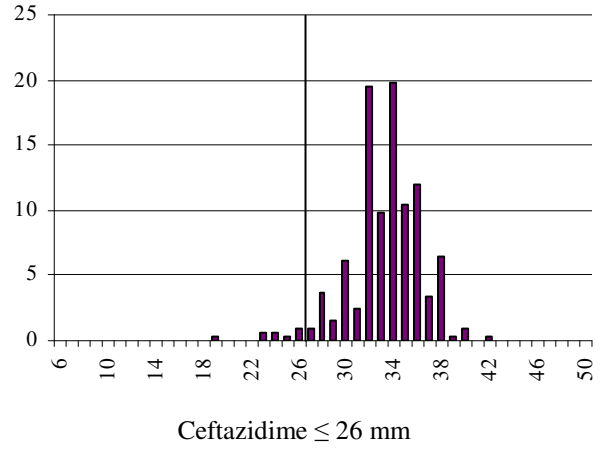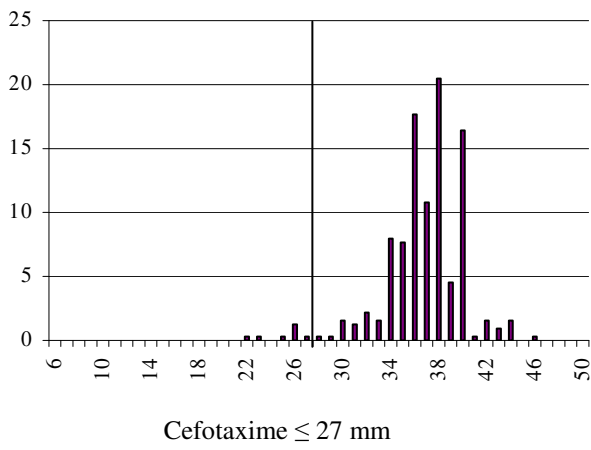Amoxicillin / Clavulanic acid
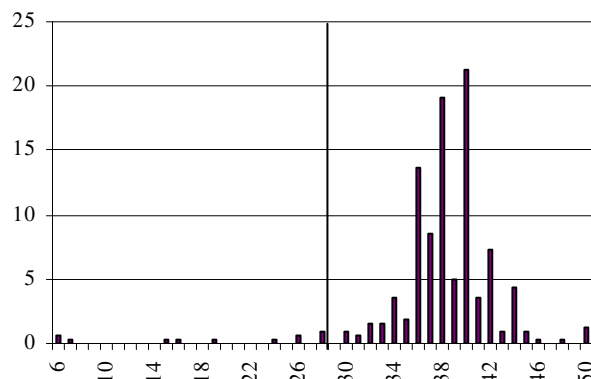
Aztreonam ≤ 28 mm

FIGURE 22. Distributions disk diffusion zone diameters (mm) of amoxicillin/clavulanic acid, aztreonam, cefotaxime, ceftazidime, cefpodoxime and cefpirom in *Klebsiella* spp. from blood cultures. Suggested cut-off values for ESBL detection are represented by vertical black bars.

Enterococcus spp. in blood cultures

TABLE 46. *Enterococcus* spp. blood culture isolates (n=294). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints mg/L		Proportion of isolates (%)*			MIC range	MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R			
Ampicillin	≤ 2	≥ 16	85.4	2.0	12.6	0.032 - ≥ 256	0.5	32
Gentamicin	≤ 128	≥ 256	79.3	–	20.7	0.25 - ≥ 1024	8	≥ 1024
Penicillin G	≤ 4	≥ 16	81.3	2.7	16.0	0.032 - ≥ 256	2	32
Streptomycin	≤ 256	≥ 512	74.1	–	25.9	0.5 - ≥ 1024	64	≥ 1024
Vancomycin Screen			98.6	–	1.4			
β-lactamase			100.0	-	0.0			

*S=Susceptible, I=Intermediately susceptible, R=Resistant.

TABLE 47. *Enterococcus* spp. blood culture isolates (n=294). Distribution (%) of MICs (mg/L).*

	≤ 0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	≥ 1024
Ampicillin		0.3	0.7	1.7	14.6	37.1	26.9	4.1	0.7	1.4	2.0	5.1	1.7	0.3	3.4		
Gentamicin				2.0	2.7	4.4	10.5	20.4	25.9	9.5	0.7	1.4	1.7	4.1	1.0		15.6
Penicillin G		0.3	0.3	0.7	0.7	5.4	19.7	38.1	16.0	2.7	2.0	4.1	1.0	0.7	8.2		
Streptomycin						0.3	0.7	1.0	2.7	3.1	9.9	17.7	25.9	9.5	3.4	1.0	24.8

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.

TABLE 48. *Enterococcus faecalis* blood culture isolates (n=227). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints mg/L		Proportion of isolates (%)*			MIC range	MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R			
Ampicillin	≤ 2	≥ 16	100.0	0.0	0.0	0.064 - 2	0.5	1
Gentamicin	≤ 128	≥ 256	78.0	–	22.0	0.25 - ≥ 1024	8	≥ 1024
Penicillin G	≤ 4	≥ 16	94.7	3.1	1.8	0.125 - 16	2	4
Streptomycin	≤ 256	≥ 512	78.4	–	21.6	0.5 - ≥ 1024	64	≥ 1024
Vancomycin Screen			100.0	–	0.0			
β-lactamase			100.0	-	0.0			

*S=Susceptible, I=Intermediately susceptible, R=Resistant.

TABLE 49. *Enterococcus faecalis* blood culture isolates (n=227). Distribution (%) of MICs (mg/L).*

	≤ 0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	≥ 1024
Ampicillin			0.9	1.3	17.6	44.1	31.3	4.4									
Gentamicin				1.8	0.9	3.1	10.1	18.9	29.1	10.6	0.4	0.9	2.2	4.8			17.2
Penicillin G			0.9			4.4	24.2	45.8	19.4	3.1	1.8						
Streptomycin						0.4		0.9	2.2	2.2	7.9	20.7	29.1	11.5	3.5		21.6

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.

TABLE 50. *Enterococcus faecium* blood culture isolates (n=52). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints mg/L		Proportion of isolates (%)*			MIC range	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Ampicillin	≤ 2	≥ 16	25.5	9.8	64.7	0.125 - ≥ 256	32	256
Gentamicin	≤ 128	≥ 256	80.4	-	19.6	0.25 - ≥ 1024	8	≥ 1024
Penicillin G	≤ 4	≥ 16	23.5	2.0	74.5	1 - ≥ 256	64	≥ 256
Streptomycin	≤ 256	≥ 512	51.0	-	49.0	1 - ≥ 1024	256	≥ 1024
Vancomycin Screen			100.0	-	0.0			
β-lactamase			100.0	-	0.0			

*S=Susceptible, I=Intermediately susceptible, R=Resistant.

TABLE 51. *Enterococcus faecium* blood culture isolates (n=52). Distribution (%) of MICs (mg/L).*

	≤ 0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	≥ 1024
Ampicillin				2.0	2.0	3.9	13.7	3.9	3.9	5.9	7.8	27.5	9.8	2.0	17.6		
Gentamicin					2.0	7.8	9.8	5.9	23.5	19.6	5.9	2.0	3.9		2.0		17.6
Penicillin G							3.9	15.7	3.9	2.0	3.9	19.6	5.9	3.9	41.2		
Streptomycin							3.9	2.0	2.0		7.8	9.8	17.6	3.9	3.9		49.0

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.

RESULTS AND COMMENTS

As in previous years, enterococci were analysed both as a genus and as separate species. The results for each species are microbiologically more valid as resistance rates differ significantly between *E. faecalis* and *E. faecium*. However, overall rates of resistance are of interest when formulating empirical treatment strategies because they include the probability of systemic infection with each enterococcal species. The overall results for enterococci are presented in Tables 46 and 47.

E. faecalis is still uniformly susceptible to ampicillin (Tables 48 and 49) whereas the prevalence of non-susceptibility to this agent has increased steadily in *E.*

faecium. As seen in Tables 50 and 51 and Figure 23, the prevalence of non-susceptibility has now reached 74.5%, with 63.7% being categorized as resistant (MIC ≥ 16 mg/L). Although the number of isolates is limited, the data are in accordance with recent Norwegian and international studies suggesting global dissemination of an ampicillin resistant *E. faecium* clone in hopital settings. Fortunately, vancomycin resistant *E. faecium* has not yet been established in Norway as only four VRE isolates were registered and all turned out to be *E. gallinarum* strains with chromosomally encoded VanC resistance.
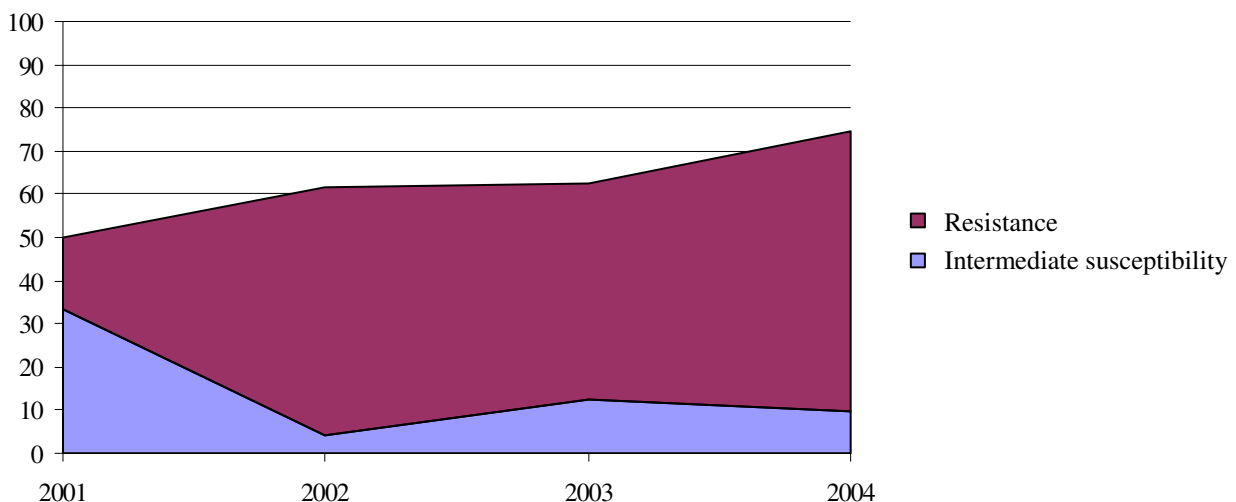

FIGURE 23. Prevalence of intermediate susceptibility and resistance to ampicillin in *E. facium* blood culture isolates. The breakpoints applied were S ≤ 1 mg/L and R ≥ 32 mg/L in 2001 and 2002, and S ≤ 2 mg/L and R ≥ 16 mg/L in 2003 and 2004.

The recommended treatment for systemic enterococcal infections includes a combination of an aminoglycoside and an agent active against the cell wall, i.e. ampicillin or vancomycin. It is therefore worrying that the prevalences of high-level resistance to gentamicin in *E. faecalis* and *E. faecium* have now reached 22.0% and 19.6%, respectively. Although the number of isolates is limited, the situation is especially problematic in *E. facium* where 17.6% of the isolates are now non-susceptible to both components of the combination regimen. Again, this may reflect the successfull spread of certain clones in Norwegian

hospitals. It should be noted that the breakpoint for high-level gentamicin resistance was decreased in 2004 from R $\geq$ 1024 mg/L to R $\geq$ 256 mg/L (officially denoted as R > 128 mg/L).

