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Usage of Antimicrobial Agents and Occurrence of Antimicrobial Resistance in Norway

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The NORM and NORM-VET programmes are part of the
Norwegian Government's Action Plan against Antimicrobial Resistance (2000 – 2004) issued in 2000, and the
National Strategy for Prevention of Infections in the Health Service and Antibiotic Resistance (2008 – 2012) issued in 2008.

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 following use of antimicrobial agents 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 drug usage and resistance in one ecological 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 data on usage and resistance in human and veterinary medicine, as well as microbes in food production.

Several countries have implemented monitoring programmes for antimicrobial resistance and usage of antimicrobial agents 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. 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 drug usage and resistance in both human and veterinary medicine and 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 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 forms the basis for containment of antimicrobial resistance in the years to come. The need for continued surveillance of both resistance and drug usage was emphasized. An integrated national strategy for prevention of infections in the health service and antibiotic resistance (2008 – 2012) was issued in the summer of 2008.

The NORM surveillance programme for antimicrobial resistance in human pathogens was established in 1999 and is coordinated by the Department of Microbiology and Infection Control 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, i.e. coccidiostatic growth promoters, are collated at the Norwegian Food Safety Authority.

This report, which is the ninth annual joint report from NORM and NORM-VET, presents data for 2008. 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 usage of antimicrobial agents 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 2009

II. SAMMENDRAG

Dette er den niende felles rapporten fra Norsk overvåkingssystem for antibiotikaresistens hos mikrober (NORM) og Norsk overvåkingsprogram for antibiotikaresistens i mikrober fra fôr, dyr og næringsmidler (NORM-VET). Rapporten presenterer data over forekomst av antibiotikaresistens og forbruk av antibiotika til mennesker og dyr i 2008. Data fra relevante prosjekter som ikke er med i de kontinuerlige overvåkingsprogrammene, presenteres også.
Både NORM og NORM-VET programmene er deler av Regjeringens tiltaksplan mot antibiotikaresistens som ble offentliggjort i 2000. NORM koordineres av Avdeling for mikrobiologi og smittevern, Universitetssykehuset Nord-Norge i Tromsø. NORM-VET koordineres av Zoonosesenteret ved Veterinærinstituttet i Oslo. 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 veterinære antibiotika til terapeutisk bruk på landdyr i 2008 var 6219 kg. Fra 1995 til 2001 ble salget av veterinære antibiotika til landdyr redusert med ca 40 %. Etter dette har forbruket holdt seg relativt konstant. Forskrivningsmønsteret har utviklet seg i gunstig retning siden 1995; det vil si at andelen penicillinbruk har økt. Rene penicillinpreparater utgjorde 46 % av salget av veterinære antibiotika til landdyr i 2008, og av dette var 86 % beta-laktamase følsomme penicilliner. Forbruket av tetracykliner utgjorde kun 4 %. Nedgangen i antibiotikaforbruket og endringene i forskrivningsmønsteret skyldes først og fremst at husdyrnæringen i andre halvdel av 1990-tallet gjennomførte systematiske kampanjer for å redusere forbruket av antibiotika til dyr og for riktig bruk av antibiotika.
Totalsalget av veterinære antibiotika til terapeutisk bruk hos oppdrettsfisk i Norge var i 2008 på 941 kg aktiv substans, hvorav 72 % var kinoloner. Forbruket av antibiotika i oppdrettsnæringen er redusert med 98 % siden 1987. Denne betydelige reduksjonen kan tilskrives innføringen av effektive vaksiner til laks og ørret samt andre infeksjonsforebyggende tiltak, herunder bedrede miljøforhold. Av all antibiotika rekvirert til oppdrettsfisk i 2008 ble 65 % brukt til torsk, 23 % til laks, 8 % til ørret og de resterende 4 % hovedsakelig til kveite og piggvar.
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 nesten fordoblet siden forbudet mot bruk av antibakterielle vekstfremmere, noe som kan forklares ved økt produksjon av broilere. Forbruksmønstret for koksidiostatika er endret fra monensin til narasin etter 1996. Narasin har de senere årene utgjort hovedparten av forbruket av ionofore koksidiostatika.

Forbruk av antibiotika hos mennesker

Totalsalget av antibiotika til systemisk bruk hos mennesker var i 2008 19,8 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. Fra 2004 har totalforbruket av antibiotika økt. Salget av penicilliner og kinoloner øker, mens salg av sulfonamider og trimetoprim synker. Det urinveisantiseptiske middelet metenamin øker også. I 2008 utgjorde metenamin 15 % av totalt salg.
I 2008 var 43 % av det totale antibiotikaforbruket i Norge penicilliner, målt i DDD. I 2008 så vi en økning for bredspektrede og penicillinase stabile penicilliner, de økte med henholdsvis 5 og 6 %. Tetracykliner utgjorde 16 % av totalforbruket i 2008. Forbruket av makrolider og linkosamider sank med 7 % i 2008 og utgjorde 11 % av totalt salg. Salget av cefalosporiner, monobaktamer og karbapenemer utgjør bare 3 % av totalsalget. Over år har det vært en markant økning i forbruket av fluorokinoloner. Denne gruppen utgjorde kun 4 % av totalforbruket i 2008, men salget er doblet siden 2000.
Bruken av antibakterielle midler varierer avhengig av kjønn, aldersgrupper og bosted. Salget til sykehus og allmennpraksis utgjorde i 2008 henholdsvis 9 og 91 % av totalsalget. Penicilliner sto for 43 % av antibiotikasalget målt i DDD til sykehus og 44 % i allmennpraksis. De viktigste andre gruppene på sykehus var cefalosporiner (20 %) og kinoloner (7 %), mens det i allmennpraksis var tetracykliner (18 %) og makrolider (12 %).

Resistens hos kliniske isolater fra dyr

De kliniske isolatene inkludert i 2008 var fra diagnostiske prøver av beta-haemolysin-produserende stafylokokker fra hud- eller øreinfeksjoner hos hund. De fleste av disse stafylokokkene ble identifisert som *Staphylococcus pseudintermedius*. Forekomsten av resistens hos beta-haemolysin-produserende stafylokokker var høy, der kun 13,5 % av isolatene var følsomme for alle testede antibiotika. Andelen multiresistente isolater (resistent mot to eller flere antibiotika) var 67 % av de undersøkte isolatene, og resistens mot penicillin og fusidin ble hyppigst identifisert.

Resistens hos indikatorbakterier fra dyr og mat

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 2008 ble avføringsprøver fra svin undersøkt for *E. coli* og *Enterococcus* spp., mens avføringsprøver fra hund ble kun undersøkt for *E. coli*.
Forekomsten av antibiotikaresistens i *E. coli* i Norge er lav hos hund og lav til moderat hos gris. Andelen av *E. coli* fra hundeavføring som var følsom for alle testede antibiotika var 83,1 %, mens i tilsvarende prøver fra gris var 77,5 % av isolatene følsomme. Den hyppigst forekommende resistensen hos *E. coli* fra hund var ampicillinresistens, mens streptomycinresistens var vanligst i *E. coli* fra gris. Videre ble en høyere andel av *E. coli* isolatene fra hund funnet å være resistent mot tre

eller flere antibiotika sammenliknet med isolater fra gris, men denne forskjellen er ikke statistisk signifikant. Ett av disse multiresistente *E. coli* isolatene fra hund hadde produksjon av bredspektret beta-laktamase (ESBL produksjon). Forekomst av bakterier med ESBL produksjon hos dyr er svært betenkelig.

Avføringsprøver fra gris viste tilstedeværelse av *Enterococcus* spp. i 45 % av prøvene, og disse ble hovedsakelig identifisert som *Enterococcus faecium*. Andelen av antibiotikaresistens i *Enterococcus* spp. var i 2008, som i tidligere år, moderat. Nesten halvparten av enterokokkene hadde nedsatt følsomhet for ett eller flere av antibiotikaene som det ble testet for. Tetracyklinresistens, tett fulgt av erytromycinresistens, var de vanligste formene for antibiotikaresistens hos *Enterococcus* spp. fra avføringsprøver fra gris.

Resistens hos zoonosebakterier og andre enteropatogene bakterier

I 2008 ble det gjort resistensbestemmelse av 15 *Salmonella* spp. isolater fra norske dyr. Tolv av isolatene var *S*. Typhimurium og syv av disse ble isolert fra hund, to fra storfe, to fra katt og ett isolat fra hest. De resterende tre *Salmonella* isolatene var *S*. London, *S*. Goldcoast og *S*. Hvittingfoss. Antibiotikaresistens hos *Salmonella* fra norske dyr er hovedsaklig, med ett unntak i 2008, knyttet til isolering av multiresistent *S*. Typhimurium DT104, noe som ble gjort i tre tilfeller. Ett tilfelle var fra en oppfølgingsprøve av storfe, mens de to resterende, samt ett ikke typbart isolat med kinolonresistens, ble isolert fra hund i 2008. Dette tyder på lav forekomst av resistens i tilfeldige funn av *Salmonella* i norske dyr.

Av de humane salmonellosetilfellene som ble rapportert i 2008, var 83,4 % oppgitt å ha blitt smittet i utlandet. Andelen *S*. Typhimurium isolater som var følsomme for alle antibiotika, var høyere for kategorien "smittet i Norge" (55,8 %) enn for kategorien "smittet i utlandet" (32,6 %). Multiresistens ble hyppigere påvist hos de utenlandssmittede (53 %) enn hos de innenlandssmittede (36 %). Resultatene for *S*. Typhimurium 2001-2008 indikerer en økende forekomst av resistens mot tetracykliner og ampicillin. Forekomsten av antibiotikaresistens var betydelig lavere hos *S*. Enteritidis enn hos *S*. Typhimurium med unntak av nalidiksinsyre. Til sammen 28,6 % av *S*. Enteritidis isolatene var resistente mot nalidiksinsyre. Sammenliknet med resultatene fra 2007, ble det påvist en signifikant reduksjon i andelen av ciprofloxacinresistente *S*. Enteritidis.

Resultatene fra 2008 viser at forekomsten av resistens hos *Campylobacter jejuni* fra norske broilere fremdeles er lav og stabil. Hele 92,2 % av isolatene var følsomme for alle undersøkte antibiotika. I likhet med *C. jejuni* isolater fra pasienter smittet i Norge ble det ikke påvist erytromycin- eller gentamicinresistente isolater fra norske broilere. Andelen kinolon- og tetracyklinresistente isolater var noe høyere i gruppen pasienter smittet i Norge enn hos norske broilere. Begge disse gruppene hadde betydelig lavere forekomst av antibiotikaresistens enn *C. jejuni* fra pasienter smittet i utlandet, hvor kun 27,0 % var følsomme for alle undersøkte antibiotika. Andelen multiresistente *C. jejuni* isolater fra pasienter smittet i utlandet er betydelig høyere enn hva som rapporteres fra "norske" isolater. Over halvparten av *C. jejuni* isolatene var ervervet i utlandet, og av disse var 47,6 % resistente mot tre eller flere antibiotika.

Smitte med *Yersinia enterocolitica* skjer hovedsakelig innenfor Norges grenser, og den vanligste serogruppen er O:3. Resistens mot nalidiksinsyre og trimetoprim-sulfamethoxazole er hyppigst identifisert. I 2008 ble alle tilfeller av *Shigella*-infeksjoner knyttet til smittekilder i utlandet. Antibiotikaresistens var utbredt hos *Shigella* isolater i likhet med hva som rapporteres fra andre land.

Resistens hos kliniske isolater fra mennesker

Forekomsten av antibiotikaresistente kliniske bakterie-isolater fra mennesker var, som i de foregående år, meget lav i 2008. Det ble påvist seks tilfeller av meticillin-resistente *Staphylococcus aureus* (MRSA) blant 871 blodkulturisolater (0,7 %) som ble inkludert i NORM-protokollen. Dette er i samsvar med at 10 av 1359 (0,7 %) *S. aureus* blodkulturisolater i laboratorienes datasystemer ble rapportert som MRSA. I 2008 var 10 av 1365 (0,7 %) *S. aureus* fra blodkultur og spinalvæske MRSA. Andelen har økt noe fra 2007 da det ble påvist 0,4 % MRSA. Meldesystemet for infeksjonssykdommer (MSIS) registrerte 348 tilfeller av MRSA-infeksjon i 2008 hvilket er uendret fra 2007 da det ble registrert 342 tilfeller. Hele 88 % av disse tilfellene var pasienter med sårinfeksjoner og abscesser. MRSA utgjør fortsatt en svært liten andel av *S. aureus* isolater fra sårprøver (7/1061, 0,7 %). MSIS registrerte videre 304 tilfeller av MRSA-kolonisering i 2008. Det totale antallet MRSA-meldinger økte fra 594 meldinger i 2007 til 652 i 2008 (+ 9,8 %). Resultatene fra overvåkingen antyder en utflating i det totale antallet MRSA-infeksjoner i Norge, men at antallet koloniserte og andelen av alvorlige infeksjoner forårsaket av MRSA fortsatt øker. Blant *S. aureus* isolater fra sårprøver fortsatte nedgangen i andelen med fucidinresistens fra 14,5 % i 2006 og 11,1 % i 2007, til 10,3 % i 2008.

Blodkulturisolater av *E. coli* og *Klebsiella* spp. var som tidligere stort sett følsomme for bredspektrede antibiotika. Forekomsten av resistens og nedsatt følsomhet for gentamicin hos *E. coli* var 2,9 %. Dette er en svak reduksjon fra 3,9 % i 2007 og på samme nivå som 2,3 % i 2006. Det er observert en økning av resistens og nedsatt følsomhet for ciprofloxacin fra 3,3 % i 2004, 5,0 % i 2005, 5,7 % i 2006 og 7,1 % i 2007, til 8,1 % i 2008. Tallene er justert i henhold til nye brytningspunkter. Det er en klar samvariasjon mellom forbruket av fluorokinoloner og nedsatt følsomhet for denne antibiotikagruppen. *Klebsiella* spp. hadde lavere forekomst av resistens mot aminoglykosider og fluorokinoloner enn *E. coli*.

Produksjon av bredspektrede beta-laktamaser (ESBL) er blitt et utbredt problem i mange land, og enkelttilfeller er også blitt rapportert fra Norge. Til sammen 19/1279 *E. coli* (1,5 %) og 10/505 (2,0 %) av *Klebsiella* spp. fra blodkulturer ble rapportert som ESBL positive. For *E. coli* var forekomsten av ESBL litt høyere enn i 2007 (1,2 %). Forekomsten av ESBL blant *Klebsiella* spp. økte fra 1,0 % i 2007 til 2,0 % i 2008, men flere av isolatene kunne ikke verifiseres ved molekylære analyser. Det er derfor ingen sikker økning av ESBL-forekomsten blant *Klebsiella* spp. Andelen av ESBL positive isolater var fortsatt høyere blant *E. coli* fra blodkulturer (1,5 %) enn fra urinprøver (0,7 %). *Enterobacter cloacae* i blodkulturer fra 2006-2008 viste som ventet en høy forekomst av derepressert AmpC beta-laktamaseproduksjon, men isolatene var forøvrig følsomme for de fleste bredspektrede antibiotika.

Det ble ikke påvist klinisk signifikant vankomycin-resistens i enterokokker i 2008. Forekomsten av nedsatt følsomhet for ampicillin i *Enterococcus faecium* ligger

fortsatt rundt 80 %, og høygradig gentamicinresistens ble påvist i 33,4 % av *E. faecalis* og 53,6 % av *E. faecium*. De aller fleste (56 av 59) *E. faecium*-isolater med høygradig gentamicinresistens hadde samtidig nedsatt følsomhet for ampicillin. Alle enterokokkisolatene var følsomme for linezolid.

Streptococcus pneumoniae fra blodkulturer var generelt følsom for alle relevante antibiotika. Femten av 507 isolater (3,0 %) hadde nedsatt følsomhet for penicillin G, og syv av disse hadde også redusert følsomhet for cefalosporiner. Andelen av isolater med nedsatt følsomhet for penicillin G er på samnme nivå som i 2007 (3,3 %). Forekomsten av makrolidresistens blant pneumokokker i blodkultur ble for første gang siden registreringen startet redusert fra 12,4 % i 2006 til 9,9 % i 2007. Nedgangen fortsatte til 8,5 % i 2008 og må sees i sammenheng med innføringen av den konjugerte pneumokokkvaksinen i barnevaksinasjonsprogrammet i juli 2006.

Moraxella catarrhalis fra luftveisprøver viste svakt økende forekomst av beta-laktamaseproduksjon fra 90,3 % i 2003 til 92,0 % i 2008. Andelen av isolater med resistens mot erytromycin sank fra 8,3 % til 5,1 % i samme periode. Resistensforholdene for *Streptococcus pyogenes* (beta-hemolytiske streptokokker gruppe A) fra sår og luftveisprøver har stort sett vært uendrede i perioden 2002–2008.

I alt 324 tilfeller av tuberkulose ble meldt til MSIS i 2008. Det ble utført resistensbestemmelse av 225 *Mycobacterium tuberculosis* isolater fra pasienter som ikke hadde blitt behandlet for tuberkulose tidligere. Fire isolater fra pasienter smittet i henholdsvis Afrika (n=2), Asia (n=1) og Europa utenfor Norge (n=1) ble klassifisert som multiresistente. Det ble også gjort resistensbestemmelse av 27 isolater fra pasienter som tidligere var blitt behandlet for tuberkulose.

Det ble utført resistensbestemmelse av 199 blodkulturisolater av *Candida albicans* (n=140), *C. glabrata* (n=37) og *C. tropicals* (n=22). Alle *C. albicans* isolater var følsomme for amphotericin B, voriconazol, caspofungin og anidulafungin, men et enkelt isolat var resistent mot fluconazol. Det ble påvist økende forekomst av resistens mot fluconazol og voriconazol blant *C. glabrata* og *C. tropicalis*. Resultatene er i samsvar med tidligere studier fra Norge.

Overvåking av resistens mot antivirale midler omfattet i 2008 både influensavirus og HIV. For influensa A(H3N2) ble det i sesongen 2008/2009 påvist høy forekomst av resistens mot M2 blokkere, men full følsomhet for neuraminidasehemmere (for eks. oseltamivir). Det er likeledes ikke blitt påvist resistens mot neuraminidase-hemmere hos den nye varianten av influensa A(H1N1) som er tilkommet i 2009 (tidligere kalt svineinfluensa). Blant pasienter med nydiagnostisert HIV-infeksjon var det i perioden 2006-2008 8,4 % som hadde virus med noen grad av resistens mot antiretrovirale medikamenter. 2,7 % av isolatene ble angitt å ha høygradig eller intermediær resistens.

Konklusjon

Antibiotikaresistens er fortsatt et begrenset problem i Norge både når det gjelder mennesker og dyr. Det lave forbruket av antibiotika og det fordelaktige forbruks-mønsteret 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 ninth 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 2008. The NORM and NORM-VET programmes are part of the Norwegian Government's Action Plan against Antimicrobial Resistance issued in 2000. NORM is coordinated by the Department of Microbiology and Infection Control, 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 2008, the total sales of antimicrobial drugs approved for therapeutic use in animals in Norway were 6,219 kg (fish not included). The annual usage of veterinary antimicrobial drugs decreased gradually by approximately 40% from 1995 to 2001, and has thereafter remained relatively stable. The patterns of use have gradually become more favourable as the proportion of penicillin-use has increased. The proportion accounted for by pure penicillin preparations rose from 24% in 1995 to 46% in 2008. Altogether, 86% of the veterinary penicillin preparations sold in 2008 were beta-lactamase sensitive penicillins. The sales of sulfonamides decreased from 14% in 1995 to 0.2% in 2008. The proportion accounted for by tetracyclines varied between 3-5% in the period 1995-2008. The reduced antimicrobial drug 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 2008, the total sale of antimicrobial drugs for therapeutic use in farmed fish was 941 kg of active substance. Quinolones accounted for 72% of this amount. The usage of antimicrobials in Norwegian aquaculture declined by approximately 98% from 1987 to 1996 and has thereafter remained relatively constant. This reduction is mainly attributed to the introduction of effective vaccines in salmonids as well as to improved health management. In 2008, 65% of the prescribed amounts of antimicrobial agents used in aquaculture were for Atlantic cod, 23% for Atlantic salmon, 8% for Rainbow trout and 4 % were almost exclusively for halibut and turbot.

In 2008, the total sales of coccidiostatic feed additives, in kilograms of active substance, was twice the amounts used prior to the ban of antimicrobial growth promoters in 1995. This is explained by increased production of broilers. While monensin was the most frequently used ionophore in poultry in 1995, the usage of coccidiostats has since then been dominated by narasin.

Usage of antimicrobial agents in humans

In 2008, the overall sales of antibacterials for systemic use in humans represented 19.8 DDD/1,000 inhabitants/day. Total sales of antibacterials have remained relatively unchanged for many years although, within subgroups of antibacterials, usage trends have changed. Since 2004 an increase has been observed. Sales of penicillins and quinolones are increasing, while the subgroup of sulphonamides and trimethoprim is decreasing. The use in DDDs of methenamine, a urinary antiseptic agent, is huge and the use now represents 15% of total DDDs of antibacterials.

In 2008, 43% of the total antimicrobial sales were penicillins. Within this group, the subgroup of beta-lactamase sensitive penicillins is the most commonly used. In 2008, the subgroups of penicillins with extended specter and beta-lactamase resistant penicillins increased by 5% and 6%, respectively. Tetracyclines represented 16% of total use in 2008. The use of macrolides and lincosamides decreased by 7% in 2008 and represented 11% of total use. The sales of cephalosporins, monobactams and carbapenems represented 3% of the total sales of antibacterials. There has been a marked increase in quinolone use. Quinolones represented only 4% of total antibacterial sales in 2008, but since 2000 the sales have doubled.

The use of antibacterials varies according to age, gender and area of residence. Antibacterial sales to hospitals and ambulatory care represented 9% and 91% of the total human sales in 2008, respectively. Penicillins accounted for around 43% of the sales to hospital and for 44% in ambulatory care. The most important other groups in hospitals were the cephalosporins (20%), followed by quinolones (7%), and in ambulatory care the tetracyclines (18%) and the macrolides and lincosamides (12%) were the most important other groups.

Resistance in animal clinical isolates

The clinical isolates included in 2008 were from diagnostic samples of beta-haemolysin-producing *Staphylococcus* spp. from skin or ear infections in dogs and consisted primarily of *Staphylococcus pseudintermedius*. The prevalence of antimicrobial resistance in beta-haemolysin-producing staphylococci was high, and only 13.5% of the isolates were susceptible to all antimicrobial agents included. The proportion of multiresistant isolates, i.e. resistant to two or more antimicrobials, was 67%, and resistance to penicillin and fusidic acid were most commonly identified.

Resistance in indicator bacteria from animals and food

The prevalence of 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. are commonly included as indicator bacteria. In 2008, faecal samples from swine were examined for *E. coli* and *Enterococcus* spp, whereas faecal samples from dogs were examined for *E. coli*.

The occurrence of resistance in *E. coli* was low from dogs and moderate to low from swine. In total, 83.1% and 77.5% of the *E. coli* isolates from dogs and swine, respectively, were susceptible to all antimicrobial agents included. A higher percentage of the *E. coli* isolates from

dogs were found to be resistant to three or more antimicrobial agents, although, the difference is not statistically significant. One isolate from a dog was identified as an extended-spectrum beta-lactamase (ESBL) producer. Occurrence of ESBL producing bacteria in Norwegian animals should be regarded with concern.
Examination of faecal samples from swine with regard to *Enterococcus* spp. demonstrated presence of such bacteria in 45% of the samples, and the majority of these were *Enterococcus faecium*. The occurrence of resistance in *Enterococcus* spp. was - as in previous years - moderate, albeit, nearly one in two isolates are resistant to one or more antimicrobial agents. Resistance to tetracycline and erythromycin was the two most common resistance types identified in *Enterococcus* spp. from swine.

Resistance in zoonotic and non-zoonotic enteropathogenic bacteria

In 2008, a total of 15 *Salmonella* spp. isolates from Norwegian animals were susceptibility tested. Twelve of the isolates were *S*. Typhimurium and seven of these were from dogs, two from cattle, two from cats and one from horse. The remaining *Salmonella* spp. isolates were *S*. London, *S*. Goldcoast and *S*. Hvittingfoss. Antimicrobial resistance in *Salmonella* from animals in Norway is, with one exception in 2008, primarily associated with isolation of pentaresistant *S*. Typhimurium DT104, which was done on three occasions. One was a follow-up sample from cattle whereas the two remaining, in addition to one non-typable isolate with quinolone resistance, were isolated from dogs. The data, although very limited, indicate that antimicrobial resistance is not widespread among random *Salmonella* isolates from animals in Norway.
In 2008, 83.4% 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" (55.8%) than for the "infected abroad" category (32.6%). Multiresistant strains, i.e. resistant to two or more antimicrobial agents, were more common in the category "infected abroad" (53.0%) than in the category "infected in Norway" (36.0%). The data from 2001-2008 indicate that the prevalence of resistance to tetracyclines and ampicillin in *S*. Typhimurium may be increasing. The prevalence of resistance was considerably lower in *S*. Enteritidis isolates than in *S*. Typhimurium except for nalidixic acid. In total, 28.6% of *S*. Enteritidis isolates were resistant to nalidixic acid. However, a significant reduction in the prevalence of resistance to ciprofloxacin compared to the 2007 data was observed.
The results obtained in 2008 show that the prevalence of resistance in *Campylobacter jejuni* from Norwegian broilers is still low and stable. A total of 92.2% of the isolates were susceptible to all antimicrobial agents. As for *C. jejuni* isolated from humans infected in Norway, no erythromycin or gentamicin resistance were detected in isolates from Norwegian broilers in 2008. *C. jejuni* isolates from the group "infected in Norway" had somewhat higher rates of quinolone and tetracycline resistance than found in Norwegian broilers. Both these groups were, however, considerably less resistant than *C. jejuni* isolates derived from patients infected abroad, where only 27.0% of isolates were susceptible to all antimicrobial agents included. The fraction of multiresistant *C. jejuni* isolates derived from patients infected abroad was also much higher compared to humans infected in Norway. More than half of *C. jejuni* infections were acquired abroad, and 47.6% of these isolates were resistant to three or more antimicrobial agents.
Infections with *Yersinia enterocolitica* are typically obtained domestically, and the most common serotype is O:3. Resistance to nalidixic acid and trimethoprim-sulfamethoxazole are most frequently identified. In 2008, all isolates of *Shigella* were acquired out of the country and - as reported by other countries - antimicrobial resistance was commonly identified.