The change of breakpoints account for 4.8 and 2.0 percentage points of the increase for *E. faecalis* and *E. faecium*, respectively. Using the old breakpoints the prvalences of high-level resistance would have been 17.2% for *E. faecalis* and 17.6% for *E. faecium* which is still a substancial increase over the previous years.
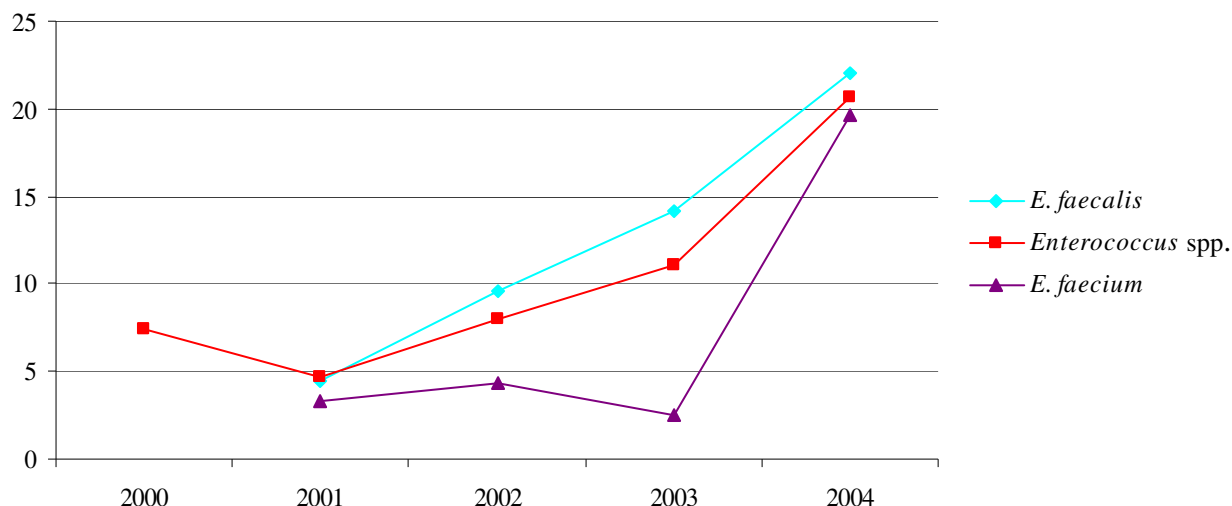

FIGURE 24. Prevalence of high-level resistance to gentamicin in blood culture isolates of *E. faecalis*, *E. faecium* and all enterococci combined 2000-2004. The breakpoint for high-level resistance was decreased from R $\geq$ 1024 mg/L to R $\geq$ 256 mg/L in 2004.

Streptococcus pneumoniae in blood cultures

TABLE 52. *Streptococcus pneumoniae* blood culture isolates (n=628). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)			MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R					
Cefotaxime	≤ 0.5	≥ 4	97.7	0.3	0.0	0.004	-	1	0.016	0.032
Cefuroxime	≤ 0.5	≥ 4	99.5	0.3	0.2	0.016	-	4	0.016	0.032
Chloramph.	≤ 4	≥ 8	99.8	0.0	0.2	0.25	-	8	2	2
Ciprofloxacin	≤ 0.125	≥ 4	0.3	98.6	1.1	0.064	-	32	1	2
Doxycycline	≤ 1	≥ 4	97.5	0.8	1.8	0.016	-	32	0.125	0.25
Erythromycin	≤ 0.5	≥ 1	90.3	0.0	9.7	0.016	-	≥ 256	0.125	0.25
Pen G**	≤ 0.064	≥ 2	98.1	1.8	0.2	0.004	-	2	0.016	0.032
TMS***	≤ 0.5	≥ 4	95.7	2.2	2.1	0.016	-	≥ 32	0.25	0.5
Vancomycin	≤ 4	≥ 8	100.0	0.0	0.0	0.125	-	4	0.5	1
Oxacillin screen	≥ 20 mm	≤ 19 mm	98.1	-	1.9					

*S=Susceptible, I=Intermediately susceptible, R=Resistant.
**Pen G=Benzylpenicillin.
***TMS= Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

TABLE 53. *Streptococcus pneumoniae* blood culture isolates (n=628). Distribution (%) of MICs (mg/L).*

	≤0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	≥ 128
Cefotaxime	0.5	3.2	70.9	22.5	1.4		1.0	0.2	0.3							
Cefuroxime			63.6	32.0	1.9	1.0	0.6	0.2	0.2	0.2	0.2					
Chloramph.							0.2	0.8	27.2	67.6	3.8	0.2				
Ciprofloxacin				0.2			1.3	14.5	54.0	28.8	1.0			0.2		
Doxycycline			0.2	0.3	7.2	44.3	38.4	6.8	0.3	0.8	0.2	0.6	0.8	0.2		
Erythromycin			0.2	1.1	28.3	52.7	8.0			0.2	0.5	2.4	4.5	1.4	0.2	0.6
Pen G**	1.4	16.9	63.9	14.5	1.4	0.5	0.5	0.5	0.3	0.2						
TMS***			0.3	0.6	0.2	12.1	65.0	17.7	1.3	1.0	0.8	0.3		1.0		
Vancomycin						0.3	2.4	49.4	38.0	9.6	0.2					

	≤ 19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	≥ 34
Oxacillin disk	1.9		1.3	2.6	3.5	7.5	13.6	16.5	13.6	11.4	7.9	10.4	4.2	1.6	1.9	2.1

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.
**Pen G=Benzylpenicillin.
***TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

The results are summarised in Tables 52 and 53. A total of 12 *S. pneumoniae* isolates were non-susceptible to penicillin G comprising 2.0% of the sample. This is an increase compared to the two previous years (0.6%) but lower than in 2001 when 3.9% of the isolates were recorded as non-susceptible to this agent. The majority of isolates (11 isolates, 1.8%) displayed reduced susceptibility (MIC 0.125 – 1 mg/L) whereas only a single isolate (0.2%) was resistant with an MIC of 2 mg/L. The penicillin resistant isolate was in addition intermediately susceptible to cefotaxime (MIC 1 mg/L) and resistant to cefuroxime (MIC 4 mg/L). Another isolate displayed resistance to cefuroxime (MIC 2 mg/L) and reduced suscpetibility to both penicillin G (1 mg/L) and cefotaxime (1 mg/L), whereas a third isolate was resistant to cefuroxime (1 mg/L), intermediately susceptible to penicillin G (MIC 1 mg/L) and susceptible to cefotaxime (MIC 0.25 mg/L). All the three isolates were highly resistant to erythromycin and doxycycline. The remaining nine penicillin non-suscpetible pneumococcal strains were

susceptible to all cephalosporins, and there were no penicillin susceptible strains with reduced susceptibility to cephalosporins. Among 624 isolates registered with results for both penicllin G and the oxacillin disk screen, six of the 12 non-susceptible strains were identified by the oxacillin disk (true positivies) in addition to six penicillin G susceptible strains (false positives). The remaining six penicillin G non-susceptible isolates were not detected by the screening test and were thus false negatives. 606 isolates were correctly identified as true negatives.

The prevalence of macrolide resistance increased to 9.7% in 2004 which is an increase of 3.7 percentage points over 2003 (Figure 25). The majority of isolates (57/61, 93.4%) displayed low-level resistance whereas only 4/61 (6.6%) were highly resistant with MIC ≥ 256 mg/L. Clindamycin was not included in the surveillance protocol, but macrolide resistant isolates should be subjected to double disk diffusion (DDD) tests for characterization of MLS phenotype. The vast majority (27/28, 96.4%) displayed the

efflux-based M phenotype whereas the ramaining isolate had a constitutively expressed MLS$_B$ phenotype. The findings are in line with a recent publication by Littauer *et al*. and may indicate further spread of the *mef*(A)-containing pneumococcal clone described in the paper.

There were no major changes for chloramphenicol, doxycycline, ciprofloxacin, vancomycin or trimethoprim/ sulfamethoxazole (Figure 26). In the 2005 protocol, ciprofloxacin will be replaced by norfloxacin and doxycycline will be replaced by tetracycline.
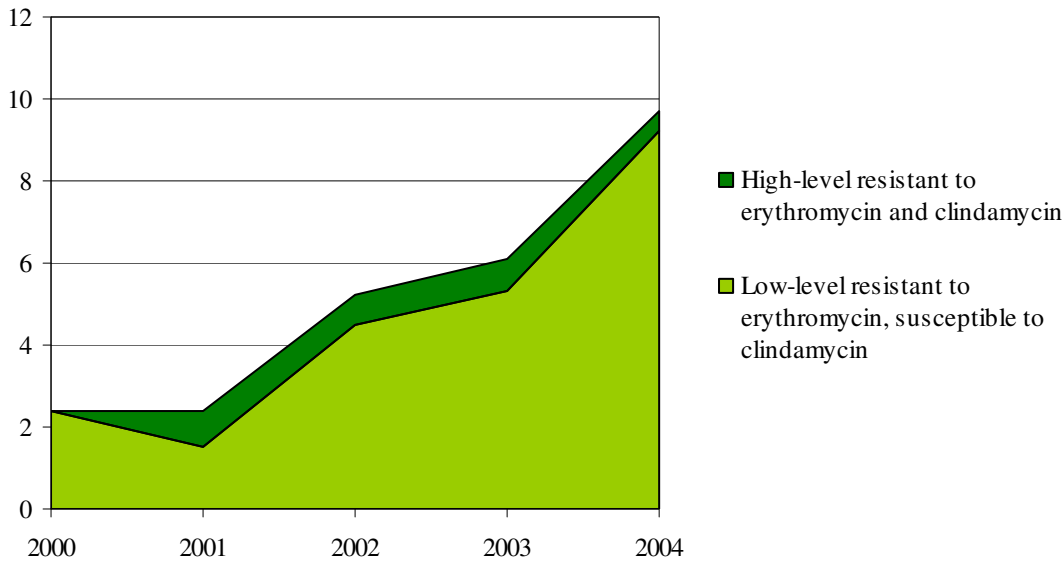
FIGURE 25. Prevalences (%) *of Streptococcus pneumoniae* with high-level resistance to erythromycin and clindamycin or low-level resistance to erythromycin and susceptibility to clindamycin 2000-2003.
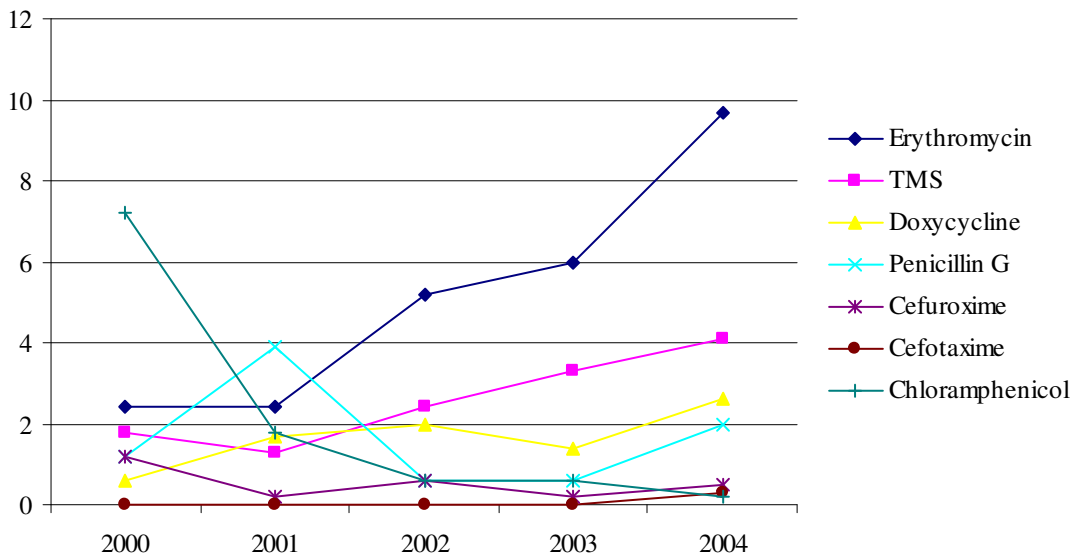

FIGURE 26. Prevalences (%) of non-susceptibility to various antimicrobials in *Streptococcus pneumoniae* blood culture isolates 2000 – 2004. The following breakpoints were adjusted in 2002: Chloramphenicol: S $\leq$ 2 mg/L and R $\geq$ 8 mg/L changed to S $\leq$ 4 mg/L and R $\geq$ 8 mg/L in 2002; cefuroxime and cefotaxime: S $\leq$ 1 mg/L and R $\geq$ 32 mg/L changed to S $\leq$ 0.5 mg/L and R $\geq$ 4 mg/L in 2002; trimethoprom/sulfamethoxazole (TMS): S $\leq$ 2 mg/L and R $\geq$ 16 mg/L changed to S $\leq$ 0.5 mg/L and R $\geq$ 4 mg/L; erythromycin: S $\leq$ 1 mg/L and R $\geq$ 4 mg/L changed to S $\leq$ 0.5 mg/L and R $\geq$ 1 mg/L.

Staphylococcus aureus in blood cultures

TABLE 54. *Staphylococcus aureus* blood culture isolates (n=660). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)		MIC_{50}	MIC_{90}
	S	R	S	I	R				
Doxycycline	≤ 1	≥ 4	95.6	1.5	2.9	0.016	- ≥ 256	0.125	0.25
Erythromycin	≤ 1	≥ 4	97.3	0.3	2.4	0.016	- ≥ 256	0.25	0.5
Fusidic acid	≤ 0.5	≥ 1	94.2	-	5.8	0.016	- ≥ 256	0.064	0.125
Gentamicin	≤ 2	≥ 8	99.8	0.0	0.2	0.016	- 16	0.25	0.5
Oxacillin	≤ 2	≥ 4	99.4	0.0	0.6	0.064	- 32	0.5	1
Oxacillin screen			99.4	-	0.6				
Penicillin G**	≤ 0.064	≥ 0.25	27.8	5.3	66.9	0.008	- ≥ 256	1	4
Beta-lactamase			31.9	-	68.1				
Vancomycin screen			100.0	-	0.0				

*S=Susceptible, I=Intermediately susceptible, R=Resistant.
**Penicillin G=Benzylpenicillin.

TABLE 55. *Staphylococcus aureus* blood culture isolates (n=660). Distribution (%) of MICs (mg/L).*

	≤0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Doxycycline		0.3	3.5	32.3	44.2	13.5	1.2	0.8	0.9	1.2	0.8	0.5	0.3			0.2
Erythromycin		0.2	1.1	3.3	31.8	53.2	7.3	0.5	0.3	0.5	0.3	0.3		0.2		1.2
Fusidic acid		0.3	15.3	50.2	24.7	3.5	0.3	1.1	0.8	1.5	1.2	0.5	0.3	0.2	0.2	0.2
Gentamicin		0.5	0.2	0.2	8.8	45.8	39.6	4.2	0.6			0.2				
Oxacillin				0.5	6.4	30.5	45.4	14.7	1.5	0.3		0.2	0.2			
Penicillin G**	0.3	5.5	18.9	3.1	5.2	4.2	11.3	23.8	17.5	5.3	2.0	1.6	0.6	0.2	0.2	0.5

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.
**Penicillin G=Benzylpenicillin.

RESULTS AND COMMENTS

A total of four *Staphylococcus aureus* isolates (0.6%) were verified as methicillin resistant *S. aureus* (MRSA) by *mecA* and *nuc* PCRs. This is in accordance with the reults from NORM 2000-2003 and the results from the national notification of systemic *S. aureus* infections where 8/1088 (0.7%) from the laboratory routine were reported as methicillin resistant. All the 8 MRSA reported from the laboratory routine were among 1085 blood culture isolates, whereas none of 11 cerebrospinal fluid isolates were MRSA.

The four MRSA strains from the NORM protocol were isolated at four different hospitals, and there were no apparent epidemiological links between them. Hopefully, a nationwide system for genetic typing of MRSA isolates will be established to determine possible clonal spread of such strains. All isolates were detected both by the oxacillin screening agar and the oxacillin Etest, but a considerable number of strains were mistakenly reported as MRSA due to typing errors and irreproducible methodological analyses. In the final analysis, no methicillin susceptible *S. aureus* (MSSA) strains had MIC values ≥ 4 mg/L or displayed growth on the oxacillin screening agar. The four MRSA strains were susceptible to all non beta-lactam antimicrobials except for resistance to fusidic acid in a single isolate.

68.1% of the isolates were beta-lactamase positive which is a decrease from 74.7% in 2003, whereas 72.2% were non-susceptible by the penicillin G Etest. A subgroup analysis of the 211 beta-lactamase negative isolates revealed 31 strains (14.7%) displaying MICs ≥ 0.125 mg/L. The corresponding figure for 2003 was 6/158 (3.4%) thus indicating increasing penicillin G MICs among beta-lactamase negative isolates. The majority of the strains were reported from only three laboratories which may indicate either local clonal dissemination or technical errors at these sites. There were no significant differences in prevalences of resistance to non beta-lactam antimicrobials between beta-lactamase negative and positive strains.

A total of 18 isolates (2.7%) were non-susceptible to erythromycin, with 2 (0.3%) being reported as intermediately susceptible and 16 (2.4%) being reported as resistant. These figures are essentially unchanged from 2004. The macrolide resistance phenotype was determined by double disk diffusion (DDD) tests in 18 isolates of which 4 (22%) were constitutively MLS_B resistant, 10 (56%) were inducibly MLS_B resistant and 3 (17%) displayed M type efflux mediated resistance. The last strain (6%) could not be classified.

The prevalences of resistance to doxycycline, gentamicin and fusidic acid were unchanged compared to 2003. No isolates displayed growth on the vancomycin agar screen. Figure 27 shows the prevalences of non-susceptibility to various non beta-lactam antimicrobials.
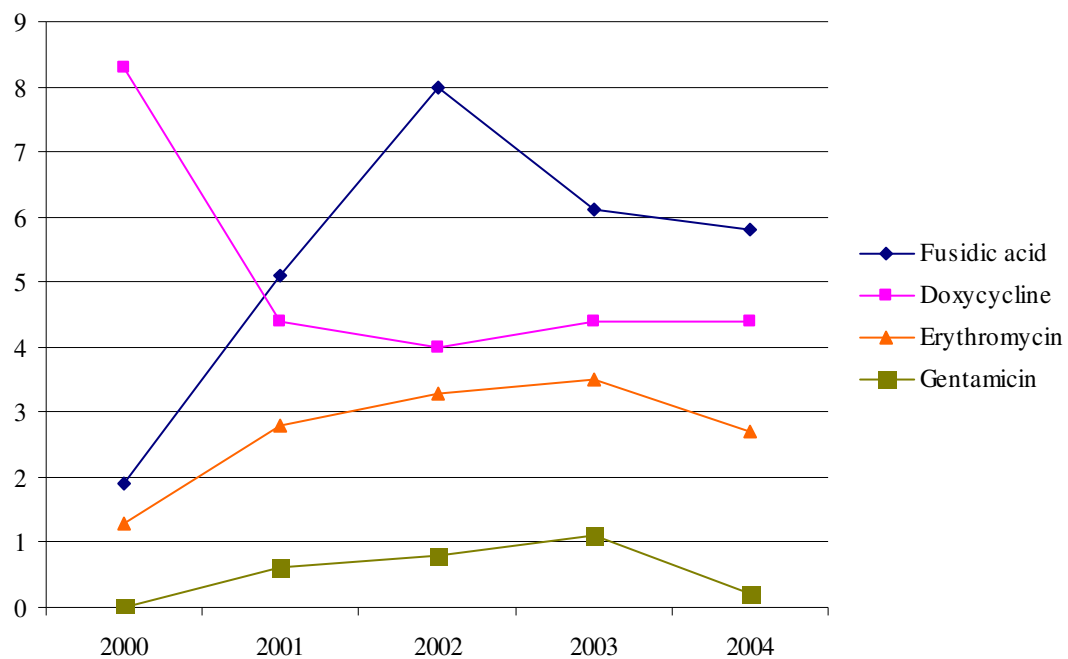
FIGURE 27. Prevalences of non-susceptibility to selected non beta-lactam antimicrobials among *Staphyloccus aureus* blood culture isolates 2000 – 2004. Breakpoints remained unchanged throughout the period.

MRSA infections in Norway

Infections with methicillin resistant *Staphylococcus aureus* (MRSA) have been notifiable in Norway since 1995 and colonisation without infection was made notifiable in 2005. Consistent discrimination between the two can be difficult.
In 2004 there was a slight increase in the number of reported cases from the previous year with 221 cases compared with 217 in 2003 (Figure 28). One hundred and thirteen (51%) were men. The mean age was 46 years (range 0-97 years). Thirty-five percent of the patients were hospitalised at the time of diagnosis.
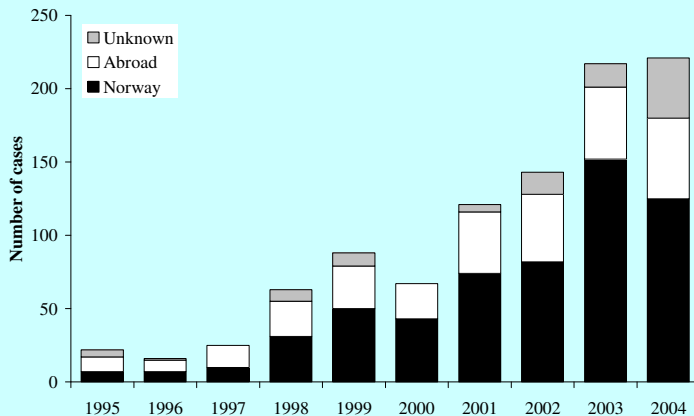

FIGURE 28. Reported cases of MRSA infection 1995–2004 and whether the infection was contracted abroad or not.

MRSA was found in blood cultures in nine patients in 2004 and only 37 for all ten years reported. The clinical picture shows a majority of wound infections or abscesses (Table 56).

TABLE 56. Clinical picture of reported cases of MRSA infection in Norway 1995–2004.

Clinical picture	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	Total
Septicaemia	1		3	3	4	2	6	4	5	10	38
Septicaemia and meningitis				1						1	2
Meningitis				1							1
Osteomyelitis	2	1		2			2	2	2	4	15
RTI[*], incl. otitis media	1	1	1	14	8	5	8	13	7	10	68
Urinary tract infection		1		4	3	3	2	9	12	13	47
Wound infection, abscess	17	14	19	36	71	54	97	115	189	176	789
Other, unknown			2	2	2	3	6		1	7	23
Total	21	17	25	63	88	67	121	143	216	221	983

[*] RTI = Respiratory tract infection

The number of reported cases of MRSA infection has increased steadily over the past ten years but seems to have levelled off this past year, at least temporarily. The overwhelming majority consists of wound infections. The number of serious infections is still very low with ten clinical septicaemias and nine positive blood cultures. Although the numbers appear to have increased they are still too small to ascertain whether there has been a true increase in serious infections.
How large the true increase in the total number of infections is, has to be interpreted with caution. The increase is mainly seen in non-hospitalised patients with minor infections and who have contracted the disease in Norway. This may indicate increased testing of patients outside hospitals.
The national surveillance of MRSA in Norway has been altered in 2005. From this year all findings of MRSA in patients are notifiable whether they are infections or mere colonisations. A national reference laboratory for MRSA has also been appointed; practical details are still to be decided. This will improve the quality of MRSA surveillance in Norway.