Resistance in human clinical isolates

The prevalence of resistance in human clinical isolates was still very low in Norway in 2008. Only six methicillin resistant *Staphylococcus aureus* (MRSA) blood culture isolates were detected among 871 strains included in the NORM protocol (0.7%), and 10 out of 1,359 (0.7%) *S. aureus* isolates were reported as MRSA from the laboratories´ information systems. The total number of systemic *S. aureus* isolates from blood cultures and cerebrospinal fluids was 1,365 including 10 MRSA strains (0.7%). This prevalence has increased slightly from 0.4% in 2007. The Norwegian Surveillance System for Communicable Diseases (MSIS) registered 348 cases of MRSA infections in 2008 which is similar to the 342 cases registered in 2007. A majority of the MRSA cases (88%) were reported to be wound infections and/or abscesses. Conversely, the prevalence of MRSA among non-invasive *S. aureus* isolates is still very low (7/1,061, 0.7%). Furthermore, MSIS registered 304 cases of MRSA colonization giving a total of 652 MRSA notifications in 2008. This is a 9.8% increase from the 594 notifications registered in 2007. The results may indicate a stable number of MRSA infections in Norway, but the prevalence of MRSA colonization and invasive disease may be increasing. The prevalence of resistance to fusidic acid among *S. aureus* wound isolates continued to decrease from 14.5% in 2006 and 11.1% in 2007, to 10.3% in 2008.
E. coli and *Klebsiella* spp. blood culture isolates were generally susceptible to broad-spectrum antimicrobials. The prevalence of gentamicin non-susceptibility in *E. coli* was 2.9% in 2008. This is a decrease from 3.9% in 2007 and on the same level as 2.3% in 2006. *E. coli* non-susceptibility to fluoroquinolones continued to increase from 3.3% in 2004, 5.0% in 2005, 5.7% in 2006 and 7.1% in 2007, to 8.1% in 2008. The figures have been adjusted for changes in microbiological breakpoints. There is a clear correlation between the total usage of fluoroquinolones and non-susceptibility to these agents. The prevalence of resistance to aminoglycosides and fluoroquinolones was lower in *Klebsiella* spp. isolates than in *E. coli*. Extended spectrum beta-lactamases (ESBL) have emerged as a significant clinical problem in many countries, and occasional cases have also been reported from Norway. A total of 19/1,279 (1.5%) *E. coli* and 10/505 (2.0%) *Klebsiella* spp. blood culture isolates were reported with this phenotype. For *E. coli*, this is a minor increase from 2007 (1.2%). The prevalence of ESBL production in *Klebsiella* spp. increased from 1.0% in 2007 to 2.0% in 2008, but several isolates could not be verified by molecular methods. Consequently, there is no documented increase in the prevalence of ESBL production in *Klebsiella* spp. The proportion of ESBL

positive isolates is still higher among *E. coli* from blood cultures (1.5%) than among urinary tract isolates (0.7%). *Enterobacter cloacae* blood culture isolates from 2006–2008 displayed, as expected, a high proportion of stably derepressed AmpC beta-lactamase production. The isolates were generally susceptible to all other broad-spectrum antimicrobial agents.

Clinically significant vancomcyin resistance was not detected in enterococci in 2008. The prevalence of non-susceptibility to ampicillin in *E. faecium* has stabilized around 80%, and high-level gentamicin resistance (HLGR) was detected in 33.4% of *E. faecalis* and 53.6% of *E. faecium*. Virtually all (56 out of 59) HLGR *E. faecium* isolates were also non-susceptible to ampicillin. All enterococcal isolates were suscpetible to linezolid.

Streptococcus pneumoniae from blood cultures were generally susceptible to all relevant antimicrobials. Fifteen out of 507 isolates (3.0%) displayed reduced susceptibility to penicillin G, and seven of these isolates were also non-susceptible to cefalosporins. This is on the same level as in 2007 (3.3%). The prevalence of macrolide resistance among pneumococcal blood culture isolates decreased for the first time from 12.4% in 2006 to 9.9% in 2007, and the decline continued with 8.5% in 2008. This reduction may be due to the conjucated pneumococcal vaccine which was introduced into the childhood vaccination programme in July 2006.

Moraxella catarrhalis from respiratory tract samples displayed a slight increase in the prevalence of beta-lactamase production from 90.3% in 2003 to 92.0% in 2008. The proportion of macrolide resistant isolates decreased from 8.3% to 5.1% in the same period. The resistance pattern for *Streptococcus pyogenes* (Group A beta-haemolytic streptococci) from wounds and respiratory tract specimens has remained unchanged from 2002-2008.

A total of 324 cases of tuberculosis were reported to MSIS in 2008. Susceptibility tests were performed on 225 *Mycobacterium tuberculosis* primary isolates. Only four isolates, originating from Africa (n=2), Asia (n=1) and Europe outside Norway (n=1) were classified as multidrug resistant (MDR). Susceptibility tests were also performed on *Mycobacterium tuberculosis* isolates from 27 previously treated patients.

Susceptibility testing was performed on 199 blood culture isolates of *Candida albicans* (n=140), *C. glabrata* (n=37) and *C. tropicals* (n=22). All *C. albicans* isolates were susceptible to amphotericin B, voriconazole and anidulafungin, but a single isolate was resistant to fluconazole. Increased prevalences of resistance were detected for fluconazole and voriconazole among *C. glabrata* and *C. tropicalis*. The results are in accordance with previous studies from Norway.

Surveillance data on resistance to antiviral agents included both influenza virus and HIV in 2008. A high prevalence of resistance to M2 blockers was noted for influenza A(H3N2) in the 2008/2009 season, but all isolates remained susceptible to neuraminidase inhibitors (e.g. oseltamivir). Similarly, all isolates of the recently discovered “new variant” influenza A(H1N1) (swine flue) have remained susceptible to neuraminidase inhibitors. Among patients newly diagnosed with HIV infections between 2006-2008, 8.4% of viral isolates displayed reduced susceptibility to antiretroviral drugs. 2.7% of isolates were classified as highly or intermediately resistant to these agents.

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 uses of antimicrobial agents in Norway increases or resistant clones are imported from abroad. A continued effort is needed to prevent the development and spread of antimicrobial resistance and thereby ensure the effectiveness of antimicrobials when such treatment is needed. The NORM/NORM-VET report is a vital component in the work aimed at preventing the development and spread of antimicrobial resistance in Norway.

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 1st, 2009.
Data provided by Statistics Norway.

Age group	All	Males	Females
0 to 4 years	298,460	152,844	145,616
5 to 14 years	613,756	314,370	299,386
15 to 24 years	613,950	314,355	299,595
25 to 44 years	1 344,100	686,989	657,111
45 to 64 years	1 224,174	622,495	601,679
65 years and older	704,812	304,000	400,812
All age groups	4 799,252	2 395,053	2 404,199

TABLE 2. Livestock population in Norway and the number of slaughtered animals in 2008.
Data provided by [1] Register of Production Subsidies as of 31 July, 2008 and [2] Register of Slaughtered Animals.

		Number of *	
Animal category	Herds	Animals	Slaughtered animals
Cattle	18,200[1]	1,011,700[1]	322,900[2]
Dairy cows only**	11,900[1]	232,400[1]	
Suckling cow only**	2,700[1]	38,200[1]	
Combined production (cow)**	1,200[1]	38,800[1]	
Goat	1,300[1]	69,600[1]	22,400[2]
Dairy goat**	450[1]	39,000[1]	
Sheep	15,100[1]	2,232,400[1]	1,140,600[2]
Breeding sheep > 1 year**	15,000[1]	891,400[1]	
Swine	2,700[1]	814,400[1]	1,497,200[2]
Breeding animal > 6 months**	1,600[1]	57,800[1]	
Fattening pigs for slaughter	2,400[1]	442,700[1]	
Poultry			
Egg laying hen (> 20 weeks of age)	1,800[1]	3,558,600[1]	988,200[2]
Flocks > 250 birds**	680[1]	3,535,800[1]	
Broiler	650[2]	-	62,234,900[2]
Turkey, ducks and geese for slaughter	100[1]	345,000[1]	1,388,600[2]
Flocks > 25 birds**	53[1]	344,600[1]	
Ostrich	5[1]	41[1]	

* Numbers >100 rounded to the nearest ten, numbers > 1000 rounded to the nearest hundred, ** Included in above total.

TABLE 3. Import of live animals and animal products (excluding fish) to Norway in 2008.
Data provided by the Norwegian Livestock Industry´s Biosecurity Unit (KOORIMP).

Species	Imported product	No. of consignments	No. of animals or products
Cattle	Live animals		7
	Semen (doses)		52,300
	Embryos		
Swine	Live animals		0
	Semen (doses)		779
Sheep	Live animals		0
	Embryos		
	Semen (doses)		
Goat	Live animals		46
	Semen (doses)		
Reindeer	Live animals for slaughter		
Fur animal	Live animals		
Poultry	Day-old chicks		106,958
	Fertilised eggs		3 304,032
Turkey	Day-old chicks	NA	NA
Duck and goose	Live birds	NA	NA

NA= Not available.

TABLE 4. Production volume of the most important species in Norwegian aquaculture during the time period 1992-2008. *Data provided by the Norwegian Directorate of Fisheries.*

Year	Atlantic salmon (ton)	Rainbow trout (ton)	Cod (ton)	Arctic char (ton)	Halibut (ton)	Blue mussels (ton)	Scallops[1] (ton)	Oysters (ton)
1992	141,000	-	-	-	-	-	-	-
1993	170,000	-	-	-	-	-	-	-
1994	204,686	14,571	569	262	63	542	-	-
1995	261,522	14,704	284	273	134	388	-	-
1996	297,557	22,966	191	221	138	184	-	-
1997	332,581	33,295	304	350	113	502	-	-
1998	361,879	48,431	203	200	291	309	-	-
1999	425,154	48,692	147	498	451	662	67	41
2000	440,061	48,778	169	129	548	851	38	8
2001	435,119	71,764	864	318	291	920	22	3
2002	462,495	83,560	1,258	319	424	2,557	5	2
2003	509,544	68,931	2,185	272	426	1,829	1	2
2004	563,915	63,401	3,165	365	648	3,747	46	3
2005	586,512	58,875	7,409	352	1,197	4,885	3	2
2006	629,888	62,702	11,087	897	1,185	3,714	4	1
2007	744,222	77,381	11,104	391	397	2,661	6	4
2008[2]	742,976	75,316	18,052	468	1,587	1,913	28	3

[1] From the wild population. [2] Preliminary figures.

V. USAGE OF ANTIMICROBIAL AGENTS

USAGE IN ANIMALS

Kari Grave

Therapeutic usage of veterinary antimicrobial agents

The data are based on sales from drug wholesalers to Norwegian pharmacies and from feed mills to fish farmers (see Appendix 1) of veterinary antimicrobial agents for therapeutic usage and includes pharmaceutical formulations approved for food animals, including horses, and/or dogs and cats. Thus, the figures represent national sales data for veterinary antimicrobial agents. Antimicrobial agents authorized for human use, but prescribed for animals, are not included (see Appendix 1 for inclusion criteria).

Table 5 summarizes the sales of veterinary antimicrobial agents for therapeutic use in domestic animals in Norway in 2008. The data are organized according to therapeutic substance groups (ATCvet groups) and show the total usage for the various routes of administration. The total annual sale of veterinary antimicrobial agents for terrestrial animals for the period 1995-2008 is given in Figure 1. Figure 2 illustrates the proportion of the total sale of the various groups of antimicrobial agents. In 2008, the sales of veterinary antimicrobial agents approved for therapeutic use in animals in Norway amounted to 6,219 kg of active substance (Table 5). The annual usage of veterinary antimicrobial agents decreased gradually by 40% from 1995 to 2008, from 2001-2006 this usage varied slightly but a 9 % increase was observed for this period. During the years 2007 and 2008 a 4% decrease in the usage was seen (Figure 1).

TABLE 5. Sales in 2008 calculated as kilograms of active substance, of veterinary antimicrobial agents approved in Norway for therapeutic use in animals (farmed fish not included, see Table 6). Numbers of sold items in 2008 for the various veterinary drug formulations, strengths and package sizes were obtained from the Norwegian Institute of Public Health and represent sales from drug wholesalers to the Norwegian pharmacies.

Groups of substances	ATCvet code	Active substance or combinations of substances	Gastro-intestinal (QA07)	Uterine (QG01)	Systemic indiv. (QJ01)	Intra-mammary (QJ51	Herds (QJ01)
Tetracyclines	QG01AA07/QJ01AA06	Oxytetracycline		3	113		137
	QJ01AA02	Doxycycline			0.6		
Amphenicols	QJ01BA99	Florfenicol+flunixin[1]			19		
Beta-lactams	QJ01CA04	Amoxicillin			131		180
	QJ01CE09/QJ51CE09	Procaine penicillin[2]			2,121	32	
	QJ01CE90	Penethamate hydroiodide[2]			1		
	QJ01CR02/QJ51RV01	Amoxicillin+clavulanic acid			357	9	
Cephalosporins	QJ01DD91	Cefovecin			1.1		
Sulfonamides	QJ01EQ06	Sulfanilamid[3]			13		
Sulfonamides and	QJ01EW10	Sulfadiazine+trimethoprim[5]			1,227		285
Trimethoprim	QJ01EW13	Sulfadoxine+trimethoprim			104		
Lincosamides	QJ01FF01	Clindamycin			20		
Aminoglycosides	QA07AA01	Neomycin	33				
	QA07AA90	Dihydrostreptomycin (DHS)	123				
	QJ01GB03	Gentamicin[5]			10		
Quinolones	QJ01MA90	Enrofloxacin[4]			30		0.3
	QJ01MA96	Ibafloxacin			1.4		
Others	QJ01XX92	Tiamulin[4]			9		120
Combinations	QJ01RA01/QJ51RC23	Procaine penicillin[1] +DHS			449	507	
	QJ51RC24	Benzylpenicillinbenzatine[2]+ DHS[6]				15	
	QJ51RC25	Penethamate hydroiodide[4] + DHS				1.1	
	QG01AE99	Sulfadimidine+procaine penicillin[2]+DHS		166			
Total per route of administration			**156**	**169**	**4,608**	**563**	**722**
Total (kg)							**6,219**

[1] Flunixin not included in figure; [2] Calculated as benzylpenicillin; [3] Represents extemporaneously prepared preparations; [4] Includes also a preparation used on exemption from market authorization; [5] Includes a premix approved for farmed fish that are used solely in terrestrial animals such as pigs and calves (Kari Grave, unpublished data); [6] Represents two preparations used on exemption from market authorization.

The proportion accounted for by pure penicillin preparations rose from 24% in 1995 to 46% in 2008. Altogether 86% of the pure penicillin preparations sold in 2008 were beta-lactamase sensitive penicillins. From 1995 to 2007, the sale of sulfonamides in combination with trimethoprim (or baquiloprim 1995-2000) increased from 11% to 26% of the total sales. The proportion of sale of the combination preparations of penicillins and aminoglycosides decreased from 34% to 16% from 1995 to 2008. The corresponding figures for the sulfonamides were 14% in 1995 and 0.2% in 2008. The proportion accounted for by tetracyclines varied between 3-5% in 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.

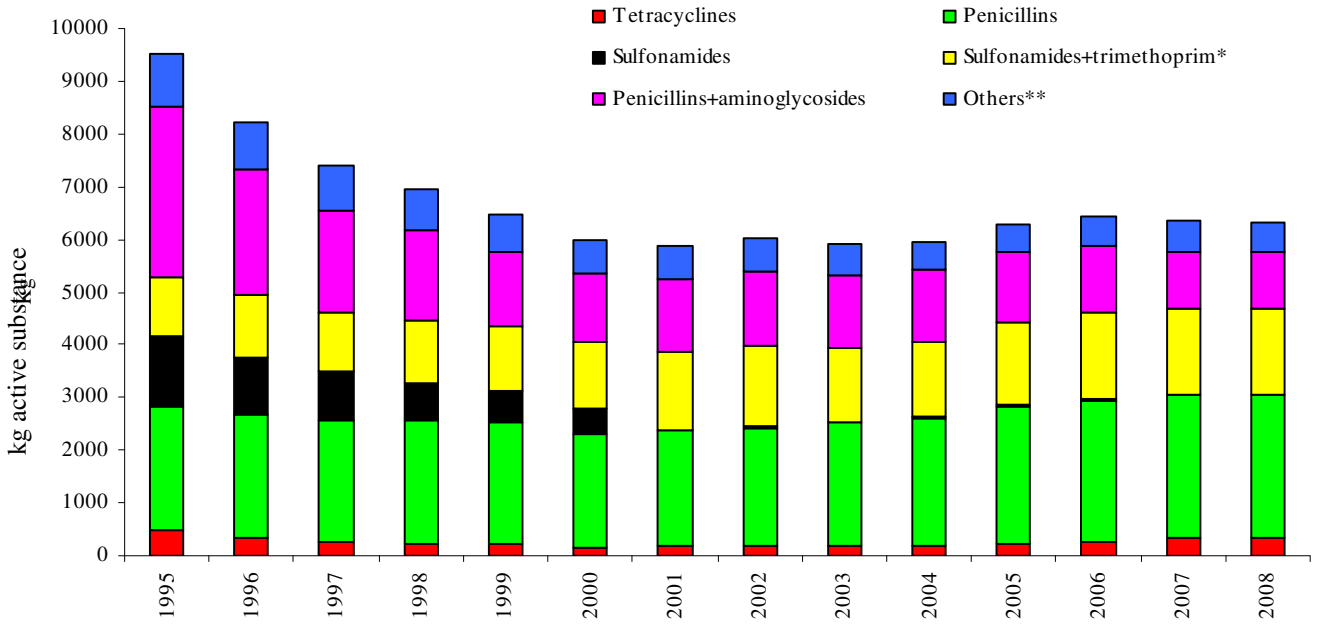

FIGURE 1. Sales (in kilograms of active substance) of veterinary antimicrobial agents (QA07AA, QG01AA, QG01AE, QJ01, QJ51) for therapeutic use in Norway 1995–2008, fish not included. Numbers of sold items in 2008 for the various veterinary drug formulations, strengths and package sizes were obtained from the Norwegian Institute of Public Health and represent sales from drug wholesalers to Norwegian pharmacies. Sulfonamides were not sold during 2001-2003.
*Includes small amounts of baquiloprim (1995-2000); includes a premix approved for farmed fish used solely in terrestrial animals such as pigs, horses and calves (Kari Grave, unpublished data). **Includes ATCvet codes: QAA7AA01; QA07AA90; QG01AE99; QJ01BA99; QJ01FA01; QJ01FF01; QJ01FF02; QJ01GB06; QJO1MA90; QJO1MA96; QJ01XX92; QJ51RC26.

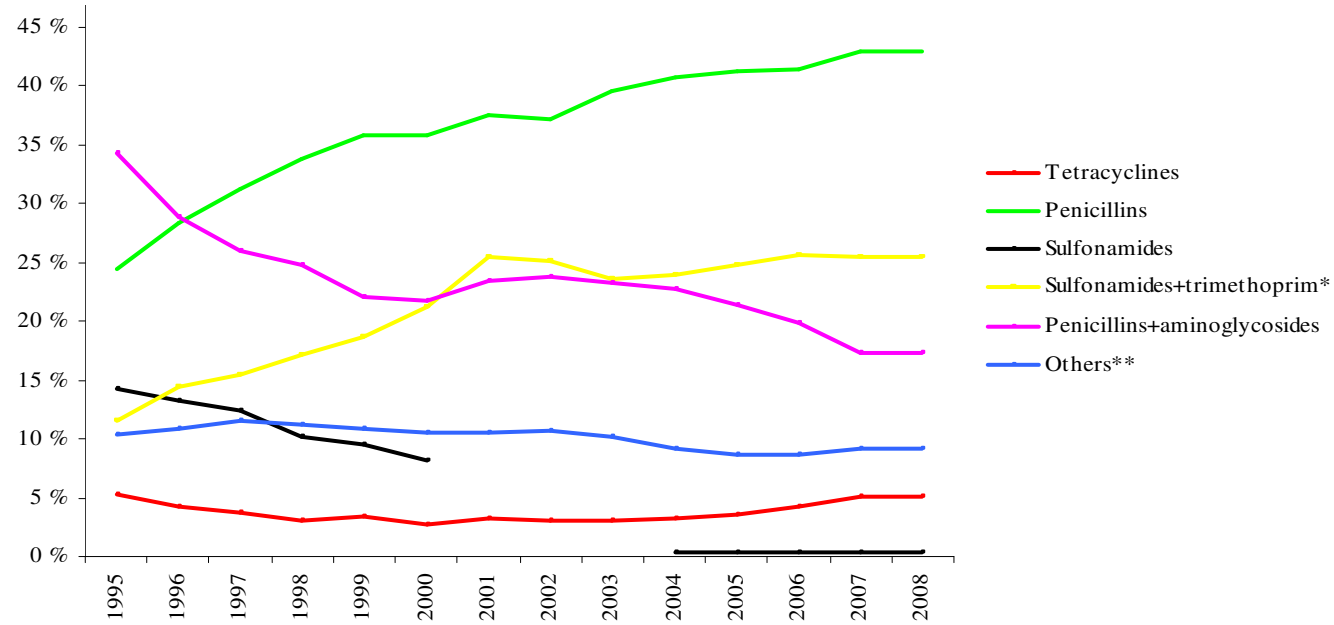

FIGURE 2. Sales (as percentage of total sales) of veterinary antimicrobial agents (QA07AA, QG01AA, QG01AE, QJ01, QJ51) in Norway 1995-2008, fish not included. Number of sold items in 2008 for the various veterinary drug formulations, strengths and package sizes were obtained from the Norwegian Institute of Public Health and represent sales from drug wholesalers to Norwegian pharmacies. Sulfonamides were not sold during 2001-2003.
*Includes small amounts of baquiloprim (1995-2000); includes a premix approved for farmed fish used solely in terrestrial animals such as pigs, horses and calves (Kari Grave, unpublished data). **Includes ATCvet codes: QAA7AA01; QA07AA90; QG01AE99; QJ01BA99; QJ01FA01; QJ01FF01; QJ01FF02; QJ01GB06; QJO1MA90; QJO1MA96; QJ01XX92; QJ51RC26.

TABLE 6. Total sales (in kilograms of active substance) of veterinary antimicrobial agents for therapeutic use in farmed fish in Norway in the period 1995-2008. 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 to fish farms.

Groups of substances/active substance	ATCvet code	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008
Tetracyclines															
Oxytetracycline	QJ01AA06	70	27	42	55	25	15	12	11	45	9	8	0	0	0
Amphenicols															
Florfenicol	QJ01BA90	64	64	123	135	65	148	109	205	154	111	202	302	139	166
Quinolones															
Flumequine	QJ01MB07	182	105	74	53	7	52	7	5	60	4	28	7	18	1
Oxolinic acid	QJ01MB91	2,800	841	507	436	494	470	517	998	546	1,035	977	1,119	406	681
Combinations															
Spectinomycin · lincomycin (2+1)	QJ01RA												50	66	70
Total		**3,116**	**1,037**	**746**	**679**	**591**	**685**	**645**	**1,219**	**805**	**1,159**	**1,215**	**1,428**	**648**	**941**

In 2008, the sales of veterinary antimicrobial agents for use in farmed fish were 941 kg active substance, of which 72% were quinolones (Table 6). The annual usage of antimicrobial agents in Norwegian fish farming peaked in 1987 when the reported sales figures amounted to approximately 48 tonnes. This implies that the usage of antimicrobial agents in Norwegian aquaculture declined by approximately 98% from 1987 to 1996. From 1987 the total production of farmed fish increased more than sixty times. This significant decrease in the usage of antimicrobial agents in Norwegian aquaculture in the period 1987 to 1996 was mainly attributed to the introduction of effective vaccines against bacterial diseases in Atlantic salmon and rainbow trout and to some extent also to improved health management.

In 2008, 65% of the prescribed amounts of antimicrobial agents in aquaculture were for Atlantic cod, 23% for Atlantic salmon and 8% for rainbow trout and 4% for other species (Figure 3).

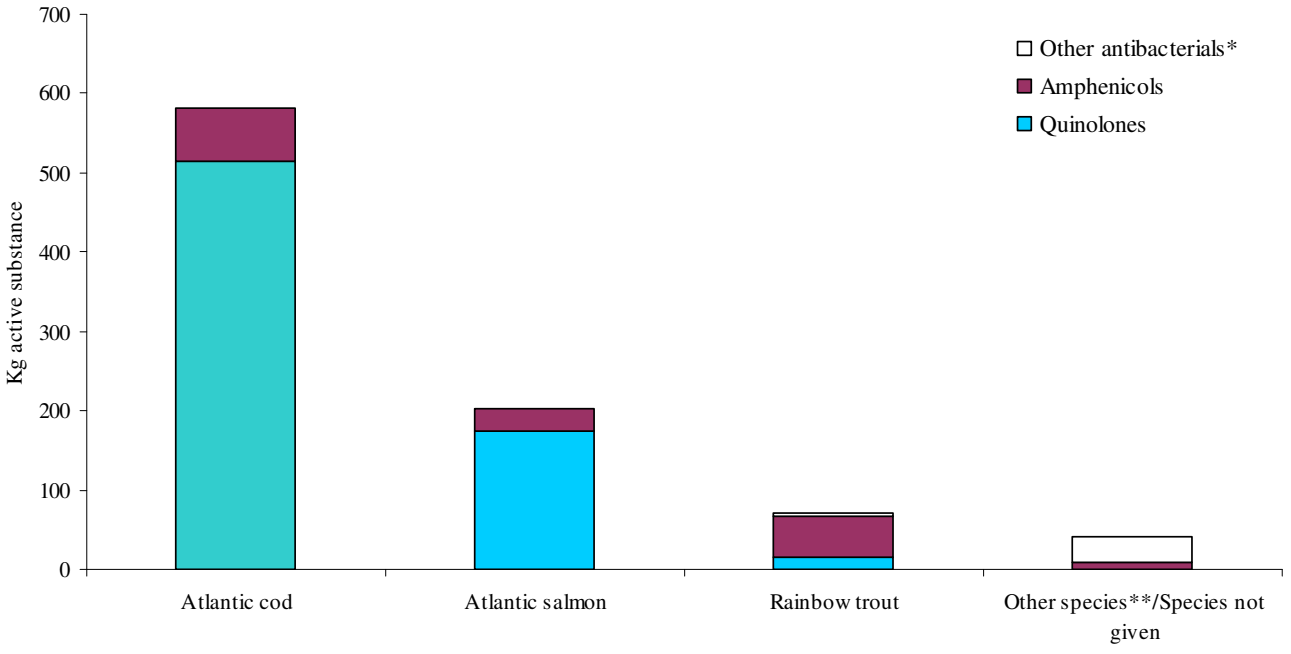

FIGURE 3. Prescribed amounts (in kilograms of active substance) of veterinary antimicrobial agents in Norwegian aquaculture in 2008 split into various fish species. Prescription data were obtained from the Norwegian Food Safety Authority (Trygve Helle, data on file). *Includes: Oxytetracycline (5.25 kg) and spectinomycin+lincomycin (33 kg for halibut fry).
** Other species: halibut (39.6 kg), turbot (0.8 kg) and species not given (0.8 kg).

Antimicrobial and coccidiostatic feed additives

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 (2003-2008). Table 7 summarizes total sales of antimicrobial growth promoters and coccidiostat feed additives in Norway in the period 1995–2008.

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. 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 1997.

Coccidiostats as feed additives are still used in Norwegian poultry production. The total sales of coccidiostats (kilograms of active substance) have been close to doubled since the ban on antimicrobial growth promoters, but the usage is highly correlated to the number of slaughtered chicken produced in this period. However, the pattern of usage has changed (Table 7). While monensin was the most frequently used ionophore in the poultry industry in 1995, the usage of coccidiostats has since then been almost totally dominated by narasin.

TABLE 7. Total sales, in kilograms of active substance, of antimicrobial growth promoters and of coccidiostats as feed additives in Norway 1995-2008. Data were obtained through annual reports from the Norwegian Agricultural Inspection Service (1995-2002) and the Norwegian Food Safety Authority (2003-2008).

Active substance	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008
Avoparcin[1]	419	P	P	P	P	P	P	P	P	P	P	P	P	P
Zincbacitracin	129	64	27	0	0	0	0	0	0	0	0	P	P	P
Virginiamycin[2]	0	0	0	0	P	P	P	P	P	P	P	P	P	P
Total antimicrobial growth promoters	**548**	**64**	**27**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
Lasalocid	996	480	471	193	208	80	96	514	108	173	37	13	17	16
Monensin	3,422	891	561	485	557	776	629	521	717	817	852	889	919	897
Salinomycin	214	27	0	0	27	233	12	0	0	0	0	0	0	0
Narasin	24	3,508	3,343	3,530	4,062	4,486	4,195	4,470	5,067	5,270	5,318	5,615	7,065	9,212
Total ionophore coccidiostats	**4,656**	**4,906**	**4,375**	**4,208**	**4,854**	**5,575**	**4,932**	**5,505**	**5,892**	**6,260**	**6,207**	**6,517**	**8,001**	**10,125**
Amprolium/etopabat	156	116	582	174	201	135	159	74	42	0.8	0	0	0	0
Total other	**156**	**116**	**582**	**174**	**201**	**135**	**159**	**74**	**42**	**0.8**	**0**	**0**	**0**	**0**

[1]Prohibited since May 31st, 1995. [2]Prohibited since 1999.