Bjørn G. Iversen

Haemophilus influenzae in respiratory tract specimens

TABLE 57. *Haemophilus influenzae* respiratory tract isolates (n=513). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints mg/L		Proportion of isolates (%)*			MIC range		MIC$_{50}$	MIC$_{90}$
	S	R	S	I	R				
Ampicillin	≤ 1	≥ 8	88.9	3.9	7.2	0.032 -	≥ 256	0.25	2
Amoxi./clav.****	≤ 2	≥ 8	96.9	2.5	0.6	0.016 -	8	0.5	1
Doxycycline	≤ 4	≥ 8	98.1	-	1.9	0.5 -	32	2	4
Erythromycin	≤ 1	≥ 16	1.6	78.8	19.7	0.032 -	32	8	16
Penicillin G**	≤ 1	≥ 8	72.7	15.6	11.7	0.032 -	≥ 256	0.5	8
Penicillin V***	≤ 0.5	≥ 8	1.6	68.2	30.2	0.064 -	≥ 256	4	64
TMS*****	≤ 0.5	≥ 4	81.2	3.1	15.6	0.008 -	≥ 32	0.063	32
β-lactamase			91.2	-	8.8				

*S=Susceptible, I=Intermediately susceptible, R=Resistant.
**Penicillin G=Benzylpenicillin.
***PenicillinV=Phenoxymethylpenicillin.
****Amoxi./clav.=Amoxicillin/clavulanic acid.
*****TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

TABLE 58. *Haemophilus influenzae* respiratory tract isolates (n=513). Distribution (%) of MICs (mg/L).*

	≤ 0.008	0.016	0.032	0.063	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Ampicillin			0.2	0.4	13.3	58.1	8.2	8.8	2.7	1.2	1.0	1.9	2.3	0.8	0.2	1.0
Amoxi./clav.****		0.2		0.4	0.8	4.9	58.5	26.3	5.8	2.5	0.4					
Doxycycline							3.3	22.8	57.7	14.2	0.8	1.0	0.2			
Erythromycin			0.2			0.2	0.4	0.8	4.5	32.0	42.3	18.5	1.2			
Penicillin G**			0.2	0.4	1.4	18.7	42.3	9.7	11.1	4.5	1.8	1.4	4.5	1.4		2.7
Penicillin V***				0.4		0.2	1.0	8.0	39.8	20.5	7.6	7.8	4.5	1.8	0.4	8.2
TMS*****	0.6	4.7	16.6	31.4	22.6	4.5	0.8	1.8	1.4	1.8	1.0	1.0	11.9			

*Shaded areas in each row indicate susceptibility (light blue), intermediate susceptibility (medium blue) and resistance (dark blue). Non-shaded areas represent MIC values that are not covered by the standard test method.
**Penicillin G=Benzylpenicillin.
***PenicillinV=Phenoxymethylpenicillin.
****Amoxi./clav.=Amoxicillin/clavulanic acid.
*****TMS=Trimethoprim/sulfamethoxazole.

RESULTS AND COMMENTS

Haemophilus influenzae isolated from respiratory tract specimens has previously been surveyed in 2000 and 2001. The prevalence of beta-lactamase production increased slightly from 7.1% in 2000 and 7.0% in 2001 to 8.8% in 2004 (Tables 57 and 58). Reduced susceptibility to amoxicillin/ clavulanic acid is normally caused by changes in penicillin binding proteins (PBPs) as opposed to beta-lactamase production. The prevalence of these beta-lactamase negative *H. influenzae* with reduced susceptibility to ampicillin (BLNAR) increased from 1.2% in 2000 and 1.3% in 2001 to 3.1% in 2004. In total, the 8.8% beta-lactamase production and the 3.1% prevalence of non-susceptibility to amoxicillin/clavulanic acid add up to 11.9% non-susceptibility to ampicillin. This is in accordance with the ampicillin MIC determination revealing 7.2% resistance and 3.9% reduced susceptibility giving a total of 11.1% non-susceptibility to this agent. The limited utility of penicillin G and V in antimicrobial susceptibility testing for beta-lactams was demonstrated by protracted distributions and poor separation of resistance categories.

Doxycycline breakpoints have been adjusted on the basis of earlier NORM results. They are specified for different media with S ≤ 2 and R ≥ 4 for HTM medium, and S ≤ 4 and R ≥ 8 for MH medium containing Isolvitalex and haemoglobin. NORM 2004 was performed on the now obsolete PDM II medium with 1% Isolvitalex and 1% haemoglobin. The data have been interpreted using the breakpoints for Mueller Hinton agar. As seen in Figure 29, the normal distribution is now closely delineated by the breakpoint and only a few scattered isolates have MICs of 8 – 32 mg/L. The MIC$_{50}$ and MIC$_{90}$ values have remained unchanged at 2 and 4 mg/L, respectively.

The Norwegian Working Group on Antibiotics (NWGA) has defined wildtype *H. influenzae* as intermediately susceptible to erythromycin and has adjusted the breakpoints accordingly. The breakpoint for susceptibility has remained at S ≤ 1 mg/L whereas the breakpoint for resistance has been increased from R ≥ 4 mg/L to R ≥ 16 mg/L. In spite of the consequent alteration in distribution between resistance categories, there are no indications of changing patterns of resistance in the bacterial population. The MIC$_{50}$ and MIC$_{90}$ values are essentially unchanged and the normal distribution still terminates around 16 mg/L.

The breakpoints for trimethoprim/sulfamethoxazole have also been changed since the last survey of *H. influenzae*. However, the sharply increasing proportion of resistant

isolates is not merely an effect of susceptibility being defined by S ≤ 0.5 mg/L (formerly MIC S ≤ 2 mg/L) and resistance by R ≥ 4 mg/L (formerly R ≥ 16 mg/L). When the results from 2000 and 2001 were recalculated using the new breakpoints there was still an increase of TMS

non-susceptibility from 9% in 2000 and 7% in 2001 to 18.7% in 2004. The prevalence of resistance in 2004 was 15.6%, and the MIC_{90} value increased from 0.25 mg/L to 32 mg/L. The emergence of TMS resistance in *H. influenzae* is illustrated in Fig 30.
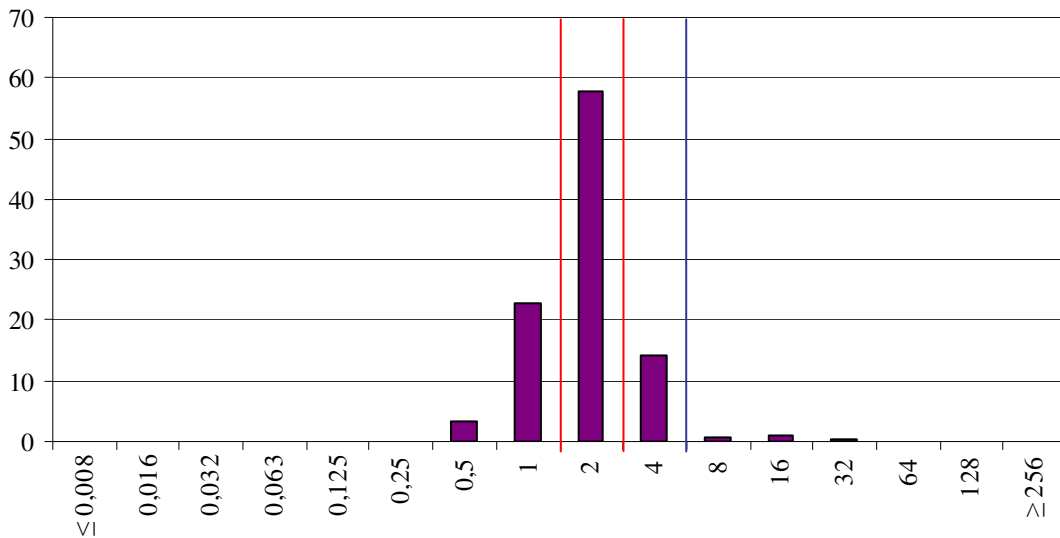
FIGURE 29. Distribution (%) of minimum inhibitory concentrations (mg/L) of doxycycline in *Haemophilus influenzae* from respiratory tract samples. The vertical bars indicate the previous breakpoints in red (S ≤ 1 mg/L and R ≥ 4 mg/L) and the new ones in blue (S ≤ 4 mg/L and R ≥ 8 mg/L).
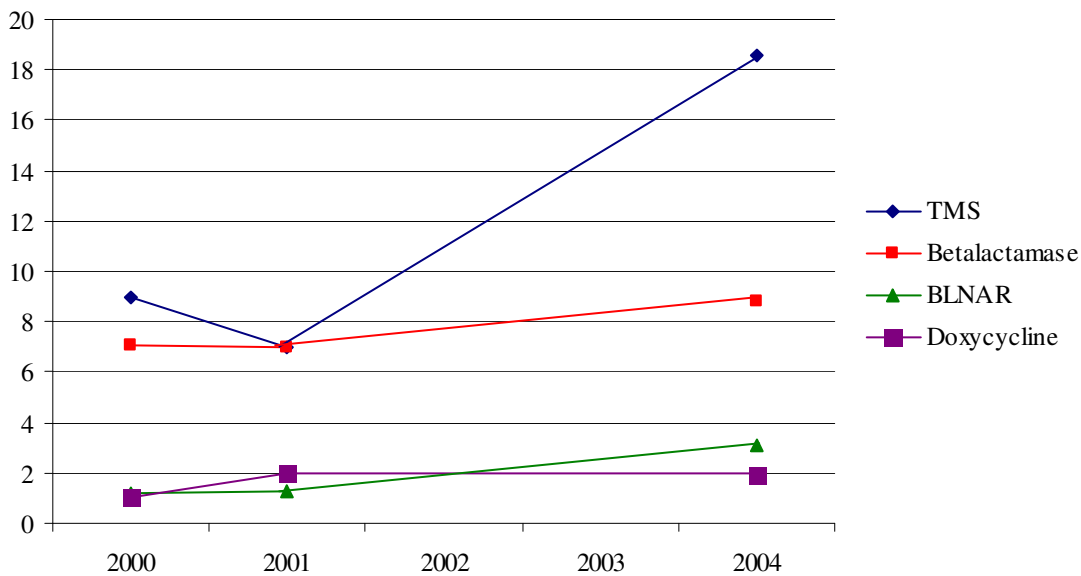

FIGURE 30. Prevalences of non-susceptibility to various antimicrobials in *Haemophilus influenzae* from respiratory tract samples. The data for doxycycline and TMS from 2000 and 2001 have been recalculated using the new breakpoints (doxycycline S ≤ 4 mg/L and R ≥ 8 mg/L, TMS S ≤ 0.5 mg/L and R ≥ 4 mg/L). TMS: trimethoprim/sulfamethoxazole, BLNAR: beta-lactamase negative non-susceptible to ampicillin.

Streptococcus pyogenes in specimens from wounds and the respiratory tract

TABLE 59. *Streptococcus pyogenes* wound specimen and respiratory tract isolates (n=977). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Doxycycline	≤ 1	≥ 4	88.3	0.0	11.7	0.016 - 32	0.125	8
Erythromycin	≤ 0.5	≥ 1	98.0	-	2.0	0.008 - ≥ 256	0.125	0.25
Penicillin G**	≤ 0.125	≥ 0.25	100.0	-	0.0	0.008 - 0.063	0.016	0.016
TMS***	≤ 0.5	≥ 4	93.5	4.8	1.6	0.016 - ≥ 32	0.125	0.5

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **Penicillin G=Benzylpenicillin.
***TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

TABLE 60. *Streptococcus pyogenes* wound specimen and respiratory tract isolates (n=977). Distribution (n) of MIC values (mg/L).

	≤ 0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	≥ 128	≥ 256
Doxycycline		0.3	1.0	11.4	48.1	26.3	0.9	0.2			0.1	2.7	7.9	1.0			
Erythromycin	0.1	0.2	0.9	20.6	58.7	16.6	0.9			0.3	0.2	0.4	0.5				0.5
Penicillin G**	8.6	87.1	3.8	0.5													
TMS***		0.3	4.3	24.7	39.4	18.0	6.9	4.0	0.8	0.4	0.2			1.0			

*Shaded areas in each row indicate susceptibility (light blue), intermediate susceptibility (medium blue) and resistance (dark blue). Non-shaded areas represent MIC values that are not covered by the standard test method. **Penicillin G=Benzylpenicillin. ***TMS=Trimethoprim/sulfamethoxazole.

TABLE 61. *Streptococcus pyogenes* wound specimen isolates (n=503). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Doxycycline	≤ 1	≥ 4	84.9	0.0	15.1	0.016 - 32	0.125	16
Erythromycin	≤ 0.5	≥ 1	97.0	-	3.0	0.008 - ≥ 256	0.125	0.25
Penicillin G**	≤ 0.125	≥ 0.25	100.0	-	0.0	0.008 - 0.032	0.016	0.016
TMS***	≤ 0.5	≥ 4	93.8	4.2	2.0	0.016 - ≥ 32	0.125	0.5

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **Penicillin G=Benzylpenicillin.
***TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

TABLE 62. *Streptococcus pyogenes* wound specimen isolates (n=503). Distribution (n) of MIC values (mg/L).

	≤ 0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Doxycycline		0.2	1.4	9.1	46.5	26.8	0.6	0.2			0.2	3.0	9.9	2.0			
Erythromycin	0.2		1.0	16.3	61.0	18.1	0.4			0.4	0.4	0.8	1.0				0.4
Penicillin G**	7.9	88.5	3.6														
TMS***		0.4	4.0	24.9	39.2	17.5	8.0	3.4	0.8	0.6	0.2			1.2			

*Shaded areas in each row indicate susceptibility (light blue), intermediate susceptibility (medium blue) and resistance (dark blue). Non-shaded areas represent MIC values that are not covered by the standard test method. **Penicillin G=Benzylpenicillin. ***TMS=Trimethoprim/sulfamethoxazole.

TABLE 63. *Streptococcus pyogenes* respiratory tract isolates (n=474). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Doxycycline	≤ 1	≥ 4	92.0	0.0	8.0	0.016 - 16	0.125	0.25
Erythromycin	≤ 0.5	≥ 1	99.2	-	0.8	0.016 - ≥ 256	0.125	0.25
Penicillin G**	≤ 0.125	≥ 0.25	100.0	-	0.0	0.008 - 0.063	0.016	0.016
TMS***	≤ 0.5	≥ 4	93.2	5.5	1.3	0.016 - ≥ 32	0.125	0.5

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **Penicillin G=Benzylpenicillin.
***TMS=Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

TABLE 64. *Streptococcus pyogenes* respiratory tract isolates (n=474). Distribution (n) of MIC values (mg/L).

	≤ 0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	≥ 128	≥ 256
Doxycycline			0.4	0.6	13.7	50.0	25.7	1.3	0.2			2.3	5.7				
Erythromycin			0.4	0.8	25.1	55.9	15.4	1.5		0.2							0.6
Penicillin G**		9.2	85.8	3.9	1.1												
TMS***			0.2	4.6	24.7	39.5	18.4	5.9	4.6	0.8	0.2	0.2		0.8			

*Shaded areas in each row indicate susceptibility (light blue), intermediate susceptibility (medium blue) and resistance (dark blue). Non-shaded areas represent MIC values that are not covered by the standard test method. **Penicillin G=Benzylpenicillin. ***TMS=Trimethoprim/sulfamethoxazole.

COMMENTS

Group A streptococci (*S. pyogenes*) were first introduced into the NORM programme in 2002, and the 2004 data represent only the second national survey of this species (Tables 59-64). As in 2002, isolates originating from both the respiratory tract (n=474) and wound specimens (n=503) were included in the protocol.

The Norwegian Working Group for Antibiotics (NWGA) has recently introduced specific breakpoints for streptococci other than *S. pneumoniae* as opposed to the former use of pneumococcal breakpoints for all streptococci. All isolates were susceptible to penicillin G irrespective of whether the the pneumococcal breakpoint of S ≤ 0.064 mg/L or the one for beta-haemolytic streptococci of S ≤ 0.125 mg/L were used.

The overall prevalence of resistance to doxycycline was slightly lower in 2004 (11.7%) than in 2002 (15.6%), see Figure 31. This may be related to the decreasing usage of tetracyclines among out-patients in Norway. As in 2002, resistance was more widespread among isolates originating from wounds (15.1%) than among respiratory tract isolates (8.0%). The doxycycline data were interpreted using pneumococcal breakpoints as no breakpoints have been defined for doxycycline in beta-haemolytic streptococci. From 2005, doxycycline will be substituted by tetracycline as recommended by NWGA.

Macrolide resistance in *S. pyogenes* has become a major problem in countries such as Finland and Italy. It is therefore remarkable that the prevalence of erythromycin resistance in Norway has decreased from 3.8% in 2002 to 2.0% in 2004. Again, there is a discrepancy between isolates from wounds (3.0% resistance) and the respiratory tract (0.8%). The changing epidemiology of macrolide resistance in *S. pyogenes* may be explained by one or more resistant clones passing through the human population before gradually being eliminated by herd immunity. Only six of the 19 erythromycin resistant isolates were further characterized by double disk diffusion. Four isolates displayed inducible MLS_B resistance whereas the other two exhibited the efflux-mediated M phenotype.

Trimethoprim/sulfamethoxazole is rarely used for treatment of *S. pyogenes* infections in Norway but may be an alternative in cases of allergy or treatment failure. The overall prevalence of reduced susceptibility to TMS increased from 0.1% in 2002 to 4.8% in 2004, and a corresponding increase of resistance from 0.3% to 1.6% was also noted. There were no apparent differences between isolates originating from wounds (6.2% non-susceptibility) and the respiratory tract (6.8% non-susceptibility). Antimicrobial susceptibility testing of TMS in *S. pyogenes* is technically challenging and may be prone to errors, but the observed changes warrant further investigations.
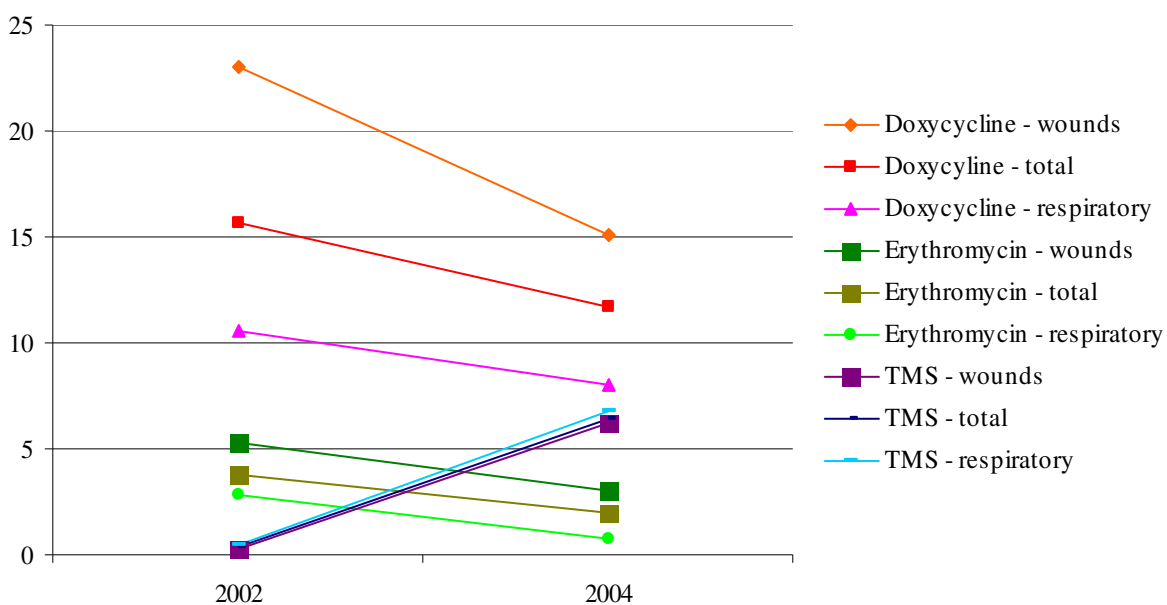

FIGURE 31. Prevalences of non-susceptibility to various antimicrobials in *Streptococcus pyogenes* from the respiratory tract and wound specimens in 2002 and 2004.

Staphylococcus aureus in wound specimens

TABLE 65. *Staphylococcus aureus* isolates from wound specimens (n=1,136). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Doxycycline	≤ 1	≥ 4	94.6	2.2	3.2	0.016 - 16	0.125	0.25
Erythromycin	≤ 1	≥ 4	95.9	0.2	4.0	0.016 - ≥ 256	0.25	0.5
Fusidic acid	≤ 0.5	≥ 1	75.0	–	25.0	0.016 - ≥ 256	0.064	4
Oxacillin	≤ 2	≥ 4	99.3	0.0	0.7	0.016 - ≥ 256	0.5	1
Oxacillin screen			99.5	–	0.5			
Penicillin V**	≤ 0.064	≥ 0.25	24.8	5.9	69.3	0.016 - ≥ 256	0.25	1
Beta-lactamase			23.8	-	76.2			
Vancomycin screen			100.0	-	0.0			

*S=Susceptible, I=Intermediately susceptible, R=Resistant.
**Penicillin V=Phenoxymethylpenicillin.

TABLE 66. *Staphylococcus aureus* isolates from wound specimens (n=1,136). Distribution (%) of MICs (mg/L).*

	≤0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Doxycycline		0.4	4.6	30.0	43.3	13.7	1.8	0.6	2.2	1.7	0.8	0.7				
Erythromycin		0.5	0.4	9.2	34.1	41.6	9.7	0.4	0.2	0.3	0.2	0.1	0.3			3.2
Fusidic acid		0.4	12.6	38.4	18.6	4.5	0.5	0.9	6.4	13.2	2.6	1.0	0.4		0.1	0.5
Oxacillin		0.4	0.7	0.6	4.1	28.7	50.3	13.0	1.5	0.3	0.1	0.2				0.2
Penicillin V**		12.2	10.1	2.6	5.7	22.4	25.9	13.0	4.1	1.5	0.9	0.3	0.1	0.1	0.1	1.1

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark). Non-shaded areas represent MIC values that are not covered by the standard test method.
**Penicillin G=Phenoxymethylpenicillin.

RESULTS AND COMMENTS

A total of 76.2% of the strains were beta-lactamase positive which is in accordance with earlier NORM results (Tables 65 and 66). Methicillin resistant *S. aureus* (MRSA) isolates were detected in six wound specimens in 2004 (0.5%). This is a slight increase from 0.3% in 2003 and corresponds to the increasing number of MRSA wound infections reported to the Norwegian Surveillance System for Communicable Diseases (MSIS) in 2004. The six isolates originated from the central eastern (4) and western (2) parts of the country. Only a single MRSA isolate was co-resistant to macrolides, but four of them were resistant to fusidic acid. One may therefore speculate about a possible connection between MRSA soft tissue infections and the clonal spread of low level fusidic acid resitant *S. aureus*. None of the MRSA isolates displayed reduced susceptibility to vancomycin. A national system for genetic typing of MRSA isolates will be a necessary next step for elucidation of MRSA epidemiology in Norway.

The prevalence of resistance to fusidic acid remained high with 25.0% in 2004 compared to 20.8% in 2001 and 23.0% in 2003. As in pevious years the majority of resistant isolates displayed low-level reistance (MIC 2 – 8 mg/L) compatible with the FusB phenotype, as oppsosed to the high-level FusA phenotype (MIC ≥ 256 mg/L), Figure 32. The fusidic acid resistant isolates had higher prevalences of resistance to doxycycline (7.0%), erythromycin (6.4%) and beta-lactamase production (91.4%) than the total sample. Fusidic acid resistant isolates are now recovered from all parts of the country.

Reduced susceptibility to erythromycin was seen in 4.2% of the isolates with the majority (4.0%) being fully resistant to this agent. The prevalence of non-susceptibility to macrolides was markedly higher in localized infections than in systemic isolates (0.3% reduced susceptibility and 2.4% resistance). 33 macrolide resistant isolates were further analysed by double disk diffusion methodology which revealed a predominance of inducible (24/33, 73%) and constitutive (4/33, 12%) MLS_B resistance as opposed to M type resistance caused by efflux mechanisms (5/33, 15%).