USAGE IN HUMANS

Hege Salvesen Blix

In 2008, the overall sale of antimicrobial agents for systemic use in humans was 19.8 DDD/1,000 inhabitants/day. Total sales of antibacterials have remained relatively unchanged for many years. However, since 2004 an increase has been observed. This change is mainly due to the penicillin group and to increased use of methenamine. The macrolides have been steadily increasing over many years, however in 2008 a decrease was observed. The use of quinolones is still increasing (Table 8, Figure 4).

TABLE 8. Human usage of antimicrobial agents in Norway 2001-2008 by ATC groups. The usage is presented as DDD (Defined Daily Doses)/1,000 inhabitants/day and in % change 2007-2008. Collection of data on human usage of antimicrobial agents is presented in Appendix 2.

ATC	Groups of substances	2001	2002	2003	2004	2005	2006	2007	2008	Change (%) 2007-2008
J01A	Tetracyclines	3.11	3.13	3.03	2.97	3.11	3.24	3.32	3.22	- 3
J01B	Amphenicols	0.003	0.002	0.002	0.001	0.001	0.002	0.001	0.001	-
J01CA	Penicillins with extended spectrum	2.1	2.23	2.29	2.37	2.53	2.74	2.93	3.09	+ 5
J01CE	Beta-lactamase sensitive penicillins	4.68	4.48	4.38	4.23	4.55	4.63	4.70	4.71	-
J01CF	Beta-lactamase resistant penicillins	0.41	0.50	0.59	0.63	0.56	0.66	0.72	0.77	+ 6
J01CR	Combination of penicillins	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	-
J01D	Cephalosporins, monobactams, carbapenems	0.55	0.58	0.62	0.61	0.57	0.60	0.60	0.60	-
J01E	Sulfonamides and trimethoprim	1.16	1.15	1.08	1.09	1.06	1.04	1.02	0.98	- 4
J01F	Macrolides, lincosamides and streptogramins	1.8	1.98	1.92	1.89	2.12	2.24	2.30	2.13	- 7
J01G	Aminoglycosides	0.06	0.06	0.07	0.06	0.07	0.07	0.07	0.07	-
J01M	Quinolones	0.40	0.44	0.48	0.52	0.57	0.62	0.67	0.70	+ 4
J01X	Other antibacterials	2.55	2.57	2.63	2.83	3.05	3.18	3.30	3.48	+ 5
	Total exclusive of methenamine	**14.7**	**15.0**	**14.9**	**14.8**	**15.6**	**16.3**	**16.9**	**16.8**	**- 1**
	Total all antimicrobial agents	**16.8**	**17.1**	**17.1**	**17.2**	**18.2**	**19.0**	**19.7**	**19.8**	**+ 1**

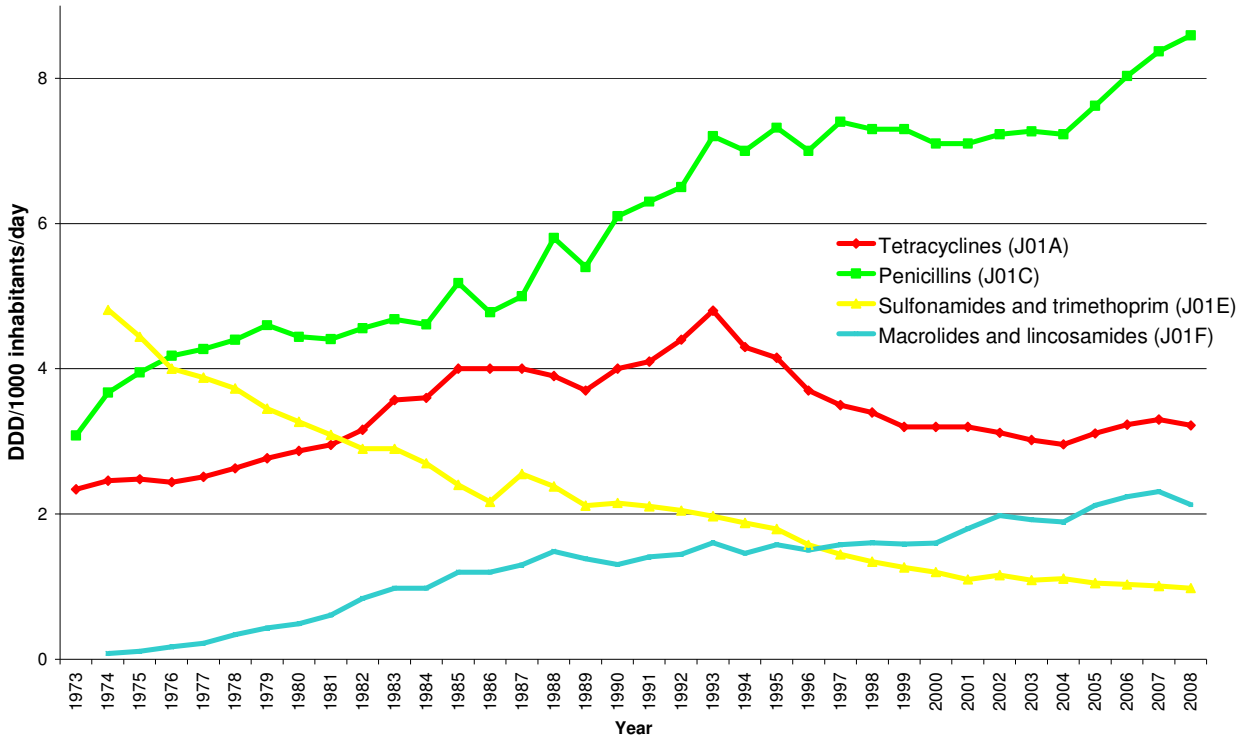

FIGURE 4. Sales of penicillins (J01C), tetracyclines (J01A), macrolides, lincosamides and streptogramines (J01F) and sulfonamides and trimethoprim (J01E) in Norway 1973-2008.

In 2008, the penicillins (ATC group J01C) represented 43% of the total antimicrobial drug use in human medicine in Norway (Figure 5). Among the penicillins the beta-lactamase sensitive penicillins (J01CE) accounted for the major proportion. It has been so over years, however, there has gradually been a shift towards use of more broadspectered penicillins. Penicillins with extended specter (J01CA) now represent 35% of the penicillin group compared to 26% a decade ago (Figure 6). This is mainly due to increasing use of pivmecillinam, which has become a predominant choice for urinary tract infections at the expense of the subgroup of sulfonamides and trimethoprim, which has decreased over the years. Use of beta-lactamase sensitive penicillins (J01CE) decreased during 2002-2004, but this use is now at the same level as in the beginning of this decade. The tetracyclines (J01A) represent 16% of total use and the sales have been relatively stable over time. The macrolides and lincosamides (J01F) represented 11% of total use in 2008. Since 2000 this use has steadily increased, however, in 2008, an 8% decrease was observed. The usage pattern within group J01F has remained relatively unchanged over the years. Erythromycin is the most frequently used agent in this group, representing 51% of sales of the subgroup (Figure 7). In the last years, sales of cephalosporins, monobactams and carbapenems have been stable, representing 3% of the total sales of antimicrobial agents. The usage pattern within this group has changed since 1996 (Figure 8). First generation cephalosporins i.e. cefalexin and cefalotin, represent 52% of sales of ATC group J01D. The use of quinolones has doubled since 2000. Still, it represents only a minor fraction (4%) of total sales of antimicrobial agents for humans. The increased sale of ATC group J01X agents is mainly due to the urinary prophylactic agent methenamine, representing 15% of total use of antimicrobial agents. The sale of methenamine has increased by 55% since 2000.

The usage of antimicrobial agents varies between the 19 Norwegian counties. The county with lowest use is using 75% (in DDDs) of the county with the highest use. There is a trend that the high-use and low-use counties are the same over the years (Figure 9).

The use of antimicrobial agents outside hospitals, including nursing homes, represents 91% of the total sale of antimicrobial drugs for humans. Antimicrobial agents are prescription-only drugs in Norway. Physicians are the main prescribers to humans, but dentists prescribe 5 % (measured in DDDs) of antimicrobial agents (J01) to humans in ambulatory care in Norway. The most frequently prescribed groups in ambulatory care are penicillins J01C (44% of DDDs), tetracyclins J01A (18%) and macrolides and lincosamides J01F (12%). Females use more antibiotics than males; 29% of females purchased at least one course of antimicrobial drug in 2008 compared to 21% of males. This pattern is observed in all regions of the country (Figure 10). The highest use is found among young children, young adults and the elderly (Figure 11).

The sales of antimicrobial drugs in DDDs to hospitals in 2008 represented 9% of total sale in the country. The patterns of antimicrobial drug use in hospitals have not change much 2006-2008 (Figure 12). Penicillins (J01C) represent around 43% of the use in hospitals followed by cephalosporins, monobactams and carbapenems (J01D) (20%) and the quinolones J01M (7%). The increase in the use of ATC group J01X in 2008 is due to methenamine.

Due to the amount of antimicrobial agents used, therapy traditions in ambulatory care have a great impact on the total burden of antimicrobials and thus on the development of bacterial resistance.

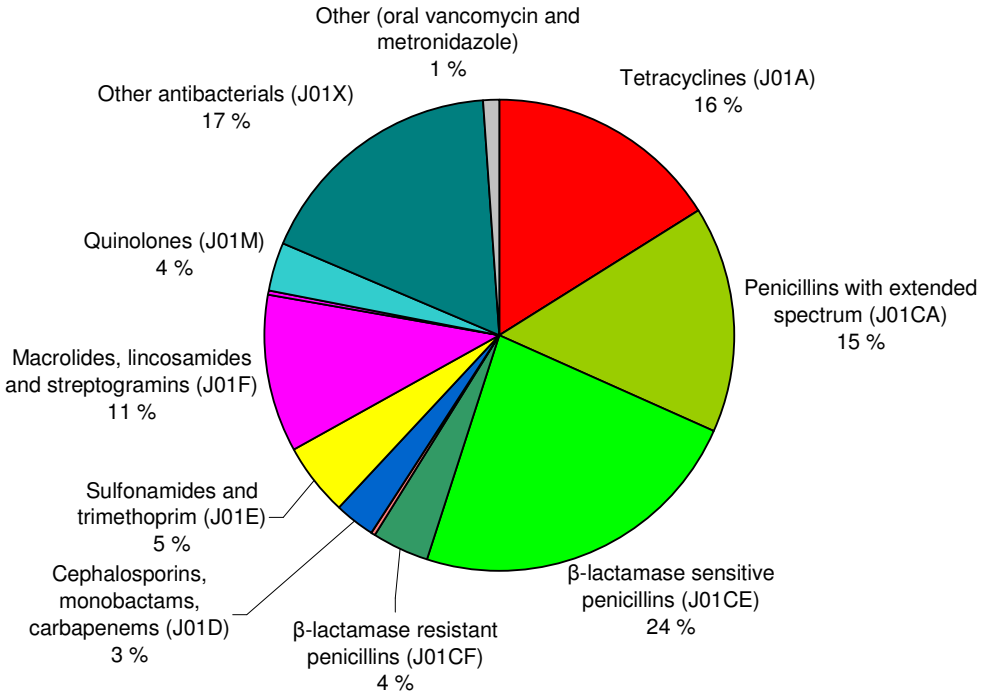

FIGURE 5. Proportional use of various groups of antimicrobial agents for systemic use in Norway 2008 in Defined Daily Doses (DDD) (total sale in the country).

TABLE 9. Human usage of single antimicrobial agents for systemic use in Norway. Sales are given in DDD/1,000 inhabitants/ day. The methodology for collection of data on human usage of antibacterial agents is presented in Appendix 2.

ATC	Substance	2000	2001	2002	2003	2004	2005	2006	2007	2008
A07A A09	Vancomycin	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
J01A A02	Doxycycline	2.10	2.1	2.03	1.93	1.80	1.89	1.97	2.0	1.9
J01A A04	Lymecycline	0.14	0.19	0.26	0.30	0.34	0.39	0.45	0.51	0.52
J01A A06	Oxytetracycline	0.24	0.22	0.21	0.19	0.20	0.20	0.19	0.18	0.17
J01A A07	Tetracycline	0.69	0.64	0.62	0.60	0.62	0.64	0.63	0.63	0.62
J01AA07*	Minocycline						0.0003	0.0003	0.0001	0.0002
J01AA12	Tigecycline							0.0001	0.0002	0.0004
J01B A01	Chloramphenicol	0.004	0.003	0.002	0.002	0.001	0.002	0.002	0.001	0.001
J01C A01	Ampicillin	0.09	0.08	0.09	0.1	0.1	0.1	0.1	0.1	0.1
J01C A02	Pivampicillin	0.13	0.11	0.11	0.09	0.08	0.07	0.06	0.01	0
J01C A04	Amoxicillin	0.83	0.89	0.94	0.95	0.94	1.06	1.11	1.26	1.34
J01C A08	Pivmecillinam	0.96	1	1.09	1.14	1.25	1.29	1.46	1.55	1.65
J01C A11	Mecillinam	0.004	0.005	0.005	0.005	0.005	0.006	0.006	0.006	0.008
J01C E01	Benzylpenicillin	0.21	0.23	0.24	0.25	0.24	0.26	0.26	0.25	0.24
J01C E02	Phenoxymethylpenicillin	4.45	4.45	4.24	4.13	3.99	4.29	4.37	4.45	4.46
J01C E08*	Benzathine benzylpenicillin	0.0001	<0.0001	0.0001	0.0001	0.0002	0.0001	0.0002	0.0001	0.0001
J01C F01	Dicloxacillin	0.25	0.31	0.39	0.48	0.51	0.41	0.54	0.61	0.64
J01C F02	Cloxacillin	0.10	0.09	0.11	0.11	0.11	0.15	0.12	0.12	0.13
J01C F05*	Flucloxacillin			0.0001	0.0002	0.0002	0.0001	0.0001	0.0003	0.0005
J01C R02*	Amoxicillin and enzyme inhibitor	0.01	0.01	0.01	0.01	0.0003	0.0000	0.0001	0.0001	0.0002
J01C R05	Piperacillin and enzyme inhibitor	0.0001	0.0006	0.0014	0.0024	0.005	0.01	0.01	0.02	0.02
J01D B01	Cefalexin	0.26	0.27	0.29	0.3	0.29	0.24	0.26	0.25	0.23
J01D B03	Cefalotin	0.05	0.05	0.05	0.06	0.06	0.06	0.06	0.07	0.07
J01D B04*	Cefazolin						0.002	0.002	0.001	0.001
J01D C01	Cefoxitin	0.0004	0.0003	0.0002	0.0001					
J01D C02	Cefuroxim	0.13	0.14	0.15	0.15	0.14	0.13	0.12	0.12	0.11
J01D D01	Cefotaxim	0.04	0.05	0.05	0.07	0.07	0.08	0.09	0.09	0.1
J01D D02	Ceftazidim	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
J01D D04	Ceftriaxone	0.011	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.02
J01D F01	Aztreonam	0.001	0.001	0.001	0.001	0.001	0.0005	0.0008	0.0008	0.0007
J01D H02	Meropenem	0.012	0.014	0.017	0.02	0.02	0.026	0.031	0.035	0.037
J01D H03	Ertapenem							0.000	0.001	0.001
J01D H51	Imipenem and enzyme inhibitor	0.006	0.005	0.005	0.006	0.005	0.005	0.004	0.004	0.003
J01E A01	Trimethoprim	0.79	0.8	0.8	0.74	0.76	0.73	0.70	0.68	0.64
J01E E01	Sulfamethoxazol and trimethoprim	0.38	0.36	0.36	0.34	0.34	0.33	0.34	0.34	0.34
J01F A01	Erythromycin	1.00	1.13	1.2	1.09	1.03	1.16	1.24	1.21	1.08
J01F A02	Spiramycin	0.02	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01
J01F A09	Clarithromycin	0.26	0.3	0.36	0.37	0.37	0.39	0.40	0.43	0.37
J01F A10	Azithromycin	0.19	0.21	0.24	0.26	0.28	0.32	0.34	0.39	0.38
J01FA15	Telithromycin			0.0001	0.0003	0.0003				
J01F F01	Clindamycin	0.12	0.14	0.16	0.19	0.20	0.23	0.25	0.26	0.28
J01GA01*	Streptomycin			0.0015	0.0004	0.0004	0.0002	0.0003	0.0002	0.0003
J01G B01	Tobramycin	0.02	0.03	0.04	0.04	0.03	0.03	0.03	0.03	0.03

ATC	Substance	2000	2001	2002	2003	2004	2005	2006	2007	2008
J01G B03	Gentamicin	0.006	0.008	0.02	0.03	0.03	0.03	0.04	0.04	0.04
J01G B06*	Amikacin			0.0009	0.0008	0.0003	0.0004	0.0009	0.0003	0.0007
J01G B07	Netilmicin	0.02	0.02	0.007				0.0001		
J01M A01	Ofloxacin	0.05	0.05	0.05	0.05	0.05	0.05	0.04	0.04	0.04
J01M A02	Ciprofloxacin	0.29	0.34	0.38	0.42	0.47	0.52	0.57	0.62	0.66
J01MA12*	Levofloxacin			0.001	0.0003		0.0003	0.0003	0.0008	0.0008
J01MA14	Moxifloxacin								0.0007	0.001
J01X A01	Vancomycin	0.005	0.005	0.006	0.006	0.007	0.007	0.008	0.01	0.01
J01X A02	Teicoplanin	0.0012	0.0013	0.0013	0.0009	0.0007	0.0008	0.0008	0.0007	0.001
J01X B01*	Colistin	0.003	0.003	0.003	0.002	0.003	0.004	0.005	0.004	0.004
J01X C01	Fusidic acid	0.003	0.01	0.01	0.007	0.008	0.006	0.006	0.006	0.006
J01X D01	Metronidazole	0.06	0.07	0.07	0.07	0.08	0.08	0.07	0.07	0.07
J01X E01	Nitrofurantoin	0.37	0.36	0.35	0.35	0.36	0.36	0.37	0.36	0.36
J01X X05	Methenamin	1.95	2.08	2.13	2.18	2.37	2.59	2.71	2.84	3.02
J01XX08	Linezolid			0.002	0.004	0.006	0.007	0.006	0.006	0.007
J01XX09	Daptomycin								0.000	0.000
D06AX09/ R01AX06*	Mupirocin in kg ointment/cream (2%)	0.3	1.0	1.3	3.0	3.0	3.4	4.3	4.0	3.9
P01AB01	Metronidazole	0.18	0.18	0.19	0.19	0.20	0.20	0.20	0.21	0.21
J04AB**	Rifampicin	0.046	0.054	0.043	0.049	0.068	0.077	0.082	0.092	0.092

* Drugs not licensed for the Norwegian marked but prescribed on exemption from marketing authorization.
** Given as the amount of rifampicin in plain and combination products.

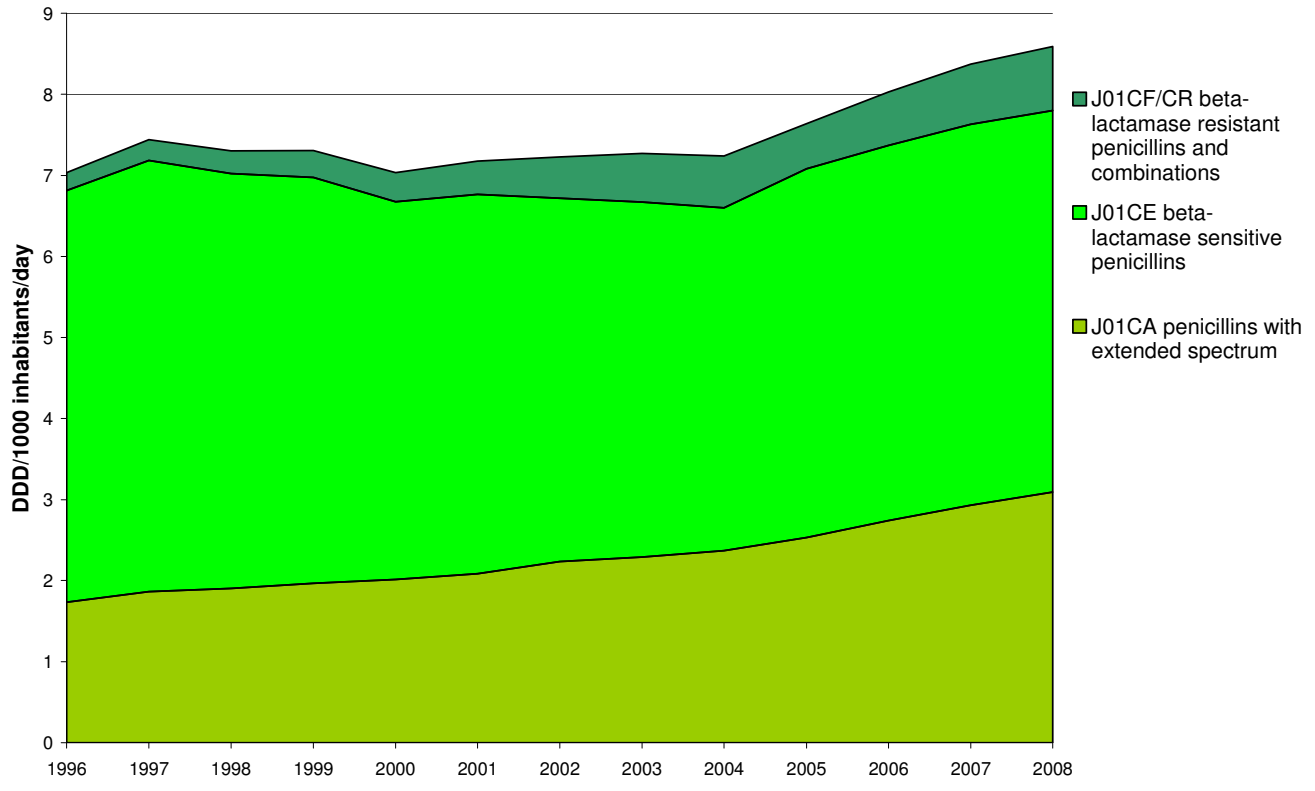

FIGURE 6. Total annual sales of penicillins (J01C) in Norway 1996-2008 and changes within the penicillin group.

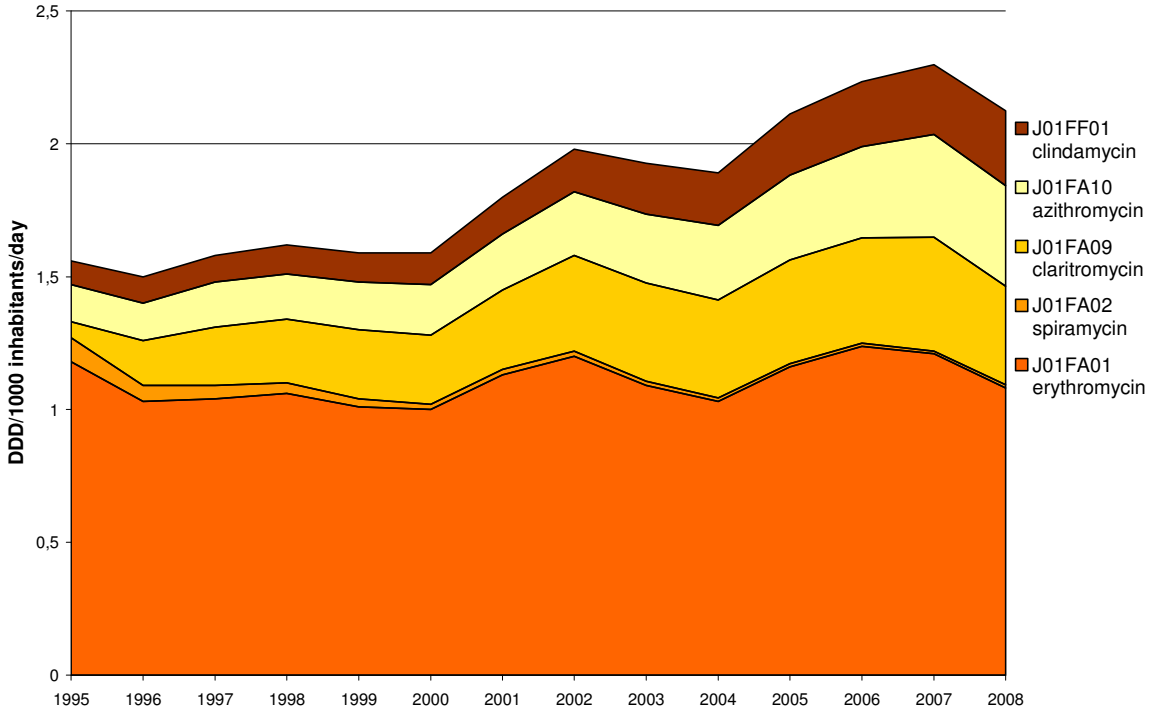

FIGURE 7. Sales of macrolides, lincosamides and streptogramins (J01F) in Norway 1996-2008.

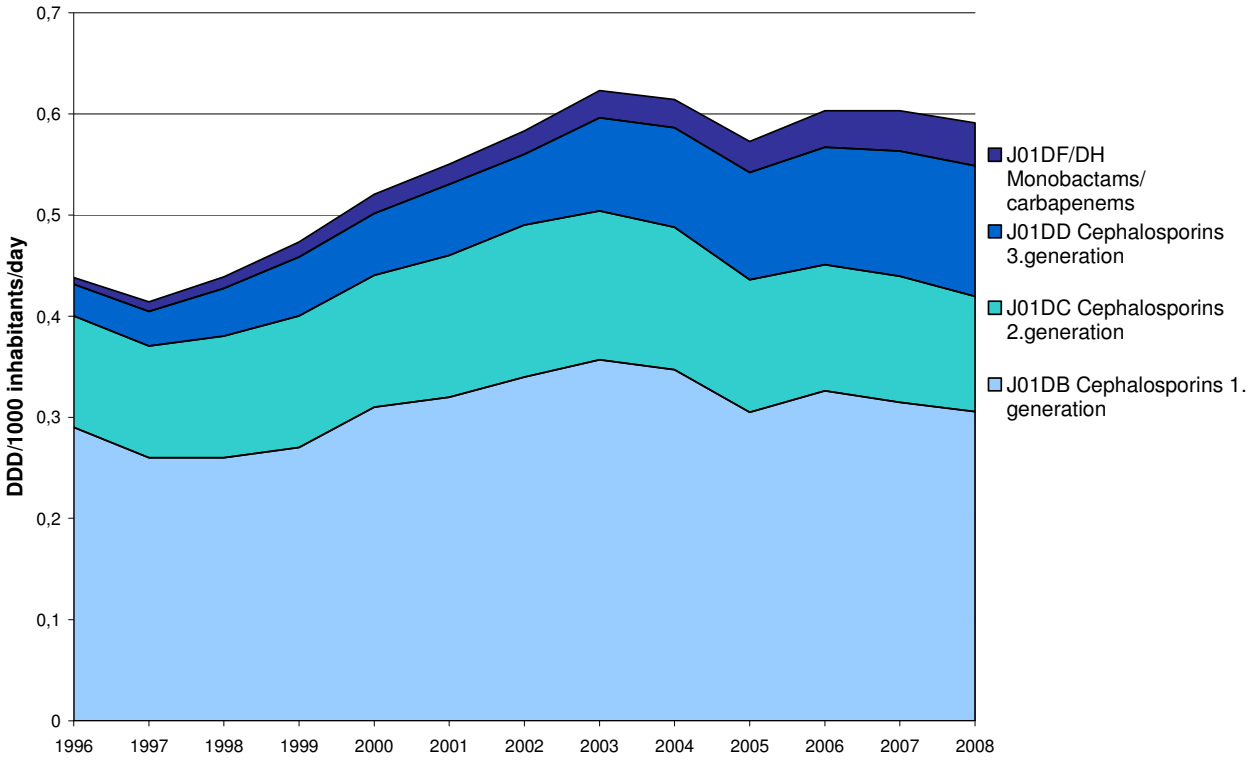

FIGURE 8. Sales of cephalosporins, monobactams and carbapenems (J01D) in Norway 1996-2008 and changes between generations of cephalosporins and monobactams/carbapenems.