Figure 33 displays the prevalences of non-susceptibility to various antibiotics other than beta-lactams.
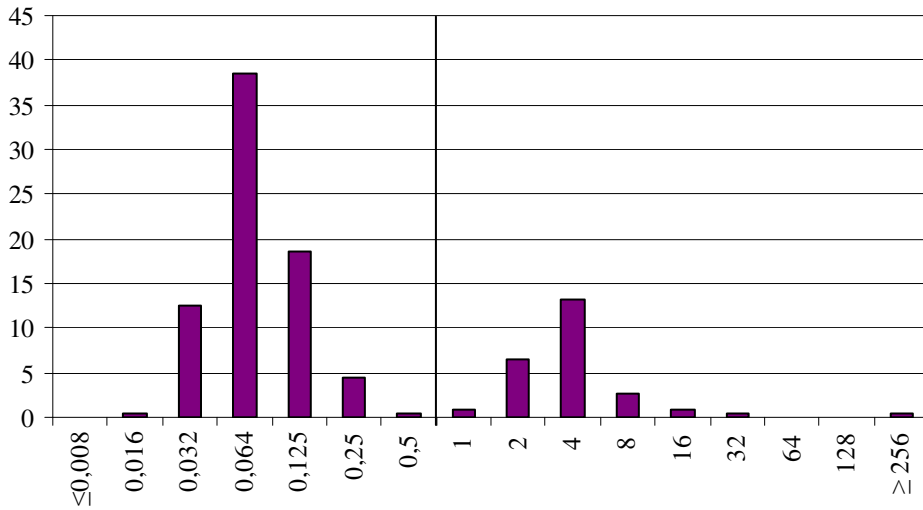
FIGURE 32. Distribution (%) of minimum inhibitory concentrations (mg/L) of fusidic acid in *Staphylococcus aureus* isolates from wound samples. The vertical bar indicates the breakpoint for resistance.
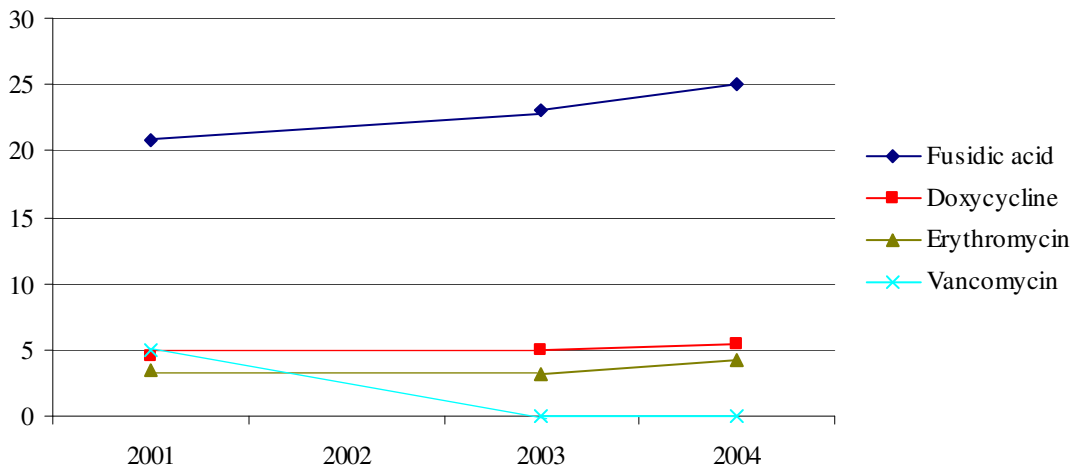

FIGURE 33. Prevalences (%) of non-susceptibility to various antibiotics other than beta-lactam in *S. aureus* isolates from wound specimens 2001 - 2004.

Escherichia coli in urine

TABLE 67. *Escherichia coli* urinary tract isolates (n=1,101). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)
	S	R	S	I	R	
Ampicillin	≥ 29	≤ 13	0.6	74.4	25.0	6 - 37
Ciprofloxacin	≥ 25	≤ 20	96.2	1.4	2.3	6 - ≥ 45
Mecillinam	≥ 20	≤ 16	92.1	5.6	2.3	6 - ≥ 45
Nalidixic acid	≥ 17	≤ 16	95.8	-	4.2	6 - ≥ 45
Nitrofurantoin	≥ 19	≤ 18	98.4	-	1.6	6 - 44
Sulfonamide	≥ 19	≤ 14	77.4	0.5	22.1	6 - 43
Trimethoprim	≥ 21	≤ 20	83.8	-	16.2	6 - 41

*S=Susceptible, I=Intermediately susceptible, R=Resistant.

TABLE 68. *Escherichia coli* urinary tract isolates (n=1,101). Distribution (%) of zone diameters (mm).*

	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Ampicillin	23.0	0.9	0.1	0.2	0.5	0.1	0.3	1.4	2.1	2.6	4.3	5.4	7.6	8.2	13.2
Ciprofloxacin	1.6	0.4	0.1			0.1		0.1	0.1						
Mecillinam	0.6			0.1			0.3	0.3	0.5	0.6	0.7	1.1	1.4	0.9	1.5
Nalidixic acid	3.7	0.2	0.2				0.1						0.3	0.1	0.3
Nitrofurantoin	0.2			0.1		0.1	0.2	0.1	0.1	0.1	0.3	0.1	0.5	0.5	1.8
Sulfonamide	20.7	1.1	0.1	0.1			0.1	0.1		0.1		0.1	0.2	0.1	0.9
Trimethoprim	14.7	0.7		0.1				0.1	0.1	0.1			0.1	0.2	0.2

	21	22	23	24	25	26	27	28	29	30	31	32	33	34	≥ 35
Ampicillin	5.6	7.0	5.4	3.9	2.7	1.9	1.0	1.0	1.1	0.2	0.2	0.1			0.1
Ciprofloxacin		0.1	0.3	0.1	1.0	0.6	0.6	0.6	0.6	3.5	2.5	4.9	8.8	12.7	61.6
Mecillinam	1.4	1.8	1.6	3.4	2.8	3.2	2.8	4.4	5.1	10.2	8.6	13.2	9.6	10.1	13.9
Nalidixic acid	0.6	1.2	2.9	5.2	8.6	13.0	14.4	17.9	9.3	12.1	3.5	3.5	1.3	1.1	0.8
Nitrofurantoin	1.7	3.9	4.7	9.8	12.2	16.7	13.8	11.8	8.3	7.6	1.8	1.6	0.7	0.3	1.4
Sulfonamide	0.7	2.0	2.9	6.1	5.9	8.5	5.1	9.5	5.1	10.6	3.3	5.3	3.4	3.0	5.3
Trimethoprim		0.1	0.3	0.5	0.8	1.6	2.1	5.8	5.1	13.8	8.4	12.7	8.7	9.1	14.8

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).

RESULTS AND COMMENTS

Urinary tract isolates of *E. coli* are analysed by disk diffusion in NORM. This protocol is used to obtain the dual aims of conducting surveillance and of improving the quality of routine susceptibility testing in Norwegian diagnostic laboratories. As a consequence of the NORM results from previous years, the breakpoint for susceptibility to ampicillin was adjusted from S ≥ 24 mm to S ≥ 29 mm and the breakpoint for resistance from R ≤ 12 mm to R ≤ 13 mm. As for blood culture isolates, the new protocol practically eliminated the ampicillin susceptible category for urinary tract *E. coli*. This does not imply that ampicillin cannot be used in the treatment of uncomplicated lower urinary tract infections. However, ampicillin should not be used as monotherapy in complicated and/or upper urinary tract infections. Approximately 25% of all isolates were high-level resistant to this agent, which is essentially unchanged from earlier years.

Extended-spectrum beta-lactamase positive isolates are occasionally recovered from routine urinary samples. Only two out of 1,101 (0.2%) were ESBL positive in NORM 2004. The technical aspects of ESBL detection are discussed in a separate section of this report.

Fluoroquinolones are used as second-line urinary tract antimicrobials in Norway. The prevalence of non-susceptibility increased from 2.6% in 2003 to 3.7% in 2004, but this change was mainly due to an increase in the category of intermediate susceptibility (0.6% in 2003, 1.4% in 2004). Nalidixic acid was used for screening of quinolone resistance, and the prevalence of resistance to nalidixic acid remained stable at 4.2% compared to 4.9% in 2003. The increase of intermediate susceptibility to ciprofloxacin may therefore be a technical issue rather than a true change in the bacterial population. Only two isolates were discrepantly reported as susceptible to nalidixic acid and resistant to ciprofloxacin.

There were no significant changes for nitrofurantoin, sulfamethoxazole and trimethoprim. Mecillinam was again a methodological challenge due to problems with standardisation and reproducibility.

Mycobacterium tuberculosis

A total of 302 cases of tuberculosis were reported to the Norwegian Surveillance System for Communicable Diseases (MSIS) in 2004. 282 of these patients had not previously been treated with antituberculosis drugs, out of which *Mycobacterium tuberculosis* was isolated from 234. All these isolates were tested for susceptibility.

TABLE 69. Antimicrobial susceptibility of 234 strains of *M. tuberculosis* complex isolated in 2004 from patients not previously treated for tuberculosis.

Geographical origin of patient	No. of isolates	Resistance to antimicrobial agents (isolates)				
		Isoniazid	Rifampicin	Ethambutol	Streptomycin	MDRTB
Norway	48				1	
Europe outside Norway	21	4	2	2	4	2
Asia	61	2			7	
Africa	101	13	2	1	12	2
America	3					
Total	234	19	4	3	24	4
Proportion of isolates resistant %		8	2	1	10	

Susceptibility tests were also performed on *Mycobacterium tuberculosis* isolates from 20 patients who had previously received antituberculosis drug treatment. Out of these, one isolate from an Asian patient was monoresistant to rifampicin and one Norwegian born patient had an isolate monoresistant to streptomycin. The rest of the isolates were sensitive to the first line drugs.

Appendix 1:
Collection of data on usage of antimicrobial agents in animals

Data sources
Feed additives
Until 2003, the Norwegian Agricultural Inspection Service was responsible for the collection of data on sales of antimicrobial growth promoters and coccidiostats as feed additives, while from that time the Norwegian Food Safety Authority has been in charge of collecting such data. Reliable data on the use of different substances and categories of feed additives was obtained from these sources.

Antimicrobial agents for therapeutic use
In Norway, veterinary antimicrobials for therapeutic use in domestic animals or farmed fish are prescription agents only. Moreover, veterinary antimicrobial agents have to be dispensed through pharmacies, which are only supplied by drug wholesalers. An exemption from the pharmacy/wholesaler monopoly has been granted for medicated feeds (i.e., feeds into which drugs for therapeutic use are mixed prior to sale). Medicated feeds have to be prescribed by veterinarians and produced by feed mills authorized by the Norwegian Medicines Agency. In Norway, medicated feeds produced and supplied by feed mills are only used for farmed fish. Medicated feeds for livestock are not produced in feed mills due to the small size of livestock herds compared to most other European countries. However, herd/flock treatment of livestock with antimicrobial agents is possible, again subject to veterinary prescription, through the drinking water or as a top dressing on the feed.
The sales figures for veterinary antimicrobials from wholesalers and feed mills are thought to roughly equal the use of these drugs. Sales figures for veterinary antimicrobials are therefore used as a synonym of veterinary antimicrobial use.
Drug wholesalers and feed mills report their sales figures to the Norwegian Institute of Public Health. This reporting

was made mandatory from January 1st 2002 to ensure that all the data are included. Crude sales data of veterinary antimicrobial agents were obtained from the Norwegian Institute of Public Health and calculated to express kg active substance.

Drug classification system
The Anatomical Therapeutic Chemical (ATC) classification system was used to categorize veterinary medicinal products (ATCvet).

Unit of measurement
The amount of active substance denoted in kilograms was chosen as the unit of measurement. Veterinary antimicrobial usage was calculated from sales figures for delivery of antimicrobials from wholesalers to pharmacies, and from feed mills. The data for benzyl penicillin salts and esters (procaine penicillin and penethamate hydriodide) were converted to the corresponding values for benzyl penicillin.

Inclusion criteria – veterinary drugs
All veterinary antimicrobial specialities included in this report belong to the following ATCvet groups: gastrointestinal infections (QA07AA), uterine infections (QG01AA+AE), and antimicrobial agents for systemic use (QJ), including intramammary dose applicators (QJ51). The QJ-group also includes medicated feeds and premixes for farmed fish that are approved by the drug authorities and classified as pharmaceutical specialities (QJ01).
Dermatological preparations (QD) and preparations intended for sensory organs (QS) are not included in the material. Human antimicrobial preparations can be used in small animal practice. However, data on the use of these drugs in animals are not included in this report as such usage cannot be separated from sales for use in humans.

Appendix 2:
Collection of data on human usage of antimicrobial agents

Data sources
In Norway, antibacterials are prescription only medicines (POM), and only allowed sold by pharmacies. The data cover total sales of antibacterials for humans in Norway and are based on sale of medicaments from drug wholesalers to pharmacies and hospitals in Norway. The figures presented should be regarded as maximum figures with the assumption that all medicaments sold are actually consumed. The actual drug consumption will probably be somewhat lower.
The data are collected by the Norwegian Institute of Public Health. Data on drug use have been collected since the beginning of the seventies.

Drug classification
The data are categorized according to the ATC classification system. Defined Daily Doses (DDDs) are employed as units of measurement. The ATC/ DDD index version 2005 is used.

Unit of measurement
The ATC/DDD system is recommended by the WHO to serve as a tool for drug utilisation research in order to improve quality of drug use. One component of this is the presentation and comparison of drug consumption statistics at international and other levels.

The use of defined daily dose, DDD, as a unit of measurement, simplifies and improves the evaluation of drug consumption over time, nationally and internationally. The DDD is a theoretical unit of measurement, and does not necessarily reflect the recommended or Prescribed Daily Dose.

The basic **definition** of the unit is:
The DDD is the assumed average maintenance dose per day for a drug used for its main indication in adults.

The DDDs for the antiinfectives are as a main rule, based on the use in infections of moderate severity. Some antiinfectives are only used in severe infections and their DDDs are assigned accordingly. The DDDs assigned are based on daily treatment.

Inclusion criteria
The antimicrobials for human use included in this report belong to ATC J01 antimicrobials for systemic use. Oral vancomycin (A07AA09) and oral and rectal metronidazole (P01AB01) are also included. Of the antimycobacterials, only rifampicin is included, as the total amount of rifampicin used. Antimicrobials used in dermatological preparations (ATC group D) and preparations intended for sensory organs (ATC group S) are not included in the material.

Appendix 3:
Sampling, microbiological methods and data processing in NORM-VET

Sampling strategy

Clinical isolates of *Staphylococcus intermedius* and *Escherichia coli* from animals included in the NORM-VET monitoring programme 2004 were collected from diagnostic submissions:

S. intermedius from dogs with "first time" skin infections were collected from five small animals clinics geographically spread throughout Norway. Each clinic was asked to submit 30 double samples (swabs from skin and faeces) from dogs with a non-treated first time pyoderma. Only dogs with no antimicrobial treatment during the last six month and only one dog from each owner were included.

E. coli isolates were collected from pigs with a diagnosis of either enteritis or oedema disease, and from poultry with a diagnosis of septicaemia. Each isolate originated from a unique flock or herd but were not always from a unique production site.

The isolates of indicator bacteria (*E. coli* and *Enterococcus* spp.) included in the NORM-VET monitoring programme 2004 were collected from healthy pigs and broilers (faecal and meat samples). The sampling period was from January to December. Personnel from the Norwegian Food Safety Authority collected the faecal and meat samples from pigs at slaughterhouses. To obtain a representative random sample from pigs, the number of samples collected at each slaughterhouse were determined by the proportion of animals slaughtered there, relative to the total number of animals slaughtered in Norway in 2003. Abattoirs that slaughtered >1% of the total delivered slaughter in 2003 were included. The faecal samples from broiler livestock were systematically sampled at four of the Regional Laboratories at the National Veterinary Institute from samples collected according to the Norwegian Salmonella control programme for live animals. The first sample on a specific weekday during the whole sampling period was collected at each laboratory. The number of samples from each laboratory was proportional to the total number of samples from broilers obtained within the Norwegian *Salmonella* Control programme for live animals for each laboratory in the previous year. The meat samples from broilers were systematically sampled from samples collected at retail level according to the *Campylobacter* action plan. Each of the four participating local Food Safety Authorities sampled appr. 50 gram of each product from the first five samples collected during the months February, April, June, August and November.

Isolation and identification of bacteria
Escherichia coli

The *E. coli* isolates included in NORM-VET 2004 were isolated and identified at the National Veterinary Institute. Meat: Five grams of material from each specimen were incubated in 45 ml of MacConkey broth (Oxoid). After incubation at 44°C for 24 h, a small amount (approx. 10 μl) of broth was plated onto the surface of lactose-saccarose-bromthymol blue agar. Faeces: Intestinal content was gathered on swabs and plated directly onto the surface of lactose-saccarose-bromthymol blue agar without broth enrichment.

After incubation of the agar plates at 37°C for 24 h, a typical colony was plated onto blood agar (Heart infusion agar (Difco) containing 5% bovine blood). Colonies were identified as *E. coli* by typical appearance, lactose and/or saccarose fermentation and a positive indole reaction.

Enterococcus **spp.**

The enterococcal isolates included in NORM-VET 2004 were isolated and identified at the National Veterinary Institute. Meat: Five grams of material from each specimen were incubated in 45 ml of Azide dextrose broth (Oxoid). After incubation at 44°C for 24 h, a small amount (approx. 10μl) of broth was plated onto the surface of Slanetz & Bartley agar (Oxoid). Faeces: Intestinal content was gathered on swabs and plated directly onto the surface of Slanetz & Bartley agar (Oxoid) without broth enrichment.

After incubation of the agar plates at 44°C for 48h, a typical colony was plated onto blood agar (Heart infusion agar (Difco) with 5% bovine blood). Typical colonies were tested by negative catalase reaction and *E. faecium* and *E. faecalis* were identified by ddl-PCR (Dutka-Malen et al., 1995).

For the selective isolation of vancomycin resistant enterococci (VRE), the samples were treated as described above, and plated out on additional Slanetz and Bartley's agar plates containing 32 mg/L vancomycin. One colony of VRE from each positive sample was selected, and the isolates confirmed as *Enterococcus* spp. by phenotypic characterisation. The isolates were further identified to species level and tested for the presence of the vanA gene using PCR (Dutka-Malen et al, 1995, Simonsen et al, 2000).

Staphylococcus intermedius.

The swab samples were submitted to the National Veterinary Institute and plated directly onto the surface of blood agar (heart infusion agar (DIFCO) with 5% bovine blood). The plates were incubated in 5% CO_2 atmosphere at 37°C for 16-24 hrs. Greyish white typical colonies with a beta-toxic zone on blood agar were isolated and tested for production of catalase and coagalase, β-galactosidase and fermentation of D-mannitol.

Susceptibility testing

Only one isolate per herd or product were tested for antimicrobial susceptibility. Bacterial isolates were tested for antimicrobial susceptibility at the National Veterinary Institute. The VetMIC[TM] microdilution method (Dept. of Antibiotics, National Veterinary Institute, Sweden) was used for the susceptibility testing of all isolates. *S. intermedius* was tested for production of beta-lactamase using the cloverleaf method. All isolates of *S. intermedius* resistant to erythromycin were tested for inducible clindamycin resistance.

Microbiological cut-off values were used to classify the isolates as resistant or susceptible (Appendix 6). The microbiological cut-off value is defined as the highest MIC-value of isolates that belong to the original genetically unchanged population (wild-type). It classifies

the isolates with a MIC-value greater than the microbiological cut-off value as resistant.

Quality assurance systems

The following bacteria were included as quality controls on a weekly basis, *E. coli* ATCC 25922, *E. faecalis* ATCC 29212 and *S. aureus* ATCC 29213. The following resistant bacteria were tested on a regular basis: *E. faecium* CCUG 33829, CCUG 36804 and *S. aureus* CCUG 35603. The results were approved according to reference values given by CLSI when available.

The participating laboratories at the National Veterinary Institute are accredited according to the requirements of NS-EN ISO/IEC 17025 and participate in external quality assurance programmes for veterinary pathogens. The programmes are organized by the VLQAS (Veterinary Laboratories Agency Quality Assurance Unit, Loughborough, England) and ARBAO-II http://www.dfvf.dk/Default.asp?ID=9753.

Data processing

Susceptibility test results were recorded and processed in WHONET 5.3, a program developed by the World Health Organization (WHO) for analysis of antimicrobial resistance data (http://www.who.int/drugresistance/whonetsoftware/en/index.html). The susceptibility data were stored as continuous values (MIC). In addition data was imported into SAS, Enterprise guide 3.0 to obtain exact 95% confidence intervals for the prevalence's of resistance. For this purpose the susceptibility data were categorised as susceptible or resistant, respectively, as defined by the relevant microbiological cut-off value. The function Proc freq, using exact binomial proportion test for one-way tables, was used for the calculation of prevalences of resistance including 95% confidence intervals.

Appendix 4:
Sampling, microbiological methods and data processing of zoonotic and non-zoonotic enteropathogenic bacteria in NORM and NORM-VET

Sampling strategy - animals
Salmonella
Samples from animals were collected according to the Norwegian Salmonella control programme for live animals. Additional samples were obtained from animals during clinical examinations or necropsies at the National Veterinary Institute. One isolate of each serovar per incident was included for susceptibility testing.

Campylobacter jejuni
As part of the Norwegian action plan against Campylobacter in broilers (www.zoonose.no), samples from chickens were collected at farm level (faecal samples) or at slaughter plants (caecal samples), and samples from fresh broiler products were collected at retail level. One isolate per positive farm or batch of products was included for susceptibility testing.

Sampling strategy - humans
Salmonella, Yersinia enterocolitica and Shigella
All the human isolates were obtained from clinical specimens. One isolate per patient was included for susceptibility testing.

Campylobacter
A total of 250 human isolates were obtained from clinical specimens. Five regional laboratories submitted the first five isolates each month to the National Reference Laboratory for Enteropathogenic Bacteria at the Norwegian Institute of Public Health. One isolate per patient was included for susceptibility testing.

Isolation and identification of bacteria
Isolation and identification of Salmonella from animals was carried out by the National Veterinary Institute according to the Nordic Committee on Food Analyses (NMKL) method number 71. Isolation of Campylobacter spp. from broiler and broiler products was carried out by the National Veterinary Institute or local food control laboratories according to the Nordic Committee on Food Analyses (NMKL) method number 119, with minor modifications. Identification of C. jejuni was carried out by the National Veterinary Institute or the Norwegian Institute of Public Health.
Isolation and identification of bacteria from humans were performed according to conventional methods described in standard reference literature (e.g. the ASM Manual of Clinical Microbiology, Edwards and Ewings Identification of Enterobacteriaceae). The identification of all isolates from animals and humans were verified at the National Reference Laboratory for Enteropathogenic Bacteria at the Norwegian Institute of Public Health.

Susceptibility testing
The isolates from animals were tested for antimicrobial susceptibility at the National Veterinary Institute. MIC values were obtained using the VetMIC^{TM} microdilution method (Dept. of Antibiotics, National Veterinary Institute, Sweden).

The Salmonella, Yersinia and Shigella isolates from humans were tested for antimicrobial susceptibility at the Norwegian Institute of Public Health by an agar disk diffusion test using PDM II agar plates and PDM disks (AB Biodisk, Solna, Sweden). The Campylobacter isolates from humans were tested for antimicrobial susceptibility at the Norwegian Institute of Public Health by using Etest (AB Biodisk).

For animal isolates, microbiological cut-off values were used to classify the isolates as resistant or susceptible (Appendix 6). The microbiological cut-off value is defined as the highest MIC-value of isolates that belong to the original genetically unchanged population (wild-type). It classifies the isolates with a MIC-value greater than the microbiological cut-off value as resistant. For human isolates, MIC-breakpoints defined by AFA (Norwegian Reference Group on Antibiotic Susceptibility Testing) were applied when available and appropriate. For disk diffusion results, population based breakpoints were used. Breakpoints for Campylobacter are preliminary and based on MIC-distributions.

Quality assurance systems
The National Veterinary Institute and the Reference Laboratory for Enteropathogenic Bacteria at the Norwegian Institute of Public Health have a quality assurance system according to the requirements of NS-EN ISO/IEC 17025.
Campylobacter jejuni subsp. jejuni CCUG 33057 and CCUG 11284 were used as quality control at the National Veterinary Institute on a weekly basis. The National Veterinary Institute participates in an external quality assurance programme for veterinary pathogens organized by the VLQAS (Veterinary Laboratories Agency Quality Assurance Unit, Loughborough, England) and participates also in the external quality assurance programmes organized by http://www.dfvf.dk/Default.asp?ID=9753 and Global Salm-Surv (GSS) http://www.who.int/salmsurv/en/. The Norwegian Institute of Public Health participates in the external quality assessment programme for Salmonella organized by Enter-Net.