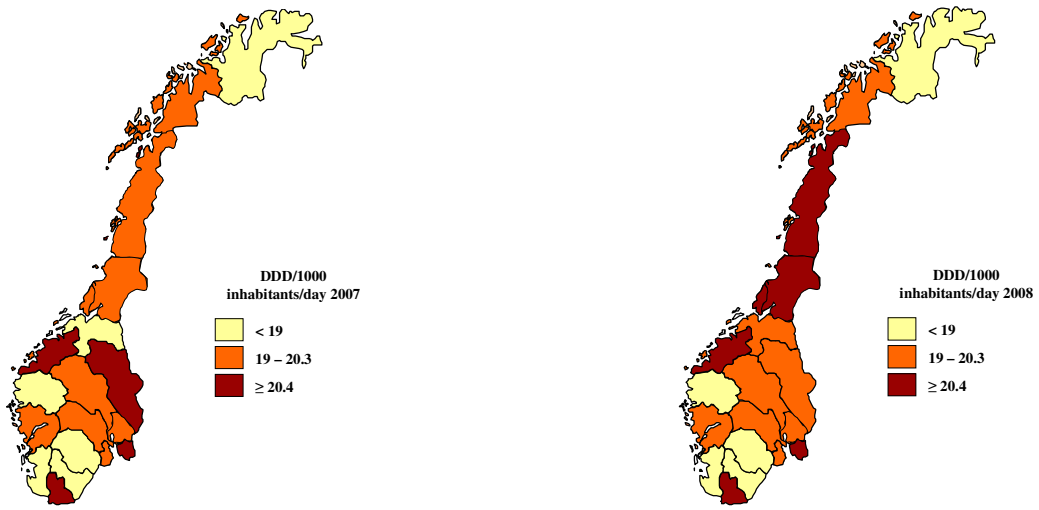

FIGURE 9. Sales of antibacterial agents for systemic use (ATC group J01) in the different counties of Norway in 2007 (left) and 2008 (right).

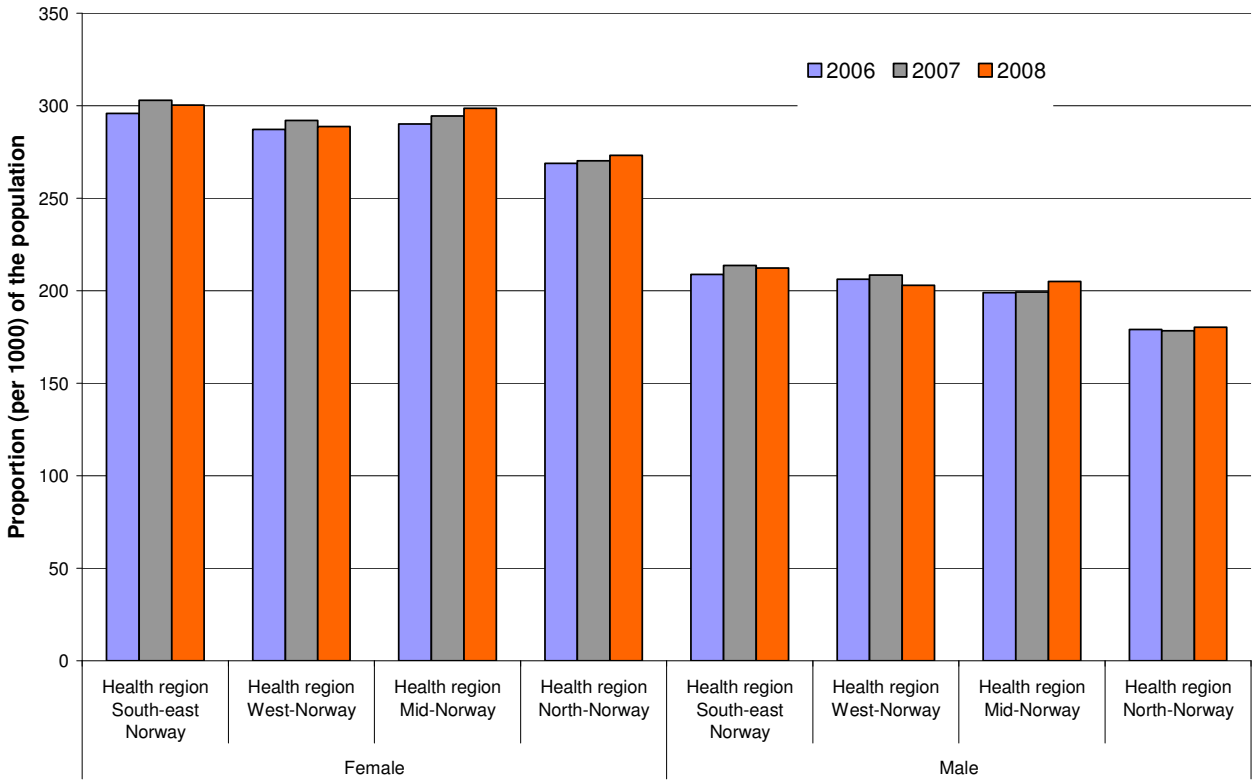

FIGURE 10. One year prevalence (%) of systemic antibacterial use in ambulatory care by gender and health region in Norway for the years 2006-2008. Antibacterials for systemic use include ATC group J01, vancomycin (A07AA09) and metronidazole (P01AB01).

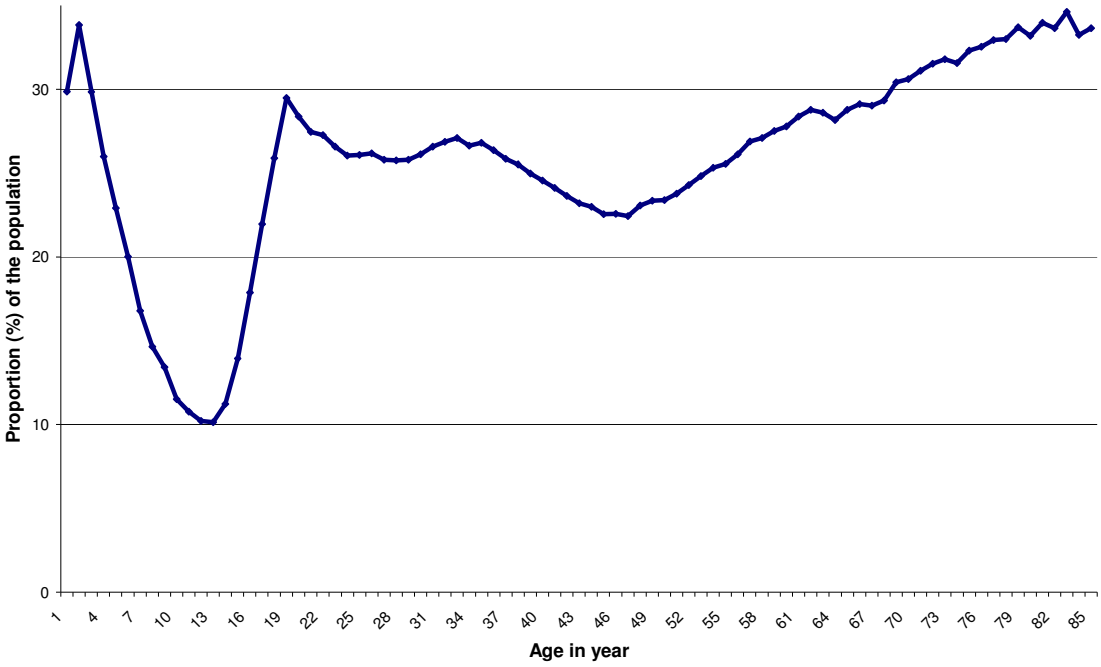

FIGURE 11. One year prevalence (%) of systemic antibacterial use in ambulatory care by age (from 1 - 85+ year) in Norway in 2008. Antibacterials for systemic use include ATC group J01, vancomycin (A07AA09) and metronidazole (P01AB01).

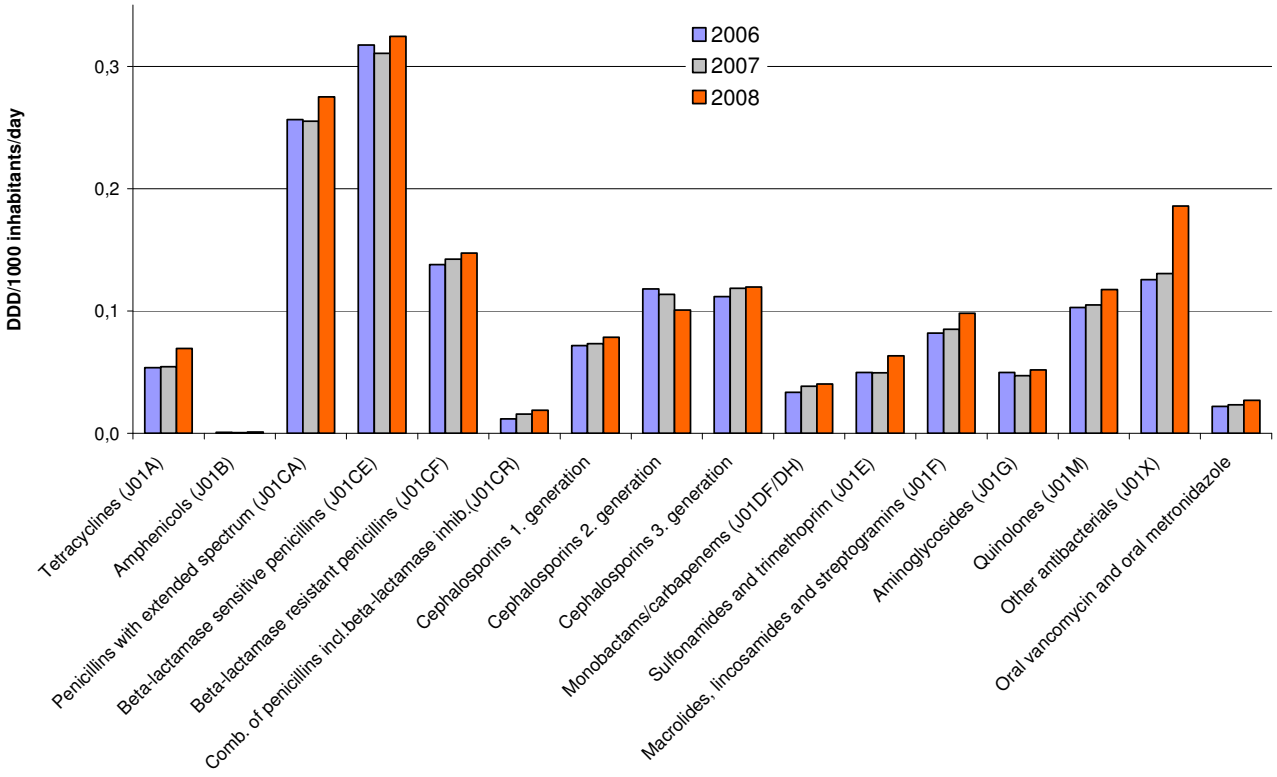

FIGURE 12. Proportions of antibacterial agents for systemic use in Norwegian hospitals 2006-2008, measured in DDD/1,000 inhabitants/day.

VI. OCCURRENCE OF ANTIMICROBIAL RESISTANCE

A. ANIMAL CLINICAL ISOLATES

Jarle Mikalsen, Madelaine Norström, Marianne Sunde

According to the NORM-VET plan, the clinical isolates included in 2008 were beta-haemolysin-producing *Staphylococcus* spp. from skin or ear infections in dogs. Sampling, laboratory methods and data processing are described in Appendix 3

Staphylococcus spp. from dog

A total of 200 isolates of beta-haemolysin-producing *Staphylococcus* spp. from clinical samples were submitted to NORM-VET. Further species identification of these isolates resulted in 185 isolates of *Staphylococcus pseudintermedius,* 14 *Staphylococcus schleiferi* spp. *coagulans* and one isolate of *Staphylococcus aureus.* See textbox "Species identification of beta-haemolysin-producing staphylococci from dogs" for more information. The results are presented in Table 10, Figures 13-14, and in the text.

TABLE 10. Beta-haemolysin-producing *Staphylococcus* spp. from dog (n=200, *Staphylococcus pseudintermedius* n=185, *Staphylococcus schleiferi* spp. *coagulans* n=14, *Staphylococcus aureus* n=1).

		Resistance (%)		Distribution (%) of MIC values (mg/L)													
Substance	Sample	%	[95% CI]	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥256
Beta-lactamase	Dog	76															
Penicillin G	Dog			23.5	0.5	2.5	9.5	8.5	5	5	8.5	37.0					
Oxacillin	Dog	0.0	[0.0-2.3]			1.5	48.0	49.0	1.5								
Cephalothin	Dog	0.0	[0.0-2.3]		8.0	90.5	1.5										
Gentamicin	Dog	1.5	[0.4-4.7]					96.0	2.5		1.0		0.5				
Kanamycin	Dog	12.5	[8.4-18.1]				1.0	11.0	60.5	14.0	1.0				12.5		
Ciprofloxacin	Dog	0.5	[0-3.2]		13.0	66.0	18.5	2.0				0.5					
Trimethoprim	Dog	10.0	[6.4-15.2]						1.0	28.0	49.0	12.0	4.5		5.5		
Clindamycin*	Dog	10.5	[6.8-15.8]				87.5	2.5	1.0		0.5	1.0	1.0	0.5	6.0		
Erythromycin	Dog	10.5	[6.8-15.8]				36.0	53.0	0.5						10.5		
Chloramphenicol	Dog	2.0	[0.6-5.4]							2.5	90.0	5.5			2.0		
Tetracycline	Dog	39.5	[32.7-46.7]					60.0	0.5			0.5		32.5	6.5		
Fusidic acid	Dog	62.5	[55.4-69.2]		1.5	12.5	20.0	3.5	1.0	6.0	6.5	29.5	19.5				

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. * Of 21 isolates resistant to clindamycin, three were classified as resistant based on inducible clindamycin resistance.

COMMENTS

The occurrence of resistance among beta-haemolysin-producing *Staphylococcus* spp. was high. Only 13.5% (27 isolates) were susceptible to all antimicrobial agents tested for. Interestingly, eight out of these 27 isolates (29.6%) were *S. schleiferi* ssp. *coagulans* isolates, for which only resistance to fusidic acid was detected. Altogether, 19.5% were resistant to one class of antibiotics (mainly penicillin or fusidic acid), 32.0% to two (mainly penicillin and fusidic acid or tetracycline), 18.5% to three (penicillin, tetracycline and fusidic acid) and 16.5% to four or more antimicrobial agents, as shown in Figure 14. Furthermore, three of the erythromycin resistant isolates did exhibit inducible clindamycin resistance.

Fusidic acid is the most commonly used preparation for local treatment of skin infections including otitis externa in dogs. However, for systemic treatment of skin infections in dogs a combination of trimethoprim-sulfamethoxazole was the most commonly used antimicrobial agent followed by erythromycin, lincomycin and clindamycin.

Compared to previous years, the overall trend shows that resistance to penicillin, fusidic acid and tetracycline remains high (Figure 13). A small drop was seen in 2004, though the data collection from that year differed substantially from the other years as the isolates were from dogs with a "first time pyodermia" and included only a small number of isolates (n=59). See textbox "Antimicrobial resistance in *Staphylococcus pseudintermedius* in the Norwegian dog population" for further information.

Levels of trimethoprim resistance have decreased from 21% in 2004 to 10% in 2008 using the current NORM-VET breakpoints as shown in Figure 13.

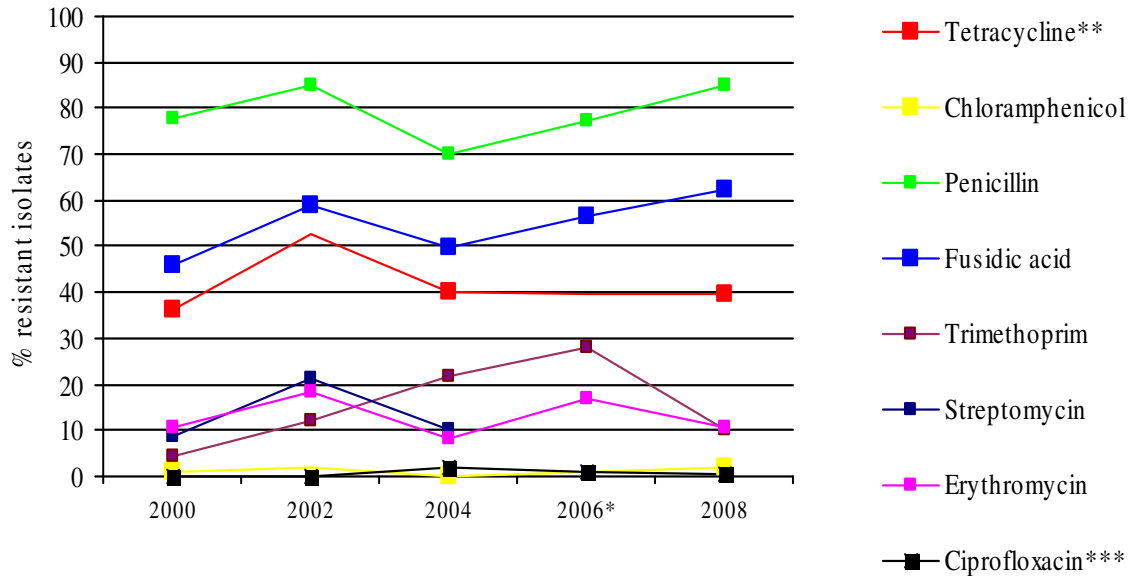

FIGURE 13. Prevalence of resistance to various antimicrobial agents in beta-haemolysin-producing *Staphylococcus* spp. from skin and ear infections in dogs 2000-2008. *Interpolated result for 2006 as no data are available (not monitored in 2006). The breakpoints in NORM-VET 2008 were applied. ** Oxytetracycline instead of tetracycline before 2008. ***Enrofloxacin instead of ciprofloxacin in 2004.

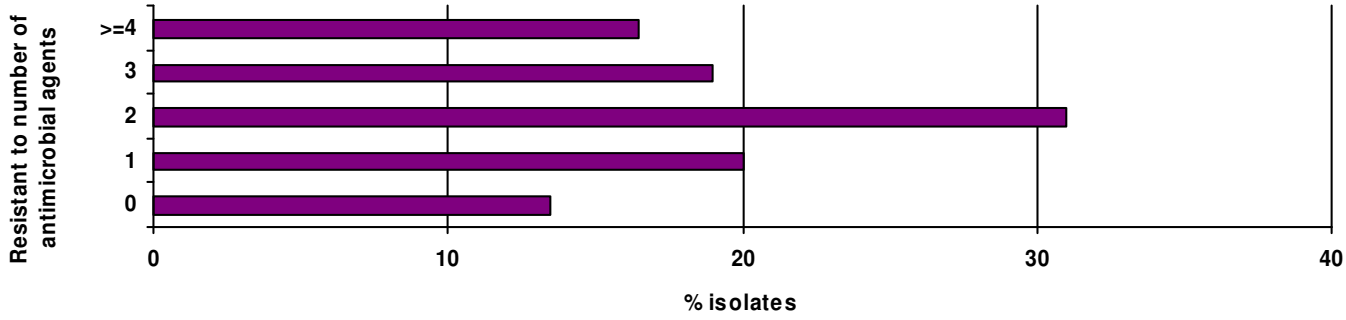

FIGURE 14. Antimicrobial resistance profile for beta-haemolysin-producing *Staphylococcus* spp. in dogs (n=200). Proportion of isolates susceptible to all antimicrobial agents included and resistant to one, two, three, and four or more antimicrobial classes.

Methicillin resistant *Staphylococcus pseudintermedius* from dogs in Norway

High resistance frequencies among *Staphylococcus pseudintermedius* isolates from dogs have been reported from Norway as well as from many other countries. So far, methicillin resistant *S. pseudintermedius* (MRSP) have been rather uncommon. However, during the last few years MRSP in dogs have been reported with increased frequencies from different European countries as well as from the United States[1]. MRSP is considered to be a serious veterinary and public health problem. In Norway MRSP in dogs was detected for the first time in 2008.

MRSP was isolated from two dogs treated at the same small animal clinic during the summer of 2008. The dogs suffered from severe infections. Puls-field gel electrophoresis (PFGE) showed equal banding patters, indicating nosocomial spread at the clinic. The isolates were extremely multiresistant, being susceptible to tetracycline only, and expressed resistance to the following antimicrobial agents: enrofloxacin, trimethoprim-sulfonamides, fusidic acid, erythromycin, clindamycin, gentamicin, kanamycin, chloramphenicol and all beta-lactams. MRSP has fortunately not been detected from animals hospitalized at the clinic since the summer of 2008.

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Species identification of beta-haemolysin-producing staphylococci from dogs

Until recently, *Staphylococcus intermedius* was presumed to be the most common staphylococcal species involved in ear and skin infections in dogs. However the taxonomy has been changed, and it is now proposed that *Staphylococcus pseudintermedius*[1], a novel species in 2005, is the most common canine species. Differentiation between *S. intermedius* and *S. pseudintermedius* by use of standard bacteriological testing may be difficult. In addition, other species of beta-haemolysin-producing staphylococci can also be involved in skin and ear infections in dogs, like *Staphylococcus aureus* and *Staphylococcus schleiferi* subsp. *coagulans*. Therefore, correct species identification of beta-haemolysin-producing staphylococci may be a challenge. To get more knowledge about the various staphylococcal species involved in skin and ear infections in dogs in Norway we decided to species determine the beta-haemolysin-producing staphylococcal isolates included in the 2008 NORM-VET program (n=200) by the use of a molecular method recently described by Bannoehr et al[2], 16S rRNA sequencing and biochemical testing. Our investigations showed the following species distribution:

S. pseudintermedius: 185 isolates (92.5 %)
S. schleiferi subsp. *coagulans*: 14 isolates (7%)
S. aureus: one isolate (0.5 %)
S. intermedius was not detected.

Interestingly, the resistance profiles of the *S. schleiferi* subsp. *coagulans* isolates differed markedly from those of *S. pseudintermedius* by being substantially more susceptible to the antimicrobial agents included in our test panel.

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Antimicrobial resistance in *Staphylococcus pseudintermedius* in the Norwegian dog population

The occurrence of antimicrobial resistance in *Staphylococcus pseudintermedius* from skin and ear infections in a representative sample of dogs not treated with antimicrobial agents prior to sampling was examined. The obtained isolates were further examined for genetic polymorphism and genetic background of resistance.

A total of 59 isolates of *S. pseudintermedius* originating from 96 samples of 91 dogs in five different regions in Norway were included in this study. Susceptibility testing was performed using a broth dilution method. Resistant isolates were subjected to polymerase chain reaction (PCR) for detection of resistance genes. All isolates were subjected to pulsed-field gel electrophoresis (PFGE) to examine the genetic polymorphism.

In total, 19% of the isolates were susceptible to all antimicrobial agents included. Resistance to penicillin was most prevalent (70%) followed by resistance to fusidic acid (49%) and oxytetracycline (42%). Resistance to quinolones or cephalosporins was not observed. Resistance to penicillin, tetracycline and erythromycin was mediated by the *blaZ* beta-lactamase gene, the *tetM* gene and the *ermB* gene, respectively. One of the fusidic acid resistant isolates harboured a *fusC* gene, whereas the mechanisms involved in resistance in the other fusidic acid resistant isolates remained unknown. PFGE showed a high genetic polymorphism of *S. pseudintermedius.*

This study indicates that the occurrence of antimicrobial resistance is common among *S. pseudintermedius* from dogs not treated with antimicrobial agents prior to sampling and furthermore that there is a high genetic polymorphism among *S. pseudintermedius.*

References

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Methicillin resistant *Staphylococcus aureus* (MRSA) from animals in Norway

Methicillin resistant *Staphylococcus aureus* (MRSA) is one of the most important human bacterial pathogens. MRSA was until year 2000 mainly associated with infections in human hospitals, but have recently emerged outside the health care settings. During the last few years a new "animal-adapted" MRSA variant has emerged among animals in several European countries, particularly among pigs[1]. The MRSA isolates are primarily classified as sequence type 398 (ST398). The animals are usually asymptomatic carriers but cases of clinical infections are described. ST398 MRSA can spread from animals to humans and is considered to represent a new zoonosis.
MRSA can also be transferred from humans to animals ("reverse zoonosis")[1]. Pets and horses are at special risk of acquiring MRSA due to the close contact with owners and handlers. Isolates from infected pets and horses are usually known human MRSA variants. Infected animals may spread MRSA to other individuals, and of special importance is the risk of re-infection of individuals under treatment for MRSA eradication. Below is an overview over MRSA cases in animals in Norway.

Year	Animal species	Genotype	Possible source of MRSA
2002[2]	Horse	ST8	Unknown
2008	Cat	ST45 - t1081	MRSA positive owner
2008	Dog	ST8 - t324	MRSA positive owner
2008	Dog	ST22 - t032	Surgery in veterinary clinic abroad
2008	Dust/pig holding	ST8 - t008	MRSA positive owner
2008	Pig	ST8 - t008	MRSA positive owner

Genotyping (except the isolate from 2002) was carried out at the National Reference Laboratory for MRSA in Norway, St Olavs hospital, Trondheim.

MRSA screening studies in animals in 2008
In 2008, the EU-initiated baseline survey on the prevalence of *Salmonella* spp. in herds of breeding pigs in the European countries also included a survey of MRSA prevalence in the same holdings. A total of 252 holdings were analysed. Only one dust sample from a production holding was positive. This gives us an estimated prevalence of MRSA of 0.40% (CI: 0.01 % - 2.19 %). The MRSA isolate was identified as a common human clone, MRSA ST8, spatype t008. MRSA screening of family members at the farm showed that two persons were positive, carrying the same MRSA variant as detected in the dust. Follow-up testing was performed twice. In the first follow-up dust samples from all rooms with pigs, and swabs from the nostrils of all pigs were tested. One pen with positive pigs and one positive dust sample were found in the first testing. Pigs in the positive pen were slaughtered and washing and disinfection performed. In the second follow-up (three months later) only dust samples representing dust from all pens in the herd were tested, none of the samples were positive. The most likely explanation for the positive finding in the pigs in this herd is human to animal transmission.

In a collaborative project between Norwegian School of Veterinary Science and National Veterinary Institute, swabs of the nostrils of 1,000 slaughter pigs were investigated for MRSA[3]. The samples were taken at 10 different slaughter houses and the animals originated from 200 different farms. MRSA was not detected.

Final remarks
The animal adapted ST398 MRSA has so far not been found in animals in Norway. Other MRSA variants have been detected in animals a few times. Most of these cases were detected in 2008. This is probably a consequence of increased focus on MRSA in animals due to the emergence of ST398 MRSA among livestock. NORM-VET has as a consequence of this emerging problem included screening of MRSA in the different animal populations, and a MRSA screening project in horses is currently running.

References

1. Weese, J. S., van Duijkeren E. Methicillin-resistant *Staphylococcus au*reus and Staphylococcus *pseudintermedius* in veterinary medicine. Veterinary Microbiology, (2009) doi:10.1016/j.vetmic.2009.01.039.
2. L'Abee-Lund, T., Wold A., Strand E., Sørum H. Post-operative MRSA-infection in a horse – a case report. Poster, Vintermøtet Norsk Forening for Mikrobiologi, 2003.
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Marianne Sunde, Hanne Tharaldsen and Madelaine Norström, National Veterinary Institute, Oslo.

B. INDICATOR BACTERIA FROM ANIMALS AND FOOD

Jarle Mikalsen, Madelaine Norström, Marianne Sunde

The prevalence of acquired antimicrobial resistance among certain bacteria of the normal enteric microflora can be used as an indicator of the selective pressure from use of antimicrobial agents in 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 to get a better overview of the resistance situation, detect trends and evaluate the effects of interventions.

In NORM-VET, *Escherichia coli* and *Enterococcus* spp. are used as indicator bacteria. In 2008, indicator bacteria from dogs and breeding swine were included in the monitoring. The substances included in the test panels might not always be substances used in veterinary medicine, but are included because of their importance for human health. Some of the cut-off values defining resistance (breakpoints) applied in NORM-VET have been changed over the years. To facilitate comparisons, data on prevalence of resistance presented in earlier reports have been recalculated using the cut-off values applied in 2008. Sampling, laboratory methods and data processing are described in Appendix 3.

Escherichia coli from dogs and breeding swine

A total of 178 and 272 faecal samples from dogs and breeding swines, respectively, were collected. For dogs, *E. coli* was isolated from 160 (89.9%) of the faecal samples. For swine, *E. coli* was isolated from 258 (94.9%) of the faecal samples. One isolate per sample positive for *E. coli* was susceptibility tested. The results are presented in Table 11, Figures 15-16, and in the text.

TABLE 11. Antimicrobial resistance in *Escherichia coli* from faecal samples from dogs (n=160) and swine (n=258) in 2008.

		Resistance %		Distribution (%) of MIC-values (mg/L)													
Substance	Sample	[95% CI]		0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	256	> 256
Tetracycline	Dog	3.8	[1.6-8.4]					42.5	53.8				1.2	1.2	1.2		
	Swine	4.3	[2.3-7.8]				1.9	58.5	35.3				1.2	1.6	1.6		
Chloramphenicol	Dog	2.5	[0.8-6.7]						1.2	54.4	41.2	0.6	0.6		1.2	0.6	
	Swine	0.0	[0.0-1.8]					0.4	6.6	64.3	28.7						
Florfenicol	Dog	0.6	[0.0-3.9]							31.2	67.5	0.6		0.6			
	Swine	0.0	[0.0-1.8]							41.9	56.6	1.6					
Ampicillin	Dog	13.8	[9.0-20.4]					4.4	54.4	27.5		0.6		13.1			
	Swine	4.3	[2.3-7.8]				3.1	23.3	59.3	10.1				4.3			
Ceftiofur	Dog	0.6	[0.0-3.9]		3.8	35.6	55.0	5.0					0.6				
	Swine	0.0	[0.0-1.8]		5.8	57.8	35.3	0.8									
Cefotaxime	Dog	0.6	[0.0-3.9]	69.4	27.5	2.5				0.6							
	Swine	0.0	[0.0-1.8]	89.5	10.5												
Trimethoprim	Dog	5.6	[2.8-10.7]			21.9	63.8	8.1	0.6					5.6			
	Swine	3.5	[1.7-6.8]			45.3	48.4	2.7		0.4				3.1			
Sulfamethoxazole	Dog	10.0	[6.0 -16.0]									55.6	31.9	2.5			10.0
	Swine	6.6	[4.1-10.5]									79.1	14.3				6.6
Streptomycin	Dog	9.4	[5.5-15.3]						1.2	51.2	35.6	2.5	0.6	4.4	2.5	1.2	0.6
	Swine	19.0	[14.5-24.4]						1.6	35.7	42.2	1.6	3.1	4.7	7.0	2.7	1.6
Gentamicin	Dog	0.6	[0.0-3.9]				51.2	45.6	2.5				0.6				
	Swine	0.0	[0.0-1.8]				44.6	51.2	4.3								
Kanamycin	Dog	1.2	[0.2-4.8]						46.9	50.0	1.2	0.6	1.2				
	Swine	0.8	[0.1-3.1]						41.5	51.6	5.8	0.4	0.8				
Nalidixic acid	Dog	0.6	[0.0-3.9]					1.2	53.1	45.0						0.6	
	Swine	0.0	[0.0-1.8]					2.3	47.3	49.2	1.2						

		Resistance (%)		Distribution (%) of MIC values (mg/L)													
Substance	Sample	[95% CI]		0.008	0.016	0.03	0.06	0.125	0.25	0.5	1	2	4	8	16	32	64
Ciprofloxacin*	Dog	1.9	0.5-5.8		0.6	71.9	25.6	1.2				0.6					
	Swine	0.0	0.0-1.8	0.4	7.8	69.0	22.9										

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. *The Cut-off value from NORM-VET 2006 was used as the distribution of the MIC values probably are one or two dilution steps higher than the EUCAST distribution.

RESULTS AND COMMENTS

The cut-off value for ciprofloxacin applied in NORM-VET 2006 was used instead of the cut-off value recommended by the European Committee on Antimicrobial Susceptibility Testing (EUCAST) as the distribution of MIC values obtained were one or two dilution steps higher than the EUCAST distribution.

DOG

The occurrence of resistance among faecal *E. coli* isolates from dogs was low. In total, 83.1% of the isolates were susceptible to all antimicrobial agents included. Altogether, 5.6% were resistant to one (predominantly ampicillin), 1.9% to two (mainly ampicillin and sulfamethoxazoles) and 9.4% to three or more antimicrobial agents (Figure 15). Resistance to streptomycin has previously been reported as the predominant resistance determinant in faecal *E. coli* samples from dog. The prevalence of streptomycin resistance has decreased from 10.1% in 2004 to 9.4% in 2008 (NORM-VET 2008 breakpoints, appendix 6), whereas the prevalence of resistance to ampicillin and sulfamethoxazole has increased from 8.8% to 13.8% and from 8.8% to 10.0% in the same period, respectively. Neither of these changes is statistically significant. Three isolates were classified as resistant to fluoroquinolones, although, two of these isolates were probably misclassified due to the chosen cut-off value, whereas one was multiresistant (se below).

One isolate (0.6%) with extended-spectrum beta-lactamase (ESBL) production was detected. This is the third ESBL positive *E. coli* isolate from animals in Norway and signifies animals as a potential zoonotic reservoir for antimicrobial resistance. PCR and sequencing of known genes involved in ESBL production identified a CTX-M-15 gene variant. This isolate had additional resistance to ciprofloxacin, nalidixic acid, tetracycline, gentamicin and kanamycin, leaving few options for antibiotic treatment. The prevalence of ESBL producing *Enterobacteriaceae* isolates are increasing in Norway and needs to be carefully monitored.

SWINE

The data indicate a moderate to low occurrence of resistance among *E. coli* from faecal samples with 77.5 % susceptible to all antimicrobial agents included. Altogether, 13.2% were resistant to one (predominantly streptomycin), 5.0% to two (mainly streptomycin and sulfamethoxazole) and 4.3% to three or more antimicrobial agents (Figure 15). Resistance to streptomycin was the most frequently identified resistance determinant, followed by resistance to sulfamethoxazole, tetracycline and ampicillin. These antimicrobial agents are commonly used for clinical purposes in swine except streptomycin as only dihydrostreptomycin is approved in combination with penicillin for use in animals in Norway. None of the isolates were resistant to chloramphenicol. Veterinary drugs containing chloramphenicol were withdrawn from the Norwegian market in 1992. Moreover, no resistance to the fluoroquinolone ciprofloxacin 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. Overall, marked reductions in the percentage of isolates being resistant to all antimicrobial agents included were seen (Figure 16).

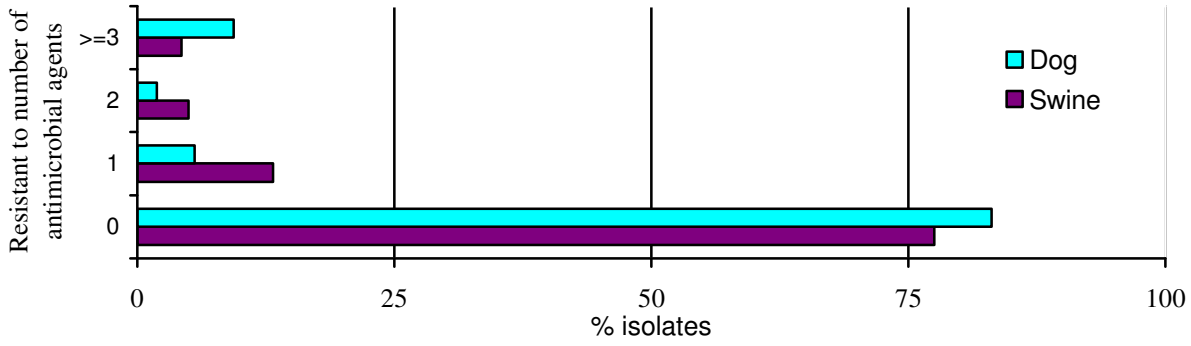

FIGURE 15. Antimicrobial resistance profile for *E. coli* from faecal isolates from dogs (n=160) and swine (n=258) in 2008. Proportions of isolates susceptible to all or resistant to one, two and three or more antimicrobial agents are illustrated.

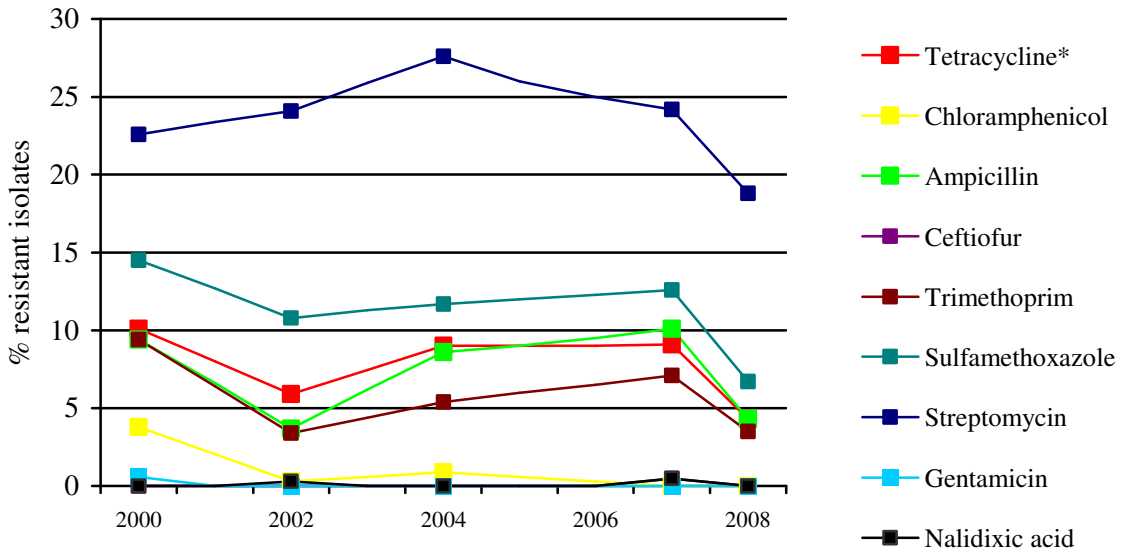

FIGURE 16. Prevalence of resistance to various antimicrobial agents in *E. coli* from swine isolates (faecal samples) 2000-2008. The breakpoints used in NORM-VET 2008 were applied. *Oxytetracycline instead of tetracycline in 2002 and 2004.

Enterococcus spp. from swine

A total of 171 faecal samples from swine were collected and *Enterococcus faecium* or *Enterococcus faecalis* was isolated from 77 samples (45.0%). One isolate per sample positive for *Enterococcus* spp. was susceptibility tested, yielding 65 and 12 isolates of *E. faecium* or *E. faecalis*, respectively. The results are presented in Tables 12-13, Figure 17, and in the text.

TABLE 12. Antimicrobial resistance in *Enterococcus faecalis* isolated from faecal samples of swine (n=12) in 2008.

		Resistant	Distribution (n) of MIC values (mg/L)														
Substance	Sample	(n)	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	≥ 2048
Tetracycline	Swine	8			4					2	4	2					
Chloramphenicol	Swine	0						6	6								
Ampicillin	Swine	0			1	10	1										
Erythromycin	Swine	2			1	2	2	5	2								
Streptomycin	Swine	4										2	4	2			4
Gentamicin	Swine	0							2	9	1						
Kanamycin	Swine	1									4	6	1				1
Vancomycin	Swine	0				2	7	3									
Bacitracin*	Swine	0						3	4	5							
Linezolide	Swine	0				3	9										
Virginiamycin	Swine	NR						1	2	7	2						
Narasin	Swine	0	1	7	3	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. * Measured in U/ml. NR=Not relevant as *E. faecalis* is known to be inherently resistant to the streptogramin virginiamycin.

TABLE 13. Antimicrobial resistance in *Enterococcus faecium* from faecal samples (n=65) from swine in 2008.

		Resistance (%)		Distribution (%) of MIC values (mg/L)													
Substance	Sample		[95%CI]	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	≥ 2048
Tetracycline	Swine	18.5	[10.3-30.5]		80.0	1.5					1.5	16.9					
Chloramphenicol	Swine	0.0	[0.0-7.0]				4.6	69.2	26.2								
Ampicillin	Swine	6.2	[2.0-15.8]	4.6	18.5	32.3	18.5	20.0	4.6			1.5					
Erythromycin	Swine	18.5	[10.3-30.5]		16.9	4.6	13.8	46.2	18.5								
Streptomycin	Swine	13.8	[6.9-25.1]							4.6	18.5	63.1		1.5	1.5	4.6	6.2
Gentamicin	Swine	0.0	[0.0-7.0]					23.1	64.6	12.3							
Kanamycin	Swine	4.6	[1.2-13.7]							1.5	7.7	21.5	44.6	15.4	4.6		4.6
Vancomycin	Swine	0.0	[0.0-7.0]			81.5	9.2	9.2									
Bacitracin*	Swine	3.1	[0.5-11.7]			4.6	3.1	6.2	15.4	66.2	1.5			3.1			
Linezolide	Swine	0.0	[0.0-7.0]		1.5	18.5	78.5	1.5									
Virginiamycin	Swine	0.0	[0.0-7.0]		38.5	23.1	18.5	20.0									
Narasin	Swine	0.0	[0.0-7.0]	7.7	75.4	16.9											

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. * Measured in U/ml.

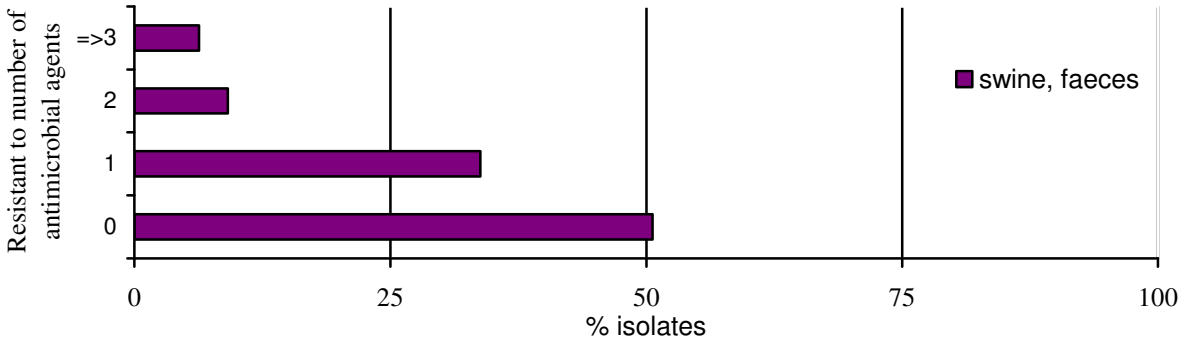

FIGURE 17. Antimicrobial resistance profile for *Enterococcus* spp. (n=77) in 2008. Proportions of isolates susceptible to all or resistant to one, two, and three or more antimicrobial agents are illustrated. Virginiamycin data not included for *E. faecalis*.

RESULTS AND 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. Resistance to virginiamycin is therefore excluded from the interpretation of the data.

The overall occurrence of resistance among *E. faecalis* and *E. faecium* from healthy swine was moderate, as in previous years. Unlike in 2007, no resistance to virginiamycin and narasin was detected in *E. faecium* from swine, and there were no significant changes in the prevalence of resistance to ampicillin, erythromycin and bacitracin. Although the prevalence of erythromycin resistance has been reduced by 11.4% from 2007 to 2008, this difference is not statistically significant due to small sample sizes (67 isolates in 2007 and 65 in 2008).

In total, 50.6% of the isolates were susceptible to all antimicrobial agents included, 33.8% were resistant to one antimicrobial agent, 9.1% were resistant to two, and 6.5% to three or more antimicrobial agents (Figure 17). Resistance to tetracycline was most common, followed by resistance to erythromycin and streptomycin.

C. ZOONOTIC AND NON-ZOONOTIC ENTEROPATHOGENIC BACTERIA

Jørgen Lassen, Trine-Lise Stavnes, Jarle Mikalsen and Madelaine Norström

Zoonotic and non-zoonotic enteropathogenic bacteria represent a considerable public health problem. Furthermore, the occurrence of acquired antimicrobial resistance in such bacteria represents 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, all *Salmonella* isolates from control programmes concerning feed samples, animals and food products, as well as a representative number of *Campylobacter* isolates from broiler are monitored for antimicrobial resistance. Regarding human isolates, all *Salmonella*, *Shigella* and *Yersinia enterocolitica* isolates are included, 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 occurrence of *Salmonella* spp. in food producing animals in Norway is very good. Such animals are virtually free from *Salmonella* spp. To document this favourable situation, Norway runs an extensive surveillance programme that covers both live animals (cattle, pigs and poultry) and meat samples (cattle, pigs, sheep and poultry). The *Salmonella* isolates examined in NORM-VET include those that are detected in this programme, in addition to selected isolates from other relevant projects, as well as clinical submissions to the National Veterinary Institute. The data are presented in Table 14 and in the text.

TABLE 14. Antimicrobial resistance in *S.* Typhimurium (n=12) and other *Salmonella* spp. (n=3) isolates from animals. Distribution (n) of MICs (mg/L).

		Distribution (n) of MIC values (mg/L)															
Substance	Resistance (n)	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Tetracycline	3							7	5			1	2				
Chloramphenicol	3									10	2				1	2	
Florfenicol	3									10	2		1	2			
Ampicillin	3							12							3		
Cefotaxime	0			4	11												
Trimethoprim	0					11	4										
Sulfamethoxazole	3												1	7	4		3
Streptomycin	3									1	7	4		2	1		
Gentamicin	0						12	3									
Kanamycin	0							1	14								
Ciprofloxacin	1		12	2			1										
Nalidixic acid	1								1	13							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.

RESULTS AND COMMENTS

In 2008, a total of 15 isolates of *Salmonella* spp. were susceptibility tested. Of the 12 *S.* Typhimurium, seven isolates were from dogs, two isolates each from cattle and cats, and one isolate from a horse. Four (33.3%) of the *S.* Typhimurium isolates had resistance to two or more classes of the antimicrobial agents tested. Three strains, two from dogs and one from cattle, exhibited penta-resistantant phenotype and were typed as *S.* Typhimurium DT104. One isolate from dog, which was not typable, displayed resistance to both ciprofloxacin and nalidixic acid.

Salmonella from human clinical specimens

In 2008 The National Reference Laboratory for Enteropathogenic Bacteria recieved a total of 1,967 *Salmonella* isolates from human infections of which 13.1% were reported acquired in Norway, 83.5% acquired abroad, whereas the place of origin is unknown for 3.4%. The incidence rate was 41 per 100,000. Altogether 958 (49%) of the isolates were *S.* Enteritidis of which only 48 (5%) were infected in Norway, while 307 (16%) of the isolates were *S.* Typhimurium of which altogether 96 (31%) were reported infected in Norway. The relatively high proportion of domestically acquired *S.* Typhimurium infections is mainly 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 salmonellosis acquired in Norway are wild birds and hedgehogs, imported food products and patients infected abroad. Thus, the isolates categorized as "infected in Norway" also partly reflect the *Salmonella* situation outside Norway.
The proportion of multiresistant *S.* Typhimurium DT104 from domestically acquired cases of *S.* Typhimurium infections was 5.8% (against 6.2% in 2007), from infections acquired abroad 5.5% (as was also the case in 2007). In total, 291 isolates of *S.* Typhimurium (of which 24 with unknown places of origin are not included in the following tables) , 892 isolates of *S.* Enteritidis, 15 isolates of *S.* Typhi, 15 isolates of *S.* Paratyphi A and two isolates of *S.* Paratyphi B, and 642 isolates of other *Salmonella* spp. were susceptibility tested. The results are presented in Tables 15-18, Figures 18-22, and in the text. Sampling, laboratory methods, and data handling are described in Appendix 4.

TABLE 15. *Salmonella* Typhimurium isolates (n=86), including multiresistant DT104 (n=5), from patients infected in Norway. Distribution (%) of antimicrobial susceptibility categories. Sampling, laboratory methods, and data handling are described in Appendix 4. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 0.5	> 8	0.0	60.5	39.5
Chloramphenicol	≤ 8	> 8	90.7	-	9.3
Tetracycline*			61.6	-	38.4
Nalidixic acid	≤ 16	> 16	97.7	-	2.3
Ciprofloxacin	≤ 0.5	> 1	100.0	0.0	0.0
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	94.2	0.0	5.8

* The epidemiological cut-off value defined by EUCAST is applied. There are no clinical breakpoints defiend by AFA as this substance is not relevant for treatment. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 16. *Salmonella* Typhimurium isolates (n=181), including multiresistant DT104 (n=10), from patients infected outside Norway. Distribution (%) of antimicrobial susceptibility categories. Sampling, laboratory methods, and data handling are described in Appendix 4. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 0.5	> 8	0.0	51.9	48.1
Chloramphenicol	≤ 8	> 8	85.1	-	14.9
Tetracycline*			34.8	-	65.2
Nalidixic acid	≤ 16	> 16	89.5	-	10.5
Ciprofloxacin	≤ 0.5	> 1	98.9	1.1	0.0
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	87.3	1.7	11.0

* The epidemiological cut-off value defined by EUCAST is applied. There are no clinical breakpoints defiend by AFA as this substance is not relevant for treatment. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 17. *Salmonella* Enteritidis isolates from patients (n=892[#]). Distribution (%) of antimicrobial susceptibility categories. Sampling, laboratory methods, and data handling are described in Appendix 4. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 0.5	> 8	0.3	94.3	5.4
Chloramphenicol	≤ 8	> 8	99.6	-	0.4
Tetracycline*			97.2	-	2.8
Nalidixic acid	≤ 16	> 16	71.4	-	28.6
Ciprofloxacin	≤ 0.5	> 1	99.9	0.0	0.1
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	99.0	0.0	1.0

Place of infection: Norway (n=43), abroad (n=828), unknown (n=21). * The epidemiological cut-off value defined by EUCAST is applied. There are no clinical breakpoints defiend by AFA as this substance is not relevant for treatment. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 18. *Salmonella* spp. (excluding *S.* Typhimurium, *S.* Enteritidis, *S.* Typhi and *S.* Paratyphi) (n=642[#]). Distribution (%) of antimicrobial susceptibility categories. Sampling, laboratory methods, and data handling are described in Appendix 4. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 0.5	> 8	0.0	87.7	12.3
Chloramphenicol	≤ 8	> 8	94.5	-	5.5
Tetracycline*			78.5	-	21.5
Nalidixic acid	≤ 16	> 16	81.3	-	18.7
Ciprofloxacin	≤ 0.5	> 1	97.2	1.4	1.4
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	88.3	0.8	10.9

Place of infection: Norway (n=81), abroad (n=535), unknown (n=26). * The epidemiological cut-off value defined by EUCAST is applied. There are no clinical breakpoints defined by AFA as this drug is not relevant for treatment. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

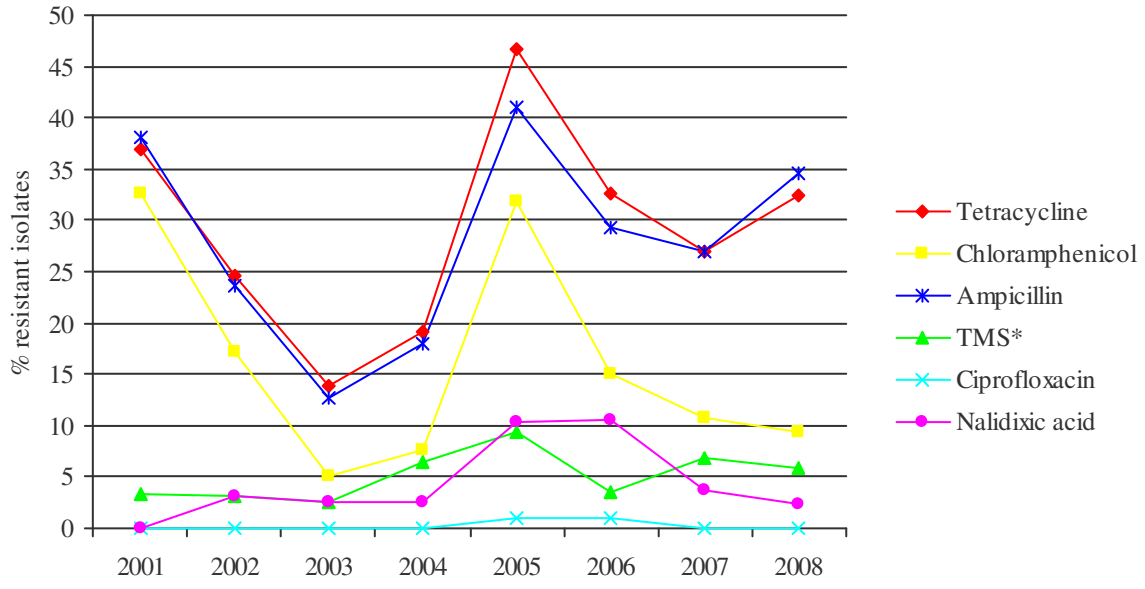

FIGURE 18. Prevalence of resistance to various antimicrobial agents in *Salmonella* Typhimurium including multiresistant DT104 from humans infected in Norway 2001-2008. *TMS=Trimethoprim-sulfamethoxazole.

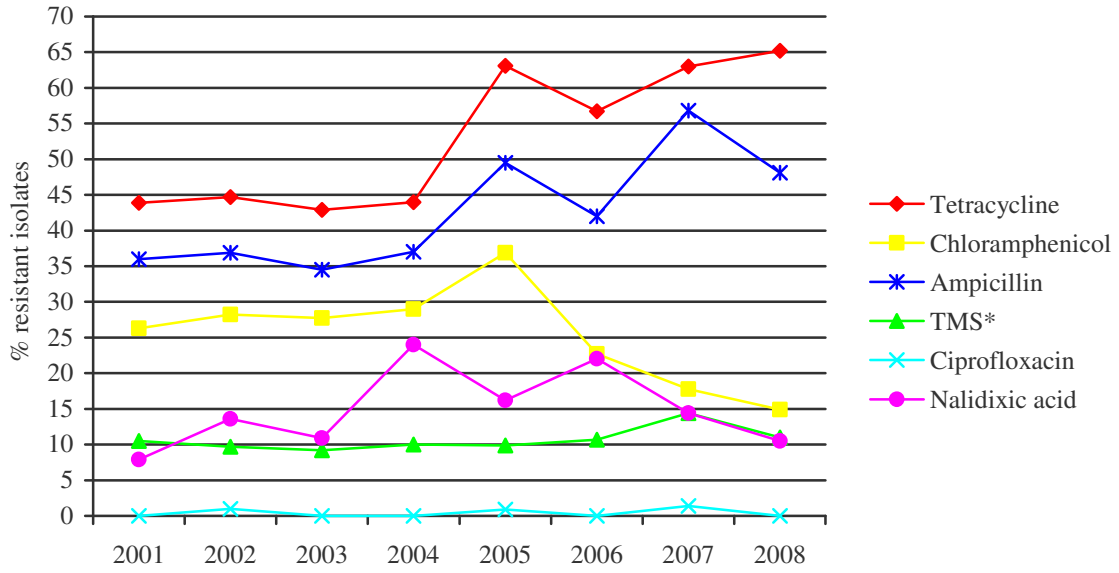

FIGURE 19. Prevalence of resistance to various antimicrobial agents in *Salmonella* Typhimurium including multiresistant DT104 from humans infected outside Norway 2001-2008. *TMS=Trimethoprim-sulfamethoxazole.

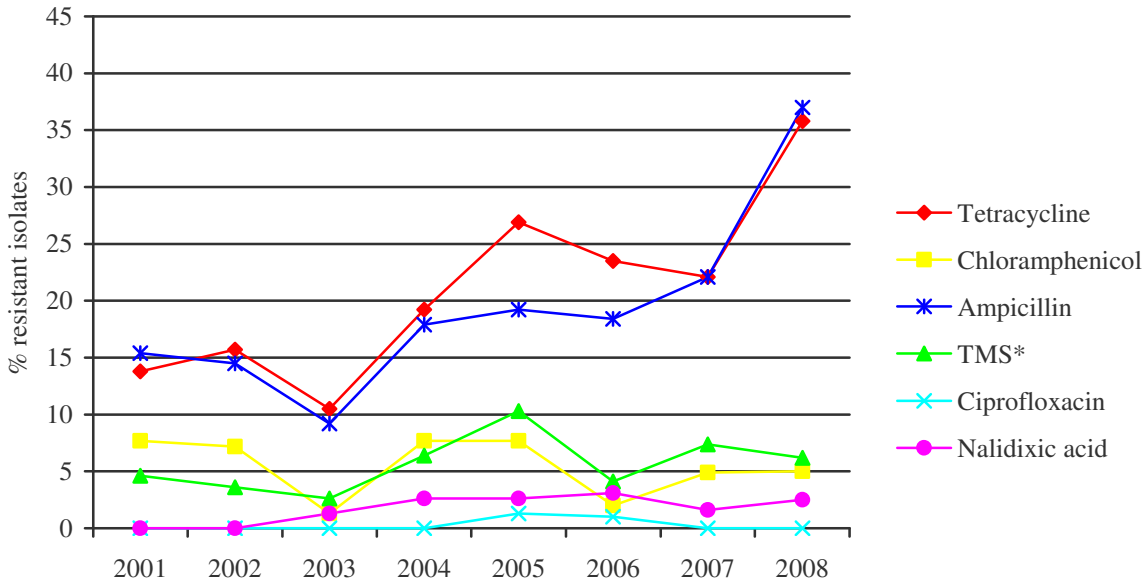

FIGURE 20. Prevalence of resistance to various antimicrobial agents in *Salmonella* Typhimurium from humans infected in Norway 2001-2008, excluding multiresistant DT104. *TMS=Trimethoprim-sulfamethoxazole.

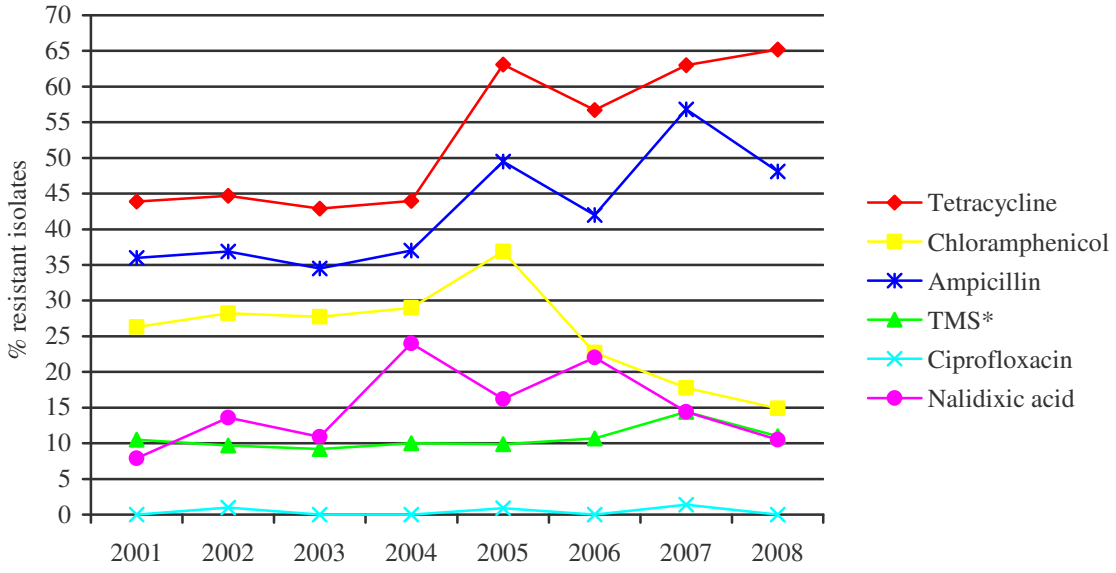

FIGURE 21. Prevalence of resistance to various antimicrobial agents in *Salmonella* Typhimurium from humans infected outside Norway 2001-2008, excluding multiresistant DT104. *TMS=Trimethoprim-sulfamethoxazole.

RESULTS AND COMMENTS

For *S.* Typhimurium, resistance to tetracycline was the most commonly observed followed by resistance to ampicillin, chloramphenicol, trimethoprim-sulfamethoxazole and nalidixic acid.

The proportion of *S.* Typhimurium isolates susceptible to all antimicrobial agents tested was higher for the category "infected in Norway" (55.8%) than for the "infected abroad" category (32.6%) (Figure 22). Multiresistant strains, defined as resistant to two or more antimicrobial agents, were more common in the category "infected abroad" (53%) than in the category "infected in Norway" (36%). The prevalence of resistance for the years 2001-2008 to various antimicrobial agents in human isolates of *S.* Typhimurium, acquired in Norway (Figure 20) shows that the prevalences of resistance to ampicillin and tetracyclin have increased compared to 2007 (from 26.9% and 26.9% to 34.5% and 38.4%, respectively). For isolates acquired abroad (Figure 21), there has been a slight increase in resistance against tetracycline (from 63.0% to 65.2%), whereas the resistance to ampicillin has decreased (from 56.8% to 48.1%),

The vast majority of *S.* Enteritidis isolates had been acquired abroad (Table 17). The proportion of *S.* Enteritidis isolates resistant to the different antimicrobial agents included was, except for nalidixic acid, considerably lower than for *S.* Typhimurium. In total, 28.6% of the isolates of *S.* Enteritidis were resistant to nalidixic acid. Resistance to ciprofloxacin was found in 0.1%, and none of the isolates were intermediately susceptible. This is a significant decrease compared to 2007 when 13.4% were found to be intermediately susceptible, but more in accordance with the results from the previous years.

With regard to *Salmonella* spp. isolates other than *S.* Typhimurium and *S.* Enteritidis, most infections had been acquired abroad and antimicrobial resistance was frequently detected (Table 18). Resistance to tetracycline was most common, followed by resistance to nalidixic acid and ampicillin. Resistance to ciprofloxacin was observed for 1.4% of the isolates and the same proportion displayed intermediate susceptibility. It is emphasized that the use of fluoroquinolones in Norway is very limited in both human and veterinary medicine.

The few isolates of *S.* Typhi (n=15), *S.* Paratyphi A (n=15) and *S.* Paratyphi B (n=2) in 2008 indicate that multiresistance, including resistance to nalidixic acid, is common in these serovars. With the exception of one case of unknown origin, all infections with these serovars were aquired abroad. Twenty-two isolates (69%, eleven of *S.* Typhi and eleven of *S.* Paratyphi A, none of *S.* Paratyphi B) were resistant to one or more of the antimicrobial agents included in the survey.

In 2008, the marker for possible extended spectrum beta-lactamases (ESBL) was based on zone diameters of cefpodoxime. All isolates with reduced susceptibility to cefpodoxime were further characterized in order to verify the presence of ESBL. A total of nine isolates displayed reduced susceptibility to cefpodoxime; all were identified as ESBL producers.

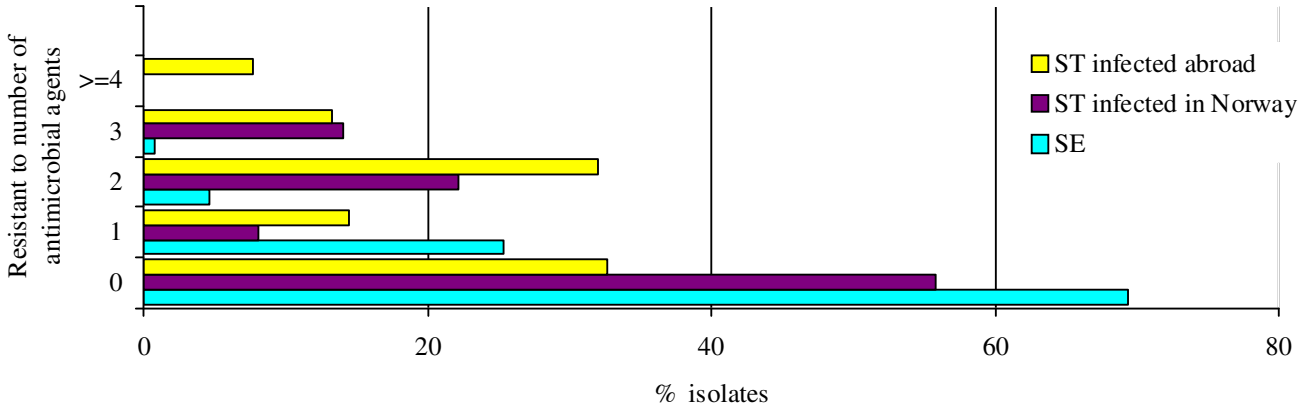

FIGURE 22. Antimicrobial resistance profiles for all *Salmonella* Enteritidis (SE) from humans (n=892) and for *Salmonella* Typhimurium (ST) from humans infected in Norway (n=86) and abroad (n=181), respectively. Proportion of isolates in 2008 resistant to none, one, two, three, or four or more antimicrobial agents are illustrated.

CAMPYLOBACTER SPP.

Campylobacter jejuni from broilers

The isolates of *Campylobacter jejuni* in broilers presented in Table 19, Figure 23 and Figure 26, originate from the Norwegian action plan against *Campylobacter* spp. in broiler meat production. All broiler flocks slaughtered before 50 days of age are tested for the presence of *Campylobacter* spp. In 2008, a total of 128 isolates from broiler flocks (caecal samples) were susceptibility tested. All results are commented in the text.

TABLE 19. Antimicrobial resistance in *Campylobacter jejuni* (n=128) from broiler flocks. Distribution (%) of MICs (mg/L).

	Resistance (%)		Distribution (%) of MIC values (mg/L)													
Substance	[95% CI]		0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Tetracycline	1.6	[0.3-6.2]		71.1	25.8	0.8		0.8	0.8			0.8				
Erythromycin	0.0	[0.0-3.6]				92.2	7.0	0.8								
Streptomycin	4.7	[1.9-10.4]				1.6	32.8	60.9	3.1				0.8	0.8		
Gentamicin	0.0	[0.0-3.6]			1.6	82.8	15.6									
Ciprofloxacin	0.8	[0.0-4.9]	0.8	57.8	33.6	7.0					0.8					
Nalidixic acid	2.3	[0.6-7.2]						1.6	48.4	43.0	4.7		0.8	1.6		

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.

RESULTS AND COMMENTS

The results show that the occurrence of antimicrobial resistance among *C. jejuni* isolates from Norwegian broilers is low. A total of 92.2% of the included isolates were susceptible to all antimicrobial agents tested. However, 3.1% of the isolates were probably misclassified as resistant to streptomycin as a result of the chosen cut-off value.

Resistance to nalidixic acid, streptomycin and tetracycline occurred at low prevalences, and one of the isolates was resistant to both nalidixic acid and ciprofloxacin. The results reflect that treatment with antimicrobial agents in poultry production is very limited. If used, the aminopenicillin amoxicillin or the tetracycline oxytetracycline are the drugs of choice. Nalidixic acid is not used in poultry, but a minor amount of enrofloxacin has been used on exemption from market authorization in recent years (K. Grave, unpublished data).

The resistance levels are similar to those presented in previous NORM/NORM-VET reports as seen in Figure 23. As in earlier years, a high agreement between the prevalence of antimicrobial resistance for *C. jejuni* from humans infected within Norway (Table 20) and isolates from Norwegian broilers are observed, although, domestically acquired human isolates have higher prevalences of resistance towards quinolones (nalidixic acid and ciprofloxacin) and tetracycline.

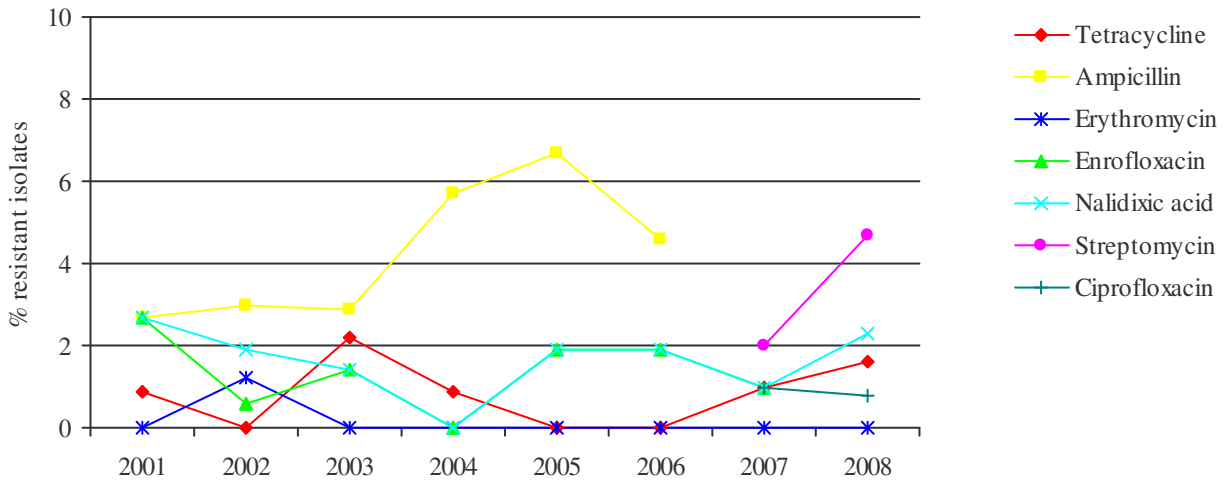

FIGURE 23. Prevalence of resistance to various antimicrobial agents in *Campylobacter jejuni* from Norwegian broilers 2001-2008. The breakpoints for resistance defined in NORM-VET 2006 were applied for the data generated before 2007.

Campylobacter spp. from human clinical specimens

Of the 2,875 cases of human campylobacteriosis registered in Norway in 2008 (incidence rate 59.9 per 100.000), 54% were reported as acquired abroad. The vast majority of cases were sporadic. Case-control studies in Norway have revealed that consumption of broiler meat purchased fresh and drinking of untreated water are important risk factors for domestically acquired campylobacteriosis. A total of 268 isolates of *C. jejuni* (95 from patients infected in Norway, 148 from patients infected abroad and 25 from patients where the origin of infection was unknown), 18 isolates of *C. coli*, six isolates of *C. lari* and one isolate of *C. upsaliensis* were susceptibility tested. The results for *C. jejuni* are presented in Tables 20-23, Figures 24-26, and in the text.

TABLE 20. *Campylobacter jejuni* isolates from patients infected in Norway (n=95). Distribution (%) of antimicrobial susceptibility categories.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Tetracycline	≤ 1	> 1	92.6	-	7.4
Erythromycin	≤ 4	> 4	100.0	-	0.0
Gentamicin	≤ 2	> 4	97.9	2.1	0.0
Nalidixic acid	≤ 16	> 16	93.7	-	6.3
Ciprofloxacin	≤ 0.5	> 1	93.7	0.0	6.3

TABLE 21. *Campylobacter jejuni* isolates from patients infected in Norway (n=95). Distribution (%) of MICs (mg/L).

	≤ 0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Tetracycline	2.1	7.4	28.4	42.1	10.5	2.1		1.1			1.1	1.0	1.0	3.2
Erythromycin			2.1	3.2	28.4	58.9	7.4							
Gentamicin		1.1	2.1	4.2	20.0	52.6	17.9	2.1						
Nalidixic acid						2.1	25.2	51.6	11.6	3.2	1.0			5.3
Ciprofloxacin		14.8	65.2	13.7	1.0						5.3			

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

TABLE 22. *Campylobacter jejuni* isolates from patients infected outside Norway (n=148). Distribution (%) of antimicrobial susceptibility categories.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Tetracycline	≤ 1	> 1	48.6	-	51.4
Erythromycin	≤ 4	> 4	97.3	-	2.7
Gentamicin	≤ 2	> 4	96.0	2.0	2.0
Nalidixic acid	≤ 16	> 16	31.1	-	68.9
Ciprofloxacin	≤ 0.5	> 1	29.0	1.4	69.6

TABLE 23. *Campylobacter jejuni* isolates from patients infected outside Norway (n=148). Distribution (%) of MICs (mg/L).

	≤ 0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Tetracycline		4.7	13.5	20.3	8.8	1.4		1.4	0.6	4.7	5.4	8.1	3.4	27.7
Erythromycin				2.7	25.0	43.9	21.6	4.0	1.4					1.4
Gentamicin			1.4	8.1	21.6	56.0	8.8	2.0		0.7				1.4
Nalidixic acid						1.4	10.1	15.5	3.4	0.7			0.7	68.2
Ciprofloxacin	0.7	8.7	16.9	2.0	0.7	1.4	0.7	1.4	2.0	0.7	64.8			

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

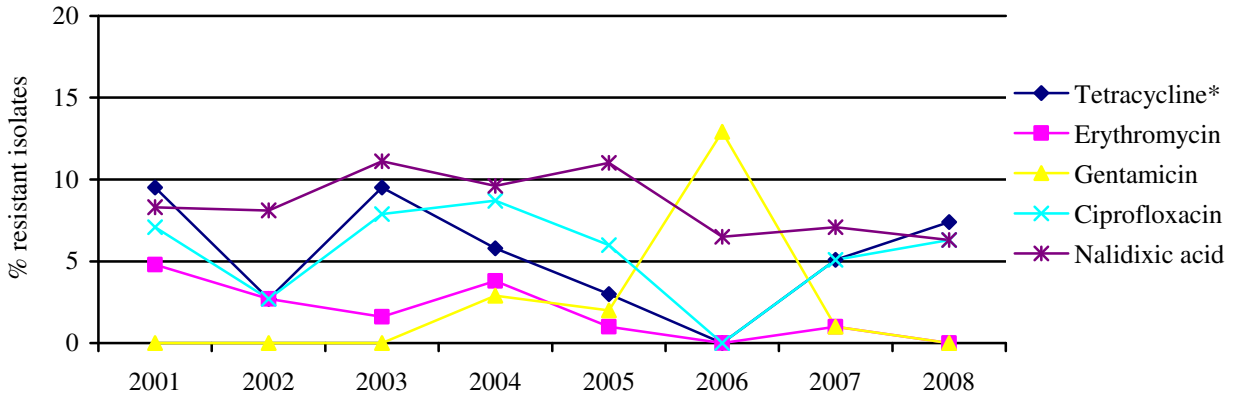

FIGURE 24. Prevalence of resistance in *Campylobacter jejuni,* isolated from humans infected in Norway 2001-2008, to various antimicrobial agents. * Doxycycline before 2006.

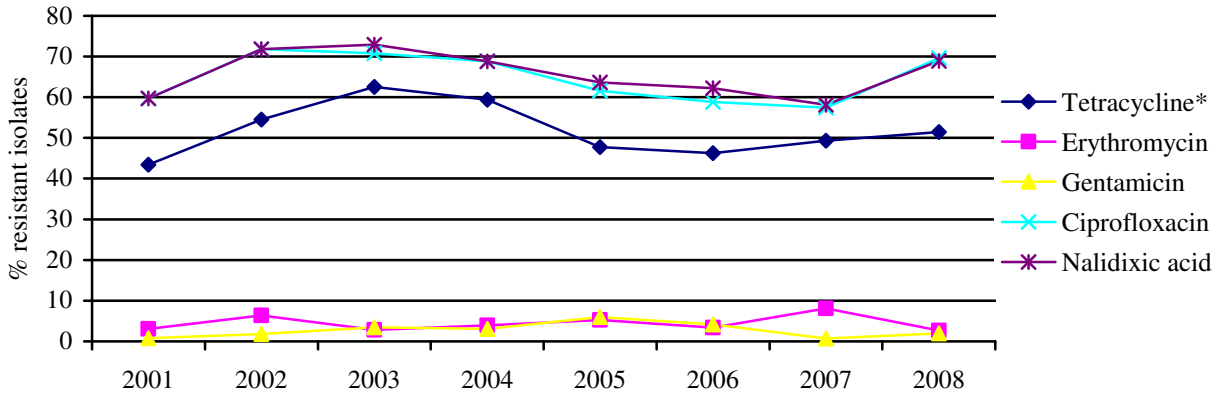

FIGURE 25. Prevalence of resistance to various antimicrobials in *Campylobacter jejuni* from humans infected outside Norway 2001-2008. * Doxycycline before 2006.

RESULTS AND COMMENTS

The data show that resistance was significantly more widespread among *C. jejuni* isolates recovered from patients infected abroad than in patients infected in Norway. Only 27.0% of isolates from infections acquired abroad were susceptible to all antimicrobial agents tested compared to 87.4% of the isolates from patients infected in Norway (Figure 26). The main differences between the two groups were seen for quinolones (ciprofloxacin/ nalidixic acid) with 69.6% resistance in isolates acquired abroad versus 6.3% resistance in isolates acquired in Norway, and tetracycline with 51.4% resistance in isolates acquired abroad versus 7.4% resistance for those acquired in Norway.

The prevalence of resistance to various antimicrobial agents for *C. jejuni* acquired in and outside Norway (Figure 24 and 25) have been fairly stable during the period 2001-2008 except for an unexplained increase in the resistance to gentamicin in indigenious isolates in 2006.

As in earlier years, a high agreement between the prevalence of resistance for *C. jejuni* from humans infected within Norway and isolates from Norwegian broilers are observed, although, domestically acquired human isolates have higher prevalences of resistance towards quinolones (nalidixic acid and ciprofloxacin) and tetracycline

Fifteen *C. coli* isolates were acquired abroad and three were acquired in Norway. Eleven of the isolate acquired abroad were resistant to at least one of the antimicrobial agents, mainly to quinolones or tetracycline. *C. coli* are typically associated with pigs and pork.

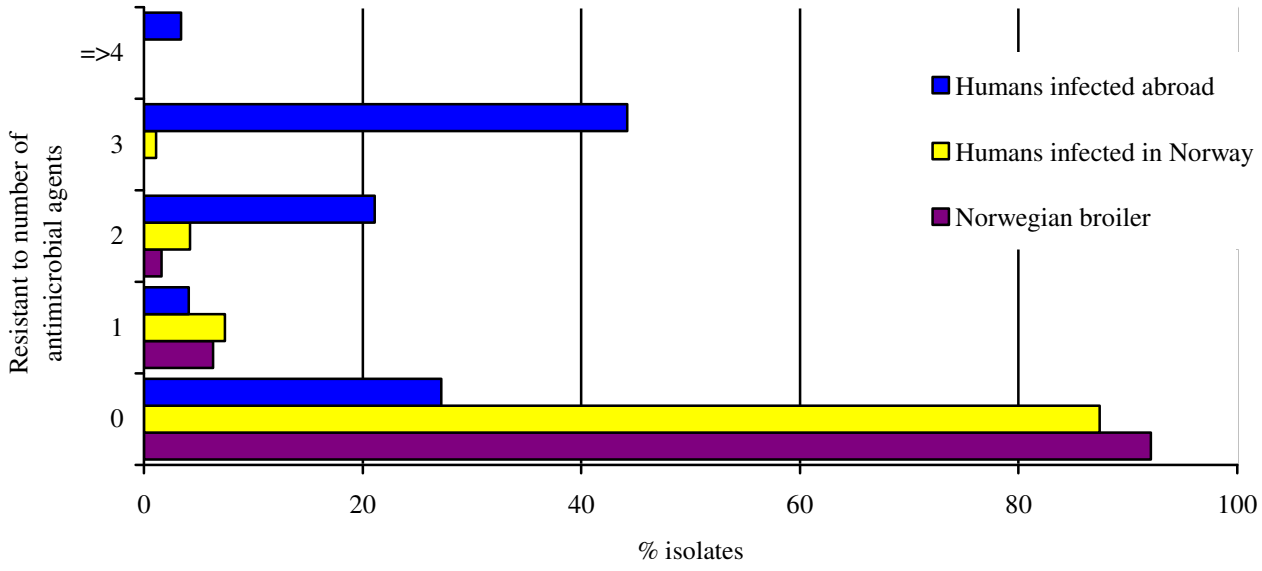

FIGURE 26. Antimicrobial resistance profiles for *Campylobacter jejuni* from Norwegian broiler (n=128), humans infected in Norway (n=95) and humans infected abroad (n=148). Proportion of isolates resistant to none, one, two, three, or four or more antimicrobial agents are illustrated. The isolates from humans were tested for susceptibility to tetracycline, erythromycin, gentamicin, ciprofloxacin and nalidixic acid, whereas the broiler isolates were additionally tested for susceptibility to streptomycin.

Yersinia enterocolitica from human clinical specimens

Most cases of *Yersinia enterocolitica* infections in Norway are domestically acquired. A total of 50 cases of yersiniosis were reported in 2008 (against 71 cases in 2007) giving an incidence rate of 1.0 per 100,000. Of the 50 cases, 46 belonged to serogroup 3 (28 acquired in Norway, 14 abroad and 4 with unknown place of infection), three to serogroup 9 (all acquired in Norway) and one to serogroup 5,27. All *Y. enterocolitica* isolates were susceptibility tested. The results for *Yersinia enterocolitica* serogroup O:3 and serogroup O:9 are presented in Table 24 and Figure 27.

TABLE 24. *Yersinia enterocolitica* serogroup O:3 and serogroup O:9 isolates from human clinical cases (n=49[#]). Distribution (%) of antimicrobial susceptibility categories. Sampling, laboratory methods, and data handling are described in Appendix 4. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 0.5	> 8	0.0	0.0	100.0
Chloramphenicol	≤ 8	> 8	87.8	-	1.2
Tetracycline*			93.9	-	6.1
Nalidixic acid	≤ 16	> 16	85.7	-	14.3
Ciprofloxacin	≤ 0.5	> 1	89.8	8.2	2.0
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	85.7	4.1	10.2

* The epidemiological cut-off value defined by EUCAST is applied. There are no clinical breakpoints defiend by AFA as this substance is not relevant for treatment. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only. [#] Place of infection: Norway (n=31), Abroad (n=14), Unknown (n=4).

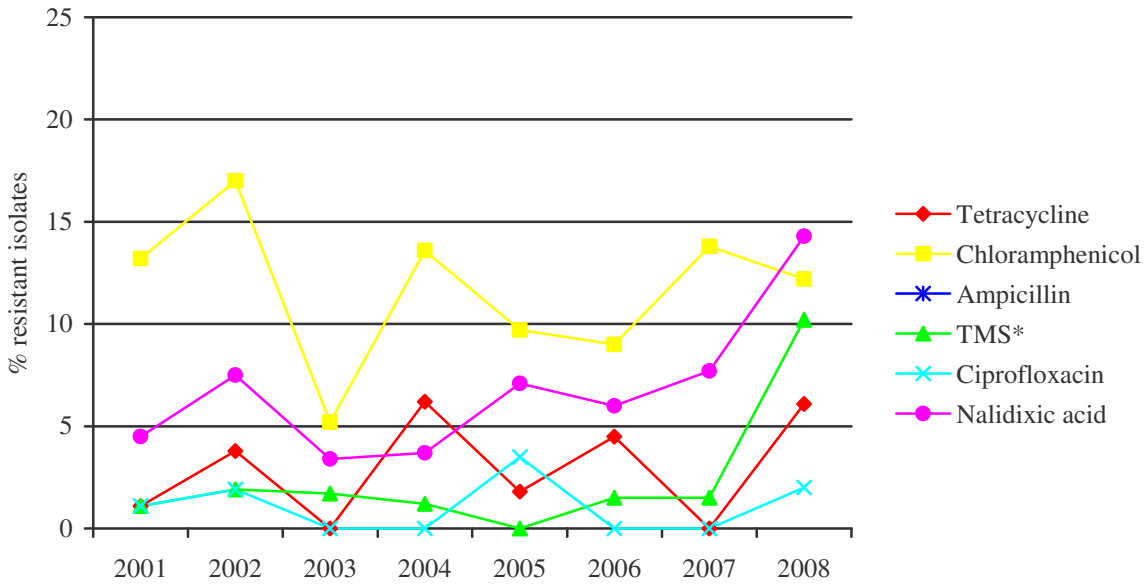

FIGURE 27. Prevalence of resistance to various antimicrobials in *Yersinia enterocolitica* from humans in Norway 2001-2008. *TMS=Trimethoprim-sulfamethoxazole.

RESULTS AND COMMENTS

The infections in 2008 were, as usual, mainly domestically acquired. All serogroup O:3 and O:9 isolates expressed intrinsic resistance to ampicillin. The prevalence of resistance to other antimicrobial agents has been fairly stable during the years 2001-2007, but in 2008 there was a tendency towards higher prevalences of resistance for all antimicrobial agents except chloramphenicol (Figure 27). None of the isolates were identified as ESBL producers in 2008.

Shigella spp. from human clinical specimens

It should be emphasized that almost all reported *Shigella* infections in Norway are acquired abroad. In 2008, 13 (9.5%) of the 134 reported cases were classified as domestically acquired. The majority of these were most probably secondary to imported cases. Thus, the prevalence of resistance presented in this report predominantly relates to isolates originating from other countries. The species distribution of the 129 *Shigella* isolates that were susceptibility tested was as follows: *S. sonnei* 59 (45.7%), *S. flexneri* 54 (41.9%), *S. boydii* 13 (10.1%), and *S. dysenteriae* 3 (2.3%). The results for *S. sonnei* and *S. flexneri* are presented in Table 25 and Figure 28 and in Table 26 and Figure 29, respectively.

TABLE 25. *Shigella sonnei* isolates from human clinical cases (n=59). Distribution (%) of antimicrobial susceptibility categories. Sampling, laboratory methods, and data handling are described in Appendix 4. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 0.5	> 8	0.0	91.5	8.5
Chloramphenicol	≤ 8	> 8	96.6	-	3.4
Tetracycline*			13.6	-	84.4
Nalidixic acid	≤ 16	> 16	69.5	-	30.5
Ciprofloxacin	≤ 0.5	> 1	100.0	0.0	0.0
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	3.4	1.7	94.9

* The epidemiological cut-off value defined by EUCAST is applied. There are no clinical breakpoints defiend by AFA as this substance is not relevant for treatment. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 26. *Shigella flexneri* isolates from human clinical cases (n=54). Distribution (%) of antimicrobial susceptibility categories. Sampling, laboratory methods, and data handling are described in Appendix 4. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 0.5	> 8	0.0	33.3	66.7
Chloramphenicol	≤ 8	> 8	38.9	-	61.1
Tetracycline*			20.4	-	79.6
Nalidixic acid	≤ 16	> 16	74.1	-	25.9
Ciprofloxacin	≤ 0.5	> 1	88.9	0.0	11.1
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	44.4	0.0	55.6

* The epidemiological cut-off value defined by EUCAST is applied. There are no clinical breakpoints defiend by AFA as this substance is not relevant for treatment. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

The resistance patterns for *S. sonnei* and *S. flexneri* have been fairly stable during the period 2001 – 2008. Resistance has been most prevalent among *S. flexneri* and least prevalent among *S. sonnei* isolates. Resistance in *S. flexneri* has been high for all the tested antimicrobial agents excluding quinolones, but from 2006 *S. flexneri* has shown a tendency towards higher prevalence of resistance also against these antibiotics. In *S. sonnei*, the prevalence of resistance has been particularly high for tetracycline and trimethoprim-sulfamethoxazole.

All the tested drugs are commonly used for various clinical purposes within human medicine in many parts of the world. The few isolates of *S. dysenteriae* (n=3) and *S. boydii* (n=13) recovered and susceptibility tested in 2008 indicate that multiresistance is also common in these species; three and ten of the isolates, respectively, were resistant to two or more antimicrobial agents. None of the isolates were susceptible to all antimicrobial agents included in the survey.

In 2008 three isolates (two *S. sonnei* and one *S. flexneri*) displayed reduced susceptibility to cefpodoxime (as a marker for possible ESBL-production) and all of them were verified as ESBL producers.

FIGURE 28. Prevalence of resistance to various antimicrobial agents in *Shigella sonnei* from humans in Norway 2001-2008. *TMS=Trimethoprim-sulfamethoxazole.

FIGURE 29. Prevalence of resistance to various antimicrobial agents in *Shigella flexneri* from humans in Norway 2001-2008. *TMS=Trimethoprim-sulfamethoxazole.

D. HUMAN CLINICAL ISOLATES

Gunnar Skov Simonsen, Karin Rønning, Ingvild Nordøy

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 a transition in the distribution from susceptible bacterial species to inherently more resistant ones will represent de facto emergence of resistance which is not easily detected. In order to complement the surveillance of individual species, NORM collects data on all positive blood cultures from the laboratory information systems of the participating institutions. A patient with a given microbial isolate was excluded from registration with a new isolate of the same species within a month from the first entry. This rule was applied irrespectively of changes in the organism´s susceptibility pattern. Isolates of a different species were included in the surveillance. It proved difficult to systematically evaluate the clinical significance of species which are commonly part of the normal skin flora. In Table 27, proportions are therefore estimated for all isolates and for all isolates excluding species considered to be common skin contaminants such as coagulase negative staphylococci, *Micrococcus* spp., *Corynebacterium* spp., *Bacillus* spp. and *Propioni-bacterium* spp. This does not imply that such isolates cannot cause infections, 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 findings although polymicrobial bloodstream infections are regularly detected in some patients. Limitations in the data extraction procedure prohibited in-depth analysis of these parameters.

TABLE 27. Number of blood culture isolates in 2008, proportion of all isolates, and proportion of isolates excluding possible skin contaminants (coagulase negative staphylococci, *Micrococcus* spp., *Bacillus* spp., *Corynebacterium* spp. and *Propionibacterium* spp.) 2004-2008. The table is based on data from the information systems of all laboratories in Norway except one in 2008.

Species	No. of isolates 2008	% of all isolates					% of isolates excluding skin flora				
		2004	2005	2006	2007	2008	2004	2005	2006	2007	2008
Staphylococcus aureus	1,245	12.3	10.3	10.3	10.1	10.6	14.0	13.3	13.7	13.3	13.9
Coagulase negative staphylococci	2,501	11.3	20.3	22.7	21.6	21.3	-	-	-	-	-
Streptococcus pneumoniae	778	11.6	9.4	7.9	7.8	6.6	13.2	12.1	10.6	10.2	8.7
Streptococcus pyogenes	150	2.3	2.2	1.3	1.1	1.3	2.6	2.8	1.7	1.5	1.7
Streptococcus agalactiae	183	2.0	1.6	1.7	1.7	1.6	2.3	2.1	2.2	2.2	2.0
Beta-haemolytic streptococci group C and G	167	0.7	0.8	1.2	0.9	1.4	0.9	1.1	1.5	1.1	1.9
Viridans- and non-haemolytic streptococci	454	4.6	3.8	3.7	3.7	3.9	5.3	5.0	5.0	4.8	5.1
Enterococcus faecalis	468	4.6	4.0	4.3	4.3	4.0	5.2	5.2	5.7	5.7	5.2
Enterococcus faecium	166	1.1	1.1	1.1	1.4	1.4	1.2	1.5	1.5	1.8	1.9
Other Gram positive aerobic bacteria	403	1.8	3.1	3.4	3.4	3.4	1.0	1.3	1.8	2.1	1.5
Escherichia coli	2,680	26.2	22.4	21.6	22.3	22.8	29.9	29.0	28.9	29.2	29.9
Klebsiella spp.	682	6.2	5.4	5.4	6.0	5.8	7.2	7.0	7.2	7.9	7.6
Enterobacter spp.	220	1.5	1.6	1.7	1.8	1.9	1.6	2.0	2.3	2.3	2.5
Proteus spp.	180	2.7	1.9	1.8	1.7	1.5	3.0	2.4	2.4	2.2	2.0
Other *Enterobacteriaceae*	247	1.8	1.8	1.9	2.2	2.1	2.0	2.3	2.5	2.9	2.8
Pseudomonas spp.	216	1.9	2.1	1.7	1.6	1.8	2.2	2.8	2.3	2.1	2.4
Other Gram negative aerobic bacteria	248	1.7	2.2	2.3	2.1	2.1	2.0	2.8	3.1	2.7	2.8
Bacteroides spp.	265	2.1	1.8	1.9	2.2	2.3	2.4	2.4	2.5	2.9	3.0
Other anaerobic bacteria	292	1.9	2.2	2.4	2.5	2.5	2.0	2.3	2.8	2.9	2.8
Yeasts	208	1.8	2.0	1.9	1.7	1.8	2.0	2.6	2.5	2.3	2.3
Total	**11,753**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**

As seen in Table 27 and Figure 30, aerobic Gram positive and Gram negative bacteria represented 54.5% and 38.1% of all isolates, respectively. The predominance of Gram positives among all isolates was at the same level as in previous years. The most common Gram positive species were coagulase negative staphylococci which represented 21.3% of all isolates. The difference between Gram positives and Gram negatives was reversed when species of the skin flora (coagulase negative staphylococci, *Micrococcus* spp., *Bacillus* spp., *Corynebacterium* spp. and *Propionibacterium* spp.) were excluded with 42.1% Gram positives and 50.0% Gram negatives.

Among the aerobic Gram positives, the prevalences of *S. pneumoniae* declined even when skin contaminants were excluded (13.2% in 2004, 12.1% in 2005, 10.6% in 2006, 10.2% in 2007 and 8.7% in 2008). The combined group of non-pneumococcal streptococci increased from 9.6% in 2007 to 10.7% in 2008. This was mainly due to an increase among group C and G streptococci to 1.9% in 2008, all figures excluding skin flora.

Among the aerobic Gram negatives, *E. coli* (29.9%) and other *Enterobacteriaceae* (14.9%) accounted for the vast majority of isolates. *Pseudomonas* spp. (2.4%) has been fairly stable after a peak in 2005 (2.8%), all figures excluding skin flora.

Anaerobic bacteria and yeasts were less prevalent. Anaerobes accounted for 4.8% (5.8% excluding skin flora) and yeasts accounted for 1.8% (2.3% excluding skin flora). The major pathogens among anaerobes were members of the *Bacteroides fragilis* group (2.3%/3.0%) and among yeasts *Candida albicans* (1.2%/1.6%). However, a multitude of other species was also represented.

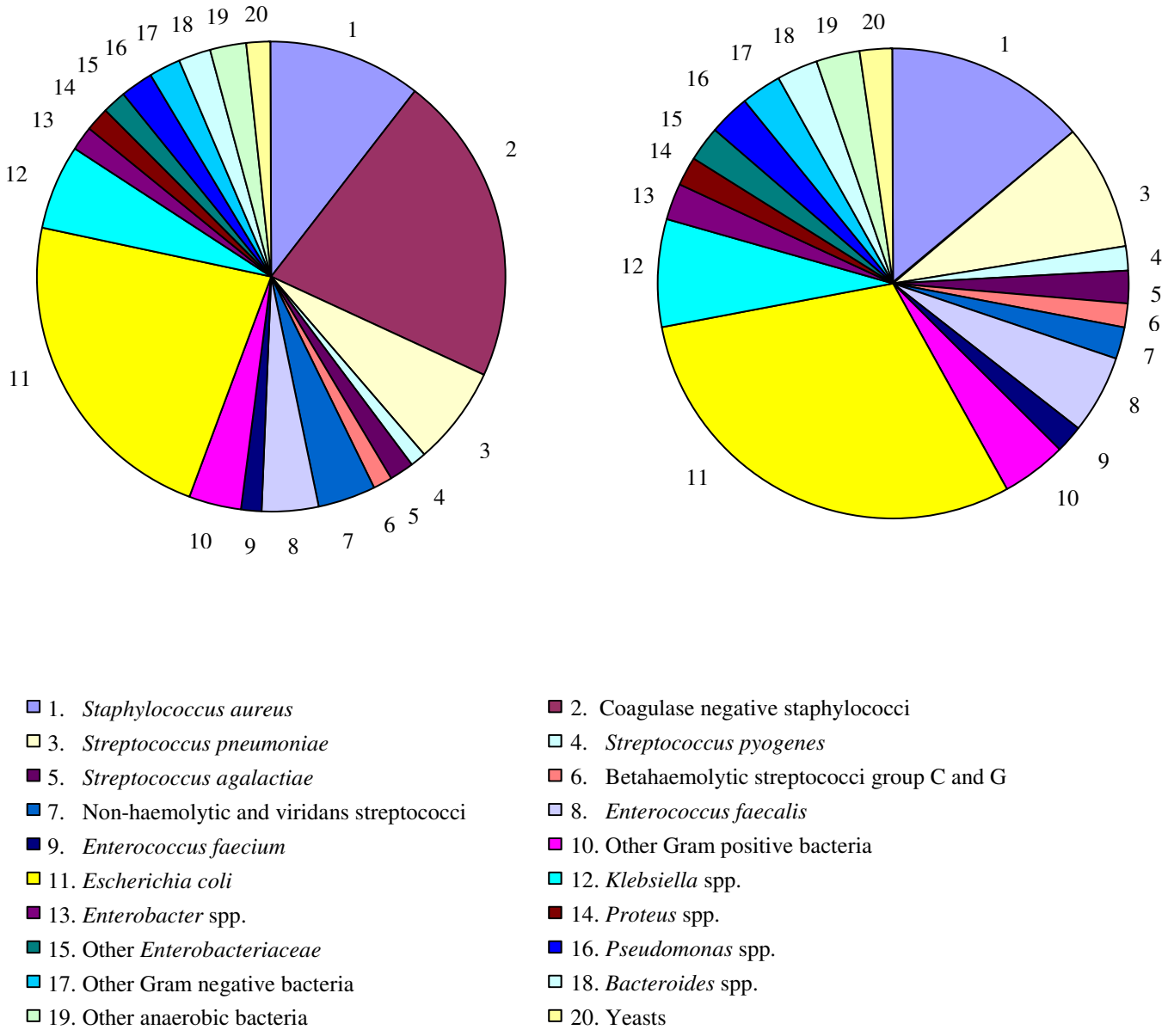

FIGURE 30. Distribution of all blood culture isolates (left, n=11,753) and blood culture isolates excluding common skin contaminants (right, n=8,949) such as coagulase negative staphylococci, *Micrococcus* spp., *Bacillus* spp., *Corynebacterium* spp. and *Propionibacterium* spp. The figure is based on data from the information systems of all Norwegian laboratories except one in 2008.

Escherichia coli in blood cultures

TABLE 28. *Escherichia coli* blood culture isolates (n=1,279). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin*	≤ 0.5	> 8	0.5	62.9	36.7
Piperacillin-tazobactam	≤ 8	> 16	95.9	2.2	1.9
Cefuroxime*	≤ 0.5	> 8	0.7	94.2	5.1
Cefotaxime	≤ 1	> 2	97.9	0.2	1.9
Ceftazidime	≤ 1	> 8	97.9	0.8	1.3
Ertapenem	≤ 0.5	> 1	99.0	0.3	0.7
Gentamicin	≤ 2	> 4	97.1	0.2	2.7
Nalidixic acid	≤ 16	> 16	87.8	-	12.2
Ciprofloxacin	≤ 0.5	> 1	91.9	0.5	7.6
Tigecycline	≤ 0.5	> 1	99.9	0.0	0.1
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	75.4	1.3	23.3
ESBL	Negative	Positive	98.5	-	1.5

*The wild type is defined as intermediately susceptible indicating that the drug is active at low dosage in urinary tract infections and at high dosage in systemic infections. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

The NORM results are interpreted according to the breakpoints of the Norwegian Working Group for Antibiotics (NWGA) at the time of analysis. The NWGA participates in the European breakpoint harmonization process, and the Norwegian breakpoints will therefore correspond to common EUCAST breakpoints when these have been established. The breakpoints for *Enterobacteriaecea* remained unchanged from 2007 to 2008 and are given in Table 28. In NORM 2008, cefpirome and aztreonam were replaced by tetracycline and tigecycline in order to provide data on resistance to the recently introduced glycylcycline. Similarly, meropenem was replaced by ertapenem which has also recently been marketed in Norway. The SIR distribution for tetracycline is not given as one of the disk suppliers has not defined breakpoints for this substance.

The vast majority of isolates remained fully susceptible to traditional broad-spectrum antimicrobial agents such as cefotaxime, ceftazidime, gentamicin and piperacillin-tazobactam (Table 28). The increase in gentamicin non-susceptibility noted from 2004 to 2007 was slightly reversed with 0.2% I (same as in 2007) and 2.7% R (3.7% in 2007) as seen in Figure 31. Further surveillance will be required to monitor aminoglycoside resistance in *E. coli*. The prevalence of non-suscpetibility to ciprofloxacin continued to increase from a total of 3.3% in 2004, 5.0% in 2005, 5.7% in 2006 and 7.1% in 2007, to 8.1% in 2008. The figures for 2006 and 2007 have been adjusted compared to previous reports due to changes in the interpretation of zone diameters from one of the disk manufacturers. The temporal association between ciprofloxacin non-susceptibility and ciprofloxacin usage is depicted in Figure 32. The prevalence of resistance to the indicator antibiotic nalidixic acid (12.2%) as well as ampicillin (36.7%) and trimethoprim-sulfamethoxazole (23.3%) remained unchanged from 2007.

Almost all isolates were fully susceptible to the new antibiotics tigecycline and ertapenem. Only a single isolate displayed reduced susceptibility to tigecycline. This isolate was resistant to tetracycline but remained susceptible to most other classes of antibiotics. Conversely, approximately 22% of all isolates had zone diameters below 10 mm for tetracycline. The surveillance programme thus confirmed that tetracycline resistance in *E. coli* does not predict resistance to glycylcyclines. A total of 13 isolates were non-susceptible to ertapenem, but the majority seemed to represent the lower end of the normal distribution. All ertapenem non-susceptible isolates will be further examined by molecular methods for the presence of metallo beta-lactamases and KPC enzymes.

In 2008, the detection of extended spectrum beta-lactamases (ESBL) was based on zone diameters of cefotaxime and ceftazidime. All isolates with reduced susceptibility to ceftazidime and/or cefotaxime were further characterized by combination Etests. A total of 19 isolates (1.5%) were reported as ESBL positive which is a slight increase from 1.2% in 2007. The 19 isolates originated from ten different hospitals, but two hospitals supplied four isolates each. The ESBL isolates were more resistant to cefotaxime (18/19 fully resistant) than to ceftazidime (9/18 fully resistant), and all were resistant to ampicillin and cefuroxime. Most isolates remained susceptible to piperacillin/tazobactam (14/19) and ertapenem (16/19), but many were co-resistant to ciprofloxacin (16/19), gentamicin (9/19) and trimethoprim-sulfamethoxazole (14/19). The ESBL isolates were molecularly characterized by PCR and DNA sequencing which revealed a predominance of CTX-M groups 9 (n=10) and 1 (n=8). A single isolate harboured a sequence belonging to the SHV family. The results are in accordance with previous surveys. It should be noted that classification of ESBLs solely on the basis of non-susceptibility to cefotaxime or ceftazidime would have overestimated the prevalence of ESBL by approximately 70% (2.6% instead of 1.5%).

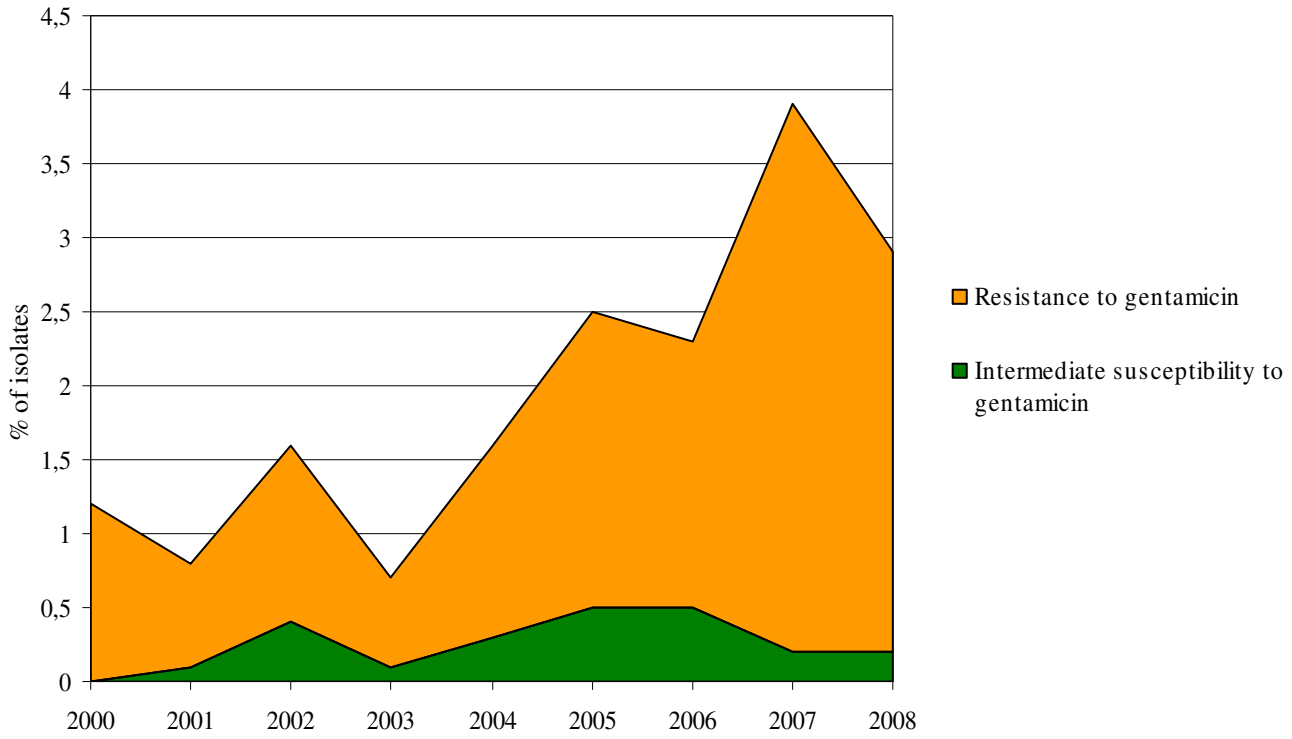

FIGURE 31. Prevalence of intermediate susceptibility and resistance to gentamicin in *Escherichia coli* blood culture isolates 2000-2008.

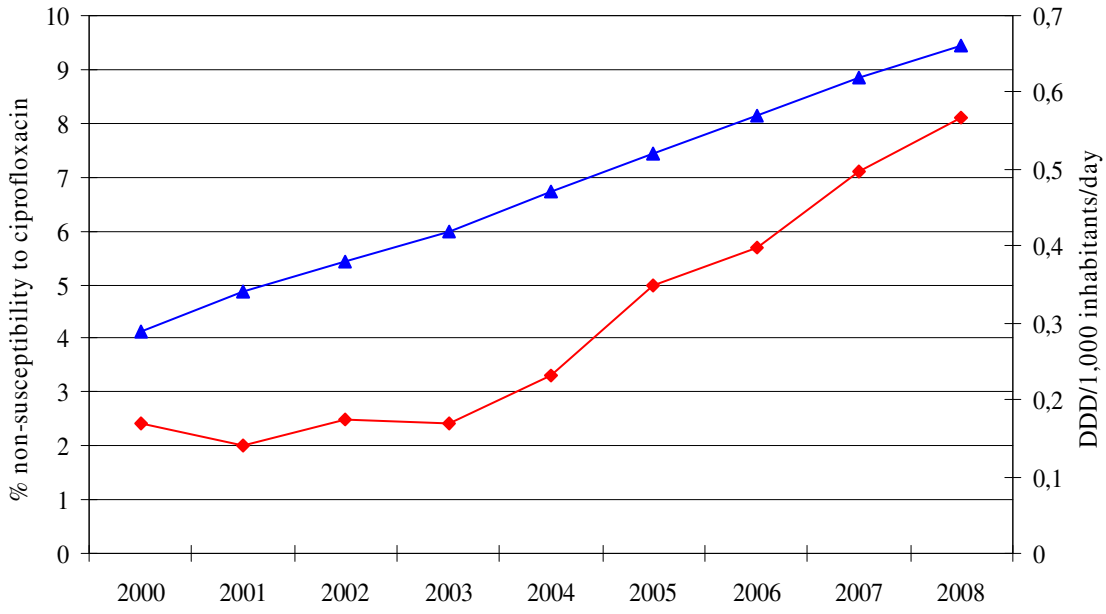

FIGURE 32. Prevalence of ciprofloxacin non-susceptibility in *Escherichia coli* blood culture isolates as defined by the 2008 breakpoint protocol (red) versus usage of ciprofloxacin (blue) 2000-2008. The figures for 2006 and 2007 have been adjusted compared to previous reports due to changed interpretation of zone diameters by one of the disk manufacturers.

Escherichia coli in urine

TABLE 29. *Escherichia coli* urinary tract isolates (n=1,165). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin*	≤ 0.5	> 8	0.7	71.2	28.1
Mecillinam	≤ 2	> 8	98.4	-	1.6
Cefuroxime*	≤ 0.5	> 8	0.7	96.8	2.5
Cefotaxime	≤ 1	> 2	98.9	0.1	1.0
Ceftazidime	≤ 1	> 8	99.2	0.2	0.6
Ertapenem	≤ 0.5	> 1	99.1	0.3	0.5
Gentamicin	≤ 2	> 4	98.2	0.3	1.5
Nalidixic acid	≤ 16	> 16	93.1	-	6.9
Ciprofloxacin	≤ 0.5	> 1	96.7	0.3	3.0
Nitrofurantoin	≤ 64	> 64	97.7	-	2.3
Trimethoprim	≤ 2	> 4	80.4	0.6	19.0
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	80.4	0.8	18.8
ESBL	Negative	Positive	99.3	-	0.7

*The wild type is defined as intermediately susceptible indicating that the drug is active at low dosage in urinary tract infections and at high dosage in systemic infections. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

Urinary tract isolates of *E. coli* have been included in the surveillance programme every year since NORM was established in 2000. The prevalences of resistance for 2008 are shown in Table 29 and the results 2000-2008 are shown in Figure 33. As for *E. coli* blood culture isolates, meropenem was replaced by ertapenem in order to generate baseline resistance data for this substance. The breakpoint for susceptibility to mecillinam was increased from S ≤ 2 mg/L to S ≤ 8 mg/L in 2009 thus eliminating the intermediate category. The mecillinam curve in figure 33 has been adjusted for this change through the whole time period. The breakpoints for susceptibility and resistance for nitrofurantoin were increased from S ≤ 32 mg/L / R > 32 mg/L to S ≤ 64 mg/L / R > 64 mg/L in 2009, but this change has not had any effect on the SIR distribution for nitrofurantoin.

The resistance rates among urinary tract isolates have remained remarkably stable over the last eight years. Approximately 30% of *E. coli* isolates were resistant to ampicillin, while the remaining 70% belong to the wild type, which in Norway is categorized as intermediately susceptible. Close to 20% of *E. coli* isolates are resistant to trimethoprim and trimethoprim-sulfamethoxazole. According to the 2009 breakpoints, resistance to mecillinam has remained below 2% over the last four years with 1.6% resistance in 2008.

Ciprofloxacin is used as a second-line agent for urinary tract infections in Norway. The prevalence of non-susceptibility has been relatively stable and was 3.3% in 2008 (0.3% intermediately susceptible and 3.0% resistant) according to the adjusted interpretation of zone diameters. The corresponding rates for blood culture isolates were 0.5% intermediate susceptibility and 7.6% resistance. The same difference was seen for nalidixic acid with 6.9% resistance in urinary tract isolates and 12.2% resistance in bloodstream infections. One may speculate that systemic infections are caused by selected pathogenic lineages with increased virulence and accumulation of mutations in gyrase and topoisomerase genes, whereas urinary tract isolates are more representative of the wild-type normal flora.

In total, eight isolates (0.7%) were confirmed as ESBL producers. This prevalence is unchanged from 2007 (0.8%). The majority of ESBL strains was non-susceptible to both cefotaxime (7/8) and ceftazidime (5/8) and displayed cross resistance to nalidixic acid (8/8), ciprofloxacin (6/8), trimethoprim (6/8) and trimethoprim-sulfamethoxazole (6/8). Most of them remained susceptible to ertapenem (7/8), nitrofurantoin (6/8) and mecillinam (7/8), although the clinical significance of in vitro susceptibility to mecillinam in ESBL producing *E. coli* is unclear. By molecular characterization it was determined that the eight ESBL isolates harboured CTX-M group 1 (n=4), CTX-M group 9 (n=3) or SHV (n=1) determinants. This is in accordance with findings in blood culture isolates and previous surveys.

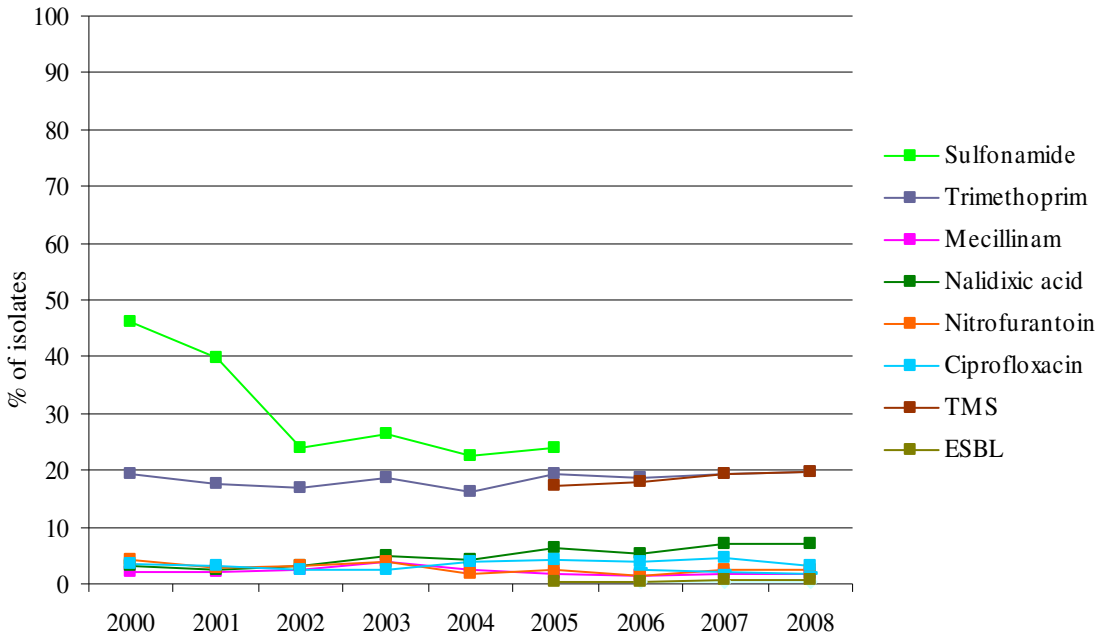

FIGURE 33. Prevalences of non-susceptibility to various antimicrobial agents in urinary tract *E. coli* isolates 2000-2008. The breakpoint for susceptibility to mecillinam was increased from S ≤ 2 mg/L to S ≤ 8 mg/L in 2009. The mecillinam data from 2000 onwards have been recalculated using the 2009 breakpoints. The breakpoints for susceptibility and resistance for nitrofurantoin were increased from S ≤ 32 mg/L and R > 32 mg/L to S ≤ 64 mg/L and R > 64 mg/L in 2009.

Klebsiella spp. in blood cultures

TABLE 30. *Klebsiella* spp. blood culture isolates (n=505). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Piperacillin-tazobactam	≤ 8	> 16	92.7	3.2	4.2
Cefuroxime*	≤ 0.5	> 8	0.4	94.1	5.5
Cefotaxime	≤ 1	> 2	98.8	0.2	1.0
Ceftazidime	≤ 1	> 8	96.2	1.2	2.6
Ertapenem	≤ 0.5	> 1	94.7	3.8	1.6
Gentamicin	≤ 2	> 4	99.0	0.4	0.6
Nalidixic acid	≤ 16	> 16	86.9	-	13.1
Ciprofloxacin	≤ 0.5	> 1	96.0	0.8	3.2
Tigecycline	≤ 1	> 2	90.7	1.4	7.9
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	86.1	3.0	10.9
ESBL	Negative	Positive	98.0	-	2.0

*The wild type is defined as intermediately susceptible indicating that the drug is active at low dosage in urinary tract infections and at high dosage in systemic infections. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 31. *Klebsiella pneumoniae* blood culture isolates (n=396). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Piperacillin-tazobactam	≤ 8	> 16	93.4	3.5	3.0
Cefuroxime*	≤ 0.5	> 8	0.5	94.4	5.1
Cefotaxime	≤ 1	> 2	97.5	0.0	2.5
Ceftazidime	≤ 1	> 8	96.7	1.8	1.5
Ertapenem	≤ 0.5	> 1	93.7	4.5	1.8
Gentamicin	≤ 2	> 4	99.2	0.3	0.5
Nalidixic acid	≤ 16	> 16	84.8	-	15.2
Ciprofloxacin	≤ 0.5	> 1	95.2	1.0	3.8
Tigecycline	≤ 1	> 2	88.6	1.8	9.6
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	83.6	3.3	13.1
ESBL	Negative	Positive	98.0	-	2.0

*The wild type is defined as intermediately susceptible indicating that the drug is active at low dosage in urinary tract infections and at high dosage in systemic infections. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 32. *Klebsiella oxytoca* blood culture isolates (n=93). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Piperacillin-tazobactam	≤ 8	> 16	90.3	2.2	7.5
Cefuroxime*	≤ 0.5	> 8	0.0	92.5	7.5
Cefotaxime	≤ 1	> 2	93.5	4.3	2.2
Ceftazidime	≤ 1	> 8	98.9	1.1	0.0
Ertapenem	≤ 0.5	> 1	100.0	0.0	0.0
Gentamicin	≤ 2	> 4	98.9	0.0	1.1
Nalidixic acid	≤ 16	> 16	94.6	-	5.4
Ciprofloxacin	≤ 0.5	> 1	100.0	0.0	0.0
Tigecycline	≤ 1	> 2	98.9	0.0	1.1
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	96.8	1.1	2.2
ESBL	Negative	Positive	98.9	-	1.1

*The wild type is defined as intermediately susceptible indicating that the drug is active at low dosage in urinary tract infections and at high dosage in systemic infections. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

The surveillance of *Klebsiella* spp. in blood cultures included 396 *K. pneumoniae* (78.4%), 93 *K. oxytoca* (18.4%) and 16 (3.2%) isolates not identified to the species level, giving a total of 505 *Klebsiella* spp. isolates (Tables 30-32). The species distribution was not significantly changed from 2007, although the proportion of isolates not identified to the species level was reduced from 9.7% to 3.2%. The breakpoints for the antimicrobial agents included in the *Klebsiella* surveillance protocol were not changed in 2009. However, one disk supplier adjusted the interpretation of zone diameters for ciprofloxacin, and the data for 2006 and 2007 have therefore been recalculated for comparison with 2008. The SIR distribution for cefpirome is not given as one of the disk suppliers has not defined breakpoints for this agent.

The vast majority of *Klebsiella* spp. isolates was fully susceptible to cefotaxime (98.8%), ceftazidime (96.2%) and the beta-lactam/beta-lactamase inhibitor combination piperacillin-tazobactam (92.7%, Figure 34). Nevertheless, the rates of non-susceptibility increased from 1.8% to 3.8% for ceftazidime and from 3.4% to 7.4% for piperacillin-tazobactam. As for *E. coli*, the detection of extended spectrum beta-lactamases (ESBL) was based on zone diameters of cefotaxime and ceftazidime disks. Isolates with reduced zone diameters were further characterized by combination Etests, and suspected isolates were confirmed by molecular characterization of resistance determinants. A total of 10 isolates (2.0%) were reported as ESBL producers from the laboratories, of which eight were *K. pneumoniae*, one was *K. oxytoca* and one was unspeciated. Most of the ESBL isolates were non-

susceptible to cefotaxime (8/10), ceftazidime (9/10) and piperacillin/tazobactam (7/10), and many displayed cross-resistance to ciprofloxacin (5/10), trimethoprim-sulfamethoxazole (5/10) or gentamicin (2/10). Molecular characterization at the Reference Centre for Detection of Antimicrobial Resistance (K-Res) confirmed the presence of CTX-M group 1 (n=1), broad spectrum SHV (n=2), or a combination of broad spectrum SHV and TEM (n=1) determinants in four of the isolates. Two isolates contained SHV-36 which is alternatively classified as either a broad-spectrum or a narrow-spectrum beta-lactamase. Three isolates were phenotypically characterized as *K. pneumoniae* hyperproducing the narrow-spectrum beta-lactamase SHV-11 (n=2) or *K. oxytoca* hyperproducing the chromosomally encoded K1 beta-lactamase. The last isolate was a *K. pneumoniae* isolate phenotypically sensitive to cephalosporins. The overall prevalence of ESBL was not adjusted in spite of the molecular data as only a very limited number of isolates were subjected to these analyses.

The carbapenem meropenem was replaced by ertapenem in order to generate baseline resistance data for this substance. The 6.3% non-susceptibility rate (4.5% reduced susceptibility and 1.8% resistance) in *K. pneumoniae* originated from users of one disk system and may result from intersection of the normal distribution as seen in the supplement at www.antibiotikaresistens.no.

The prevalence of intermediate susceptibility and resistance to gentamicin increased from 0.0% to 0.4% and from 0.2% to 0.6% between 2007 and 2008, respectively. Further surveillance is needed to determine whether this indicates an emergence of aminoglycoside resistance in *Klebsiella* spp. in Norway. The overall prevalence of resistance to ciprofloxacin has been stable between 3-4% when taking into account the changes in breakpoints and interpretive rules. Non-susceptibility to trimethoprim-sulfamethoxazole is apparently increasing, but the change from 13.6% in 2007 to 13.9% in 2008 was minor compared to previous years. There was a significant difference in the prevalence of non-susceptibility to ciprofloxacin and trimethoprim-sulfamethoxazole between *K. pneumoniae* and *K. oxytoca*. No isolates of *K. oxytoca* were non-susceptible to ciprofloxacin, and only 3.3% were non-susceptible to trimethoprim-sulfamethoxazole compared to 4.8% and 16.4% for *K. pneumoniae,* respectively. Conversely, the prevalence of non-susceptibility to cefotaxime and piperacillin-tazobactam was higher in *K. oxytoca*, presumably due to the chromosomal K1 beta-lactamase in this species.

Tetracycline and tigecycline were included in the surveillance protocol for the first time in 2008. The 9.3% non-susceptibility to tigecycline was caused by a high prevalence of resistance (14.6%) to this substance in *K. pneumoniae* among laboratories using one of the disk systems. Most of the tigecycline resistant isolates were fully susceptible to tetracycline, and the distribution of zone diameters clearly indicates that the breakpoint is above the lower end of the normal distribution (see www.antibiotikaresistens.no).

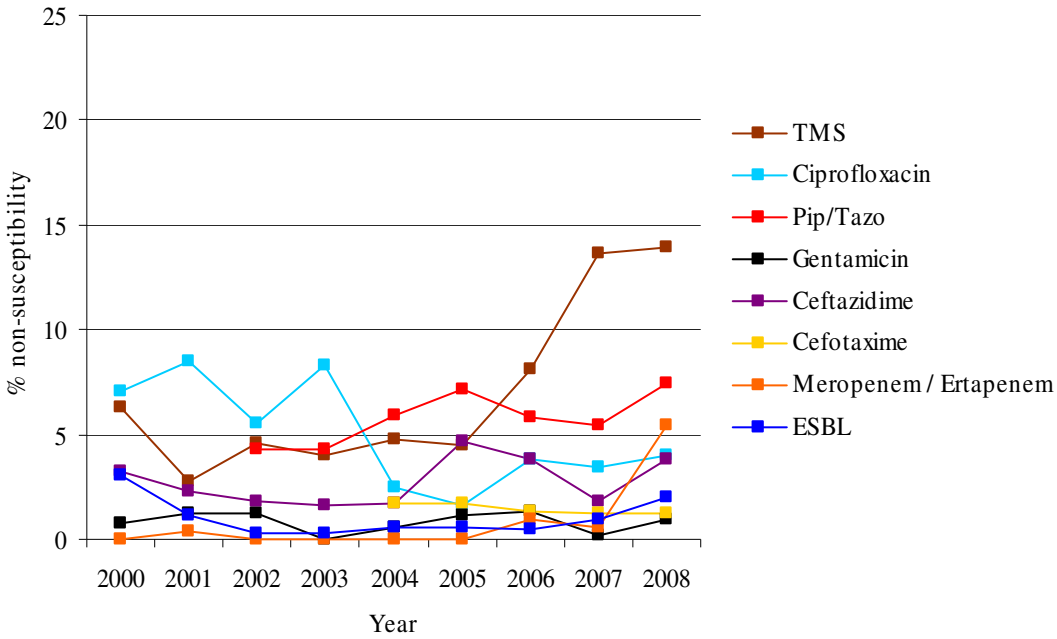

FIGURE 34. Prevalence of non-susceptibility to various antimicrobial agents in *Klebsiella* spp. blood culture isolates 2000-2008. The figures for ciprofloxacin for 2006 and 2007 have been adjusted compared to previous reports due to adjustment of zone diameter interpretations by one of the disk manufacturers. Meropenem was replaced by ertapenem in 2008.

Enterobacter cloacae in blood cultures

TABLE 33. *Enterobacter cloacae* blood culture isolates (n=264). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Piperacillin-tazobactam	≤ 8	> 16	79.8	5.7	14.8
Cefuroxime*	≤ 0.5	> 8	0.0	59.8	40.2
Cefotaxime	≤ 1	> 2	71.6	1.1	27.3
Ceftazidime	≤ 1	> 8	73.1	3.4	23.5
Ertapenem	≤ 0.5	> 1	64.8	5.7	29.5
Gentamicin	≤ 2	> 4	100.0	0.0	0.0
Nalidixic acid	≤ 16	> 16	89.0	-	11.0
Ciprofloxacin	≤ 0.5	> 1	97.0	0.3	2.7
Tigecycline	≤ 1	> 2	92.8	0.8	6.4
Trimethoprim-sulfamethoxazole**	≤ 2	> 4	91.7	2.3	6.1

*The wild type is defined as intermediately susceptible indicating that the drug is active at low dosage in urinary tract infections and at high dosage in systemic infections. **Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

Enterobacter cloacae blood culture isolates were included in the NORM surveillance programme for the first time in 2008. All blood culture isolates from 2006, 2007 and the first nine months of 2008 were included. The present report is the first large-scale survey of antimicrobial resistance in *E. cloacae* in Norway.

E. cloacae wild-type strains contain a chromosomal AmpC beta-lactamase which is negatively regulated by the repressor AmpR. This mechanism is liable to escape mutants leading to high-level resistance to all penicillins and cephalosporins except cefpirome and cefepime. The spectrum of resistance may be expanded to include 4th generation cephalosporins as well as carbapenems when derepressed AmpC is combined with porin loss. AmpC strains are generally not susceptible to beta-lactam / beta-lactamase inhibitor combinations.
Cephalosporins may be used in the treatment of systemic infections with susceptible *E. cloacae* strains, but derepressed AmpC mutants may arise during therapy and monotherapy is therefore not advisable. One recent study suggests that combination therapy with aminoglycosides may increase the rate of transition to derepression, and EUCAST has therefore recommended combinations with fluoroquinolones instead of aminoglycosides.

A primary objective of the *E. cloacae* surveillance protocol was to determine the prevalence of stable AmpC derepression in Norway. As seen in Table 33, 25-30% of all isolates were resistant to cefotaxime and ceftazidime. This is in accordance with international studies reporting 20-25% stably derepressed isolates in unselected materials. It is surprising that almost 30% of isolates were resistant to the carbapenem ertapenem, but this may be due to intersection of the normal distribution by the general *Enterobacteriaceae* breakpoints. As seen at www.antibiotikaresistens.no, all isolates had ertapenem zone diameters above 10 mm. Alternatively, additional resistance mechanisms may be present in these isolates. Further characterization by molecular methods is required to draw any definite conclusions.

Notably, all isolates were susceptible to gentamicin, and the majority was also susceptible to ciprofloxacin (97.0%) and trimethoprim-sulfamethoxazole (91.7%). Some isolates displayed high-level resistance to nalidixic acid thus indicating first-step quinolone mutations. However, both nalidixic acid and tigecycline resistance rates may be overestimated due to breakpoints interfering with the wild-type distribution of this species.

Moraxella catarrhalis in respiratory tract specimens

TABLE 34. *Moraxella catarrhalis* respiratory tract isolates (n=175). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Erythromycin	≤ 0.25	> 0.5	94.9	1.7	3.4
Tetracycline	≤ 1	> 2	99.4	0.0	0.6
Ciprofloxacin	≤ 0.5	> 0.5	98.3	-	1.7
Beta-lactamase	Negative	Positive	8.0	-	92.0

RESULTS AND COMMENTS

Moraxella catarrhalis was previously surveyed in NORM in 2003 when 287 respiratory tract isolates were included in the programme. A comparison was at that time made between MIC determination by Etest and zone diameters by disk diffusion. The results underlined the technical difficulties with disk diffusion for beta-lactam antibiotics in this species. In the present survey, beta-lactam susceptibility was only examined by beta-lactamase testing. Clinically relevant non-beta-lactam antimicrobial agents were examined when both disk diffusion systems could provide zone diameter breakpoints. The results are presented in Table 34 and Figure 35.

The prevalence of beta-lactamase production was relatively stable from 2003 (9.7%) to 2008 (8.0%). Internationally, beta-lactamase production in *M. catarrhalis* has been reported to be encoded by the predominant high-efficiency BroI beta-lactamase and the less prevalent low-efficiency BroII enzyme. There are no published reports on molecular characterization of Norwegian *M. catarrhalis* isolates.

The breakpoints for erythromycin and ciprofloxacin have been adjusted since 2003. Erythromycin breakpoints have been reduced from S ≤ 1 / R > 2 to S ≤ 0.25 / R > 0.5, whereas ciprofloxacin breakpoints have been adjusted from S ≤ 0.125 / R > 2 to S ≤ 0.5 / R > 0.5. Tetracycline breakpoints have remained unchanged at S ≤ 1 / R > 2. The 2003 results were recorded as MIC values, and the SIR distributions could thus easily be recalculated using the 2009 protocol. As seen in Figure 36, the rate of non-suseptibility to erythromycin has decreased from 8.3% to 5.1%, while the rate of resistance to ciprofloxacin has increased from 0% to 1.7%. Tetracycline resistance has remained unchanged at 0.6% of isolates.

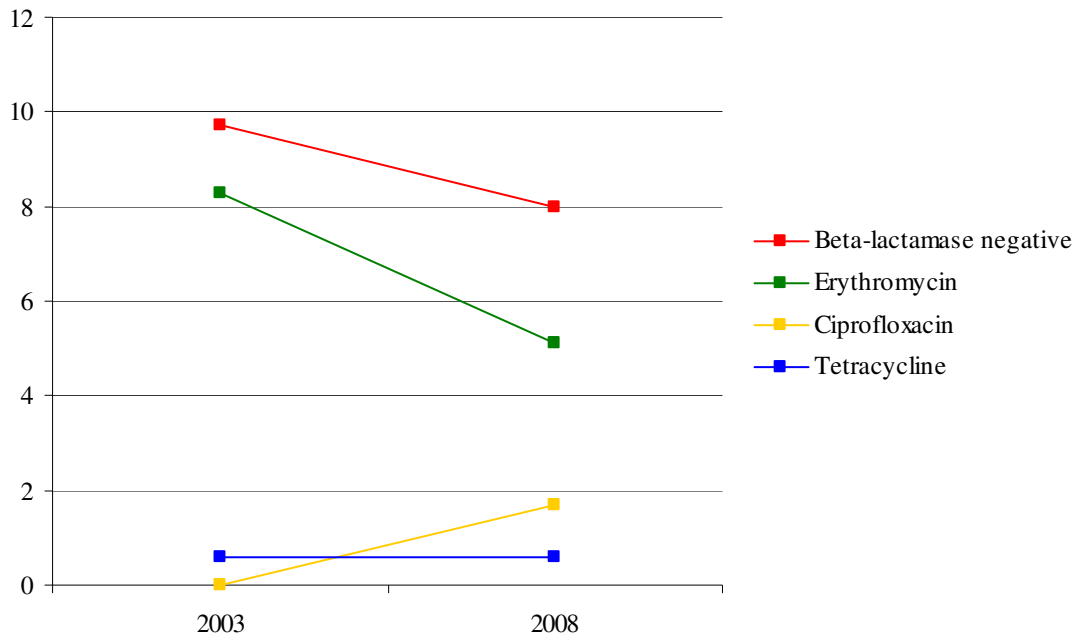

FIGURE 35. Prevalence of non-susceptibility to erythromycin, ciprofloxacin and tetracycline and prevalence of beta-lactamase negativity in respiratory tract isolates of *M. catarrhalis* in 2003 and 2008. All prevalences were calculated according to the breakpoint protocol of 2009.

Antibiotic resistance in *Neisseria meningitidis*

Apart from resistance to sulfonamides, which developed already in the 1960s and is currently present in more than 25% of patient isolates, *Neisseria meningitidis* has remained essentially susceptible to the antimicrobial agents traditionally used for treatment and prophylaxis. This is quite remarkable considering the high rate of asymptomatic meningococcal carriage and, thus, the massive exposure of the species to antimicrobial agents used in the community for other indications. In spite of this general susceptibility to currently used agents, close monitoring of the changes in the degree of susceptibility is essential, as early treatment of meningococcal disease with antimicrobial agents is crucial for keeping the case fatality rate and the risk of sequelae as low as possible. In the present note, recent developments in antimicrobial resistance of *N. meningitidis* to the main antimicrobial agents used for treatment and chemoprophylaxis are briefly described.

Penicillin

In contrast to the high proportion of beta-lactamase-producing strains of its close relative, the gonococcus, *N. meningitidis* isolates with high minimal inhibitory concentrations (MICs) for penicillin due to the production of beta-lactamase are very rare and their recovery rather anecdotal (1,2). This seems to be due to the poor capability of *N. meningitidis* to harbor plasmids in a stable manner.

On the other hand, strains with reduced penicillin susceptibility (MIC > 0.06 to < 1 mg/L) have been described since the mid 1980s (3). This reduced susceptibility has been mainly attributed to alterations in the structure of penicillin-binding proteins (PBPs), especially PBP2, which is encoded by the *penA* gene. Gradually, the frequency of such isolates has increased in many countries of the world, representing more than 30% of the isolates in Southern Europe (4). Strains with reduced penicillin susceptibility have been nearly absent in patients from Norway until a few years ago. Recently, a slight increase has been noticed, with 2 to 4 cases of meningococcal disease every year caused by isolates with reduced penicillin susceptibility. Whether infection with a strain with reduced penicillin susceptibility has an impact on failure of treatment with penicillin and disease outcome, has not yet been established. Several PCR strategies have been developed to rapidly detect altered *penA* sequences. However, the existence of mechanisms other than PBP2 alterations complicates the detection of intermediate resistance using molecular tools (5).

Chloramphenicol

Chloramphenicol is not used for treatment of meningococcal disease in developed countries, but in Africa, the general recommendation for treatment during epidemics is a single dose of chloramphenicol in oil. High level of resistance to chloramphenicol was first reported by Galimand in isolates from Vietnam and France (6), and later was also identified in Australia (7). Resistance is determined by the presence of the *catP* gene, encoding the enzyme chloramphenicol acetyltransferase. Serogroup A isolates from Africa referred to the WHO Collaborative Centers for Reference and Research on Meningococci in Atlanta (USA), Marseilles (France) and Oslo (Norway) are systematically tested for susceptibility to chloramphenicol. So far, no chloramphenicol resistance has been evidenced in Africa.

Ceftriaxone

Ceftriaxone has been proposed as an alternative to oily chloramphenicol for treatment of epidemic meningitis in sub-Saharan Africa (8). The emergence of non-susceptibility to ceftriaxone reported from India in 2006 (9) was therefore worrisome. The data were, however, quite extraordinary and have been questioned (10).

Rifampicin

In the Western world, chemoprophylaxis of persons who have been in contact with a case is often recommended, and rifampicin is the most commonly used antimicrobial agent in adults. High level resistance to rifampicin is provoked by point mutations in the *rpoB* gene encoding for the beta-subunit of the RNA polymerase. In *N. meningitidis*, resistance to rifampicin is only occasionally detected, usually in secondary cases, after prophylactic treatment of a patient's near contacts. While occurrence of rifampicin resistant meningococci after prophylaxis has been reported many times, the frequency of such isolates remains very low (11). Mutations in *rpoB* appear to represent a major biological cost for the bacteria. Thus, with lower biological fitness, these isolates are unable to clonally expand (12).

Ciprofloxacin

The quinolone group, with ciprofloxacin and ofloxacin, is an alternative in prophylaxis, particularly for infants and older individuals. There have been several reports, since the beginning of the 1990s, of isolates with reduced susceptibility to ciprofloxacin (MIC > 0.06 mg/L). More recently, outbreaks of ciprofloxacin-resistant meningococci (MIC > 0.12 mg/L) have been identified, associated with a serogroup A strain in India (13) and with a serogroup B strain in the United States (14). Alterations of the *gyrA* gene, as identified among these ciprofloxacin resistant isolates, do not seem to significantly affect the fitness of the strains, as apparently these can be maintained in the carrier population for several months (13,14).

In conclusion, while the situation regarding antimicrobial resistance of the meningococcus is still highly favourable, careful monitoring of the recent and future changes are essential. In Norway, except for a few isolates with intermediate susceptibility to penicillin, no sign of resistance towards any of the drugs tested has so far been evidenced among systemic isolates.

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Staphylococcus aureus in blood cultures

TABLE 35. *Staphylococcus aureus* blood culture isolates (n=871). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Erythromycin	≤ 