Data processing
Susceptibility data were recorded and processed in WHONET5.3, a program developed by the World Health Organization (WHO) for analysis of antimicrobial resistance data (http://www.who.int/drugresistance/whonetsoftware/en/index.html). The susceptibility data were stored as continuous values (MIC). In addition the animal data concerning C. jejuni was imported into SAS, Enterprise guide 3.0 to obtain exact 95% confidence intervals for the prevalence's of resistance. For this purpose the susceptibility data were categorised as susceptible or resistant, respectively, as defined by the relevant cut-off value. The function Proc freq, using exact binomial proportion test for one-way tables, was used for the calculation of prevalences of resistance including 95% confidence intervals.

Appendix 5:
Sampling, microbiological methods and data processing in NORM

General considerations
NORM is based upon periodic sampling of bacteria from patients with respiratory tract infections, wound infections, urinary tract infections, or septicaemiae. For enteric infections see Appendix 4. 2004 was the fifth year of surveillance, and all twenty-four laboratories in Norway participated in the surveillance system in addition to the Norwegian Institute of Public Health. The surveillance strategy is based on sampling and local testing of bacterial isolates from defined clinical conditions. All laboratories follow the same sampling strategy and use identical criteria for the inclusion of bacterial strains. Only one isolate per patient and infectious episode was included. All bacteria were identified using conventional methods as described in the ASM Manual of Clinical Microbiology. The surveillance period started in the beginning of January, and consecutive bacterial isolates were included up to a defined maximum of isolates for each surveillance category. The surveillance categories in 2004 were: *E. coli*, *Klebsiella* spp., *Staphylococcus aureus*, *Streptococcus pneumoniae* and *Enterococcus* spp. from blood cultures; *Streptococcus pyogenes* and *Haemophilus influenzae* from respiratory tract infections, *S. aureus* and *S. pyogenes* from wound infections, and *E. coli* from urinary tract infections. Blood culture isolates, respiratory tract isolates and isolates from wound specimens were tested using Etest, while isolates from urinary tract infections were examined by a disk diffusion method in accordance with the Norwegian Reference Group on Antibiotic Susceptibility Testing (AFA). All resistance values were recorded either as MICs or mm inhibition zone sizes in order to monitor trends in the occurrence of resistance. Suspected MRSA (*S. aureus* with oxacillin MIC $\geq$ 4 mg/L) were examined by *mecA* PCR, and suspected VRE (enterococci growing on BHI with 6 mg/L vancomycin) were examined by PCRs for *van* genes. The NORM computer program was used for the registration of patient data, sample data and resistance data. Data were analyzed by WHONET5 with the aid of the NORMlink program thus converting the data base structure of NORM to a single file format. Both WHONET and NORMlink were developed by John Stelling. The distribution of bacterial species in blood culture was based on extraction of routine data from the laboratory information systems of the participants. All isolates of the same species recovered within 1 month after the initial finding was considered duplicates and omitted from the survey. No attempt was made to evaluate the clinical significance of each finding.

Blood culture isolates
Consecutive isolates of up to 50 each of *E. coli*, *S. aureus*, and *S. pneumoniae*, up to 25 isolates of *Klebsiella* spp., and up to 20 isolates of *Enterococcus* spp. were included in the surveillance from January until testing time in October. All isolates were tested using Etest (AB Biodisk, Solna, Sweden). A total of 982 isolates of *E. coli*, 359 isolates of *Klebsiella* spp, 637 isolates of *S. aureus* and 294 isolates of enterococci were tested on PDM agar at 35°C in ambient air, while 628 isolates of pneumococci were tested on PDM (AB Biodisk) agar with 5% lysed horse blood at 35°C in 5% CO_2. All *E. coli* and *Klebsiella*

spp. isolates were tested for ESBL production using a disk approximation test including amoxicillin/clavulanic acid, aztreonam, ceftazidime, cefotaxime, cefpodoxime and cefpirome. All *S. aureus* isolates were tested for beta-lactamase production using the nitrocefin disk, the acidometric agar plate (3.6 mg/L penicillin G and phenol red) or the clover leaf method. All *S. aureus* isolates were screened for methicillin resistance using MH agar (Difco) with 4% NaCl and oxacillin 4 mg/L and a spot inoculum of 10^6 cfu/spot. All enterococci were screened for vancomycin resistance using BHI agar (Difco) and vancomycin 6 mg/L. The following strains were used for quality control: *E. coli* ATCC 25922, *K. pneumoniae* ATCC 700603 (ESBL positive), *E. faecalis* ATCC 29212, *S. pneumoniae* ATCC 49619, *S. aureus* ATCC 29213, *S. aureus* ATCC 43300 (heterogeneous MRSA), and *S. aureus* CCUG 35600 (homogeneous MRSA).

Respiratory tract isolates
Up to 25 consecutive isolates each of *S. pyogenes* and *H. influenzae* from patients with respiratory tract infections were collected in each laboratory from January to March. All isolates were kept in a freezer and tested in batch using Etest on PDM II agar supplemented with 5% lysed horse blood (*S. pyogenes*) or 1% haemoglobin and 1% Isovitalex (*H. influenzae)* followed by incubation at 35°C in 5% CO_2. A total of 474 *S. pyogenes* and 513 *H. influenzae* isolates were included. *S. pneumoniae* ATCC 49619 and *H. influenzae* ATCC 49247 were used for quality control.

Wound specimens
Up to 50 consecutive isolates of *S. aureus* and 25 isolates of *S. pyogenes* from patients with wound infections were collected in each laboratory from January to March. All isolates were kept in a freezer and tested in batch using Etest. A total of 1,136 *S. aureus* and 503 *S. pyogenes* were included in the study. The isolates were analysed as described for blood culture isolates (*S. aureus*) and respiratory tract isolates (*S. pyogenes*).

Urinary tract isolates
Up to 50 consecutive isolates of *E. coli* from patients with urinary tract infections were collected in each lab during January and February. All isolates were either kept on bench or in a freezer until tested in batch using a disk diffusion method with PDM II agar and paper disks (AB Biodisk) at 35°C in ambient air. ESBL production was examined by the disk approximation test described for blood culture isolates. The study included 1,101 isolates, and *E. coli* ATCC 25922 was used for quality control.

Mycobacterium tuberculosis
In the year 2004, antimicrobial susceptibility testing of *M. tuberculosis* was performed at the following institutions: Norwegian Institute of Public Health, Oslo, Ullevål University Hospital, Oslo, National Hospital, Oslo, and Haukeland Hospital, Bergen. The majority of isolates were tested using the BACTEC (Norwegian Institute of Public Health and Ullevål University Hospital) or MGIT systems (National Hospital). All four laboratories participate in an external quality control program organized by the WHO.

Appendix 6:
Breakpoints NORM-VET

For classification as resistant or susceptible, the following microbiological cut-off values were applied in this report. A microbiological cut-off value is defined as the highest MIC-value of isolates that belong to the original genetically unchanged population (wild-type). It classifies the isolates with a MIC-value greater than the microbiological cut-off value as resistant. For details regarding bacteria and antimicrobial panels, see the tables in the text.

Antimicrobials	Resistant (MIC values, mg/L)	Campylobacter	E. coli / Salmonella	Staphylococcus intermedius	Enterococcus
Oxytetracycline	> 2	■		■	
	> 4				■
	> 8		■		
Chloramphenicol	> 16		■	■	■
Florfenicol	> 16		■		
Ampicillin	> 8		■		■
	> 16	■			
Penicillins	Based upon beta-lactamase production			■	
Oxacillin	> 2			■	
Cephalothin	> 1			■	
Ceftiofur	> 2		■		
Trimethoprim	> 4		■		
	> 8				
Sulfonamides	> 256		■		
Erythromycin	> 2			■	
	> 4				■
	> 8	■			
Clindamycin	> 2			■	
Streptomycin	> 8		■*		
	> 32		■*	■	
	> 1024				■
Gentamicin	> 2			■	
	> 4	■	■		
	> 512				■
Neomycin	> 2			■	
	> 4		■		
	> 512				■
Enrofloxacin	> 0.25		■		
	> 0.5	■		■	
Nalidixic acid	> 16	■	■		
Vancomycin	> 4			■	■
Fusidic acid	> 0.5			■	
Avilamycin	> 16			■	■
Bacitracin	> 32				■
Flavomycin	> 32				■
Virginiamycin	> 4			■	
	> 8				■
Narasin	> 2				■

*> 8 for *Escherichia coli*, > 32 for *Salmonella* spp.

Appendix 7:
Breakpoints NORM

Breakpoints for antimicrobial resistance used in this report. NORM data are categorized according to the breakpoints of the Norwegian Reference Group on Antibiotic Susceptibility Testing – AFA (isolates from humans). Breakpoints for *Campylobacter* are preliminary and based on MIC-distributions. For details regarding bacteria and antimicrobial panels, see tables in text.

Antimicrobials	MIC values mg/L		*E. coli/Klebsiella*	*Staphylococcus*	*S. pneumoniae*	*S. pyogenes*	*Enterococcus*	*Campyloacter*
	S	R						
Amoxi./clav.	≤ 0.5	≥ 32	■					
Ampicillin	≤ 1	≥ 32	■					
	≤ 2	≥ 16					■	
Cefpirome	≤ 1	≥ 32	■					
Cefotaxime	≤ 0.5	≥ 4			■			
Ceftazidime	≤ 1	≥ 32	■					
Cefuroxime	≤ 0.5	≥ 4			■			
	≤ 1	≥ 32	■					
	≤ 2	≥ 8		■				
Chloramphenicol	≤ 4	≥ 8			■			
Ciprofloxacin	≤ 0.125	≥ 4	■		■			
	≤ 1	≥ 4						■
Clindamycin	≤ 0.25	≥ 4			■	■		
	≤ 1	≥ 4		■				
Doxycycline	≤ 1	≥ 4		■	■	■		
	≤ 2	≥ 4						■
Erythromycin	≤ 0.5	≥ 1			■	■		
	≤ 1	≥ 4		■				
	≤ 0.5	≥ 8						■
Fusidic acid	≤ 0.5	≥ 1		■				
Gentamicin	≤ 2	≥ 8	■	■				
	≤ 512	≥ 1024					■	
	≤ 4	≥ 8						■
Meropenem	≤ 0.5	≥ 4	■					
Nalidixid acid	≤ 16	≥ 32						■
Oxacillin	≤ 2	≥ 4		■				
Penicillin	≤ 0.064	≥ 0.25		■				
	≤ 0.064	≥ 2			■	■		
	≤ 4	≥ 16					■	
Piperacillin/tazo.	≤ 8	≥ 32	■					
Streptomycin	≤ 512	≥ 1024					■	
TMS	≤ 0.5	≥ 4			■	■		
	≤ 2	≥ 16	■					
Vancomycin	≤ 2	≥ 8			■			
	≤ 4	≥ 16		■			■	

Disk diffusion testing Antimicrobials (amount in disks)	Breakpoints (mm)		*Salmonella/ Shigella/ Yersinia*	Urine *E.coli Klebsiella Enterococcus*
	S	R		
Ampicillin (10 µg)	≥ 32	≤ 12	▪	▪
Chloramphenicol (30 µg)	≥ 20	≤ 19	▪	
Ciprofloxacin (10 µg)	≥ 27	≤ 18	▪	▪
Mecillinam (10 µg)	≥ 20	≤ 16		▪
Nalidixic acid (30 µg)	≥ 17	≤16	▪	▪
Nitrofurantoin (100 µg)	≥ 19	≤ 18		▪
Sulfonamide (250 µg)	≥ 19	≤ 14		▪
Tetracycline (30 µg)	≥ 20	≤ 16	▪	
Trimetoprim (5 µg)	≥ 21	≤ 19		▪
Trimethoprim /Sulfa (1,2/23,8 µg)	≥ 20	≤ 12	▪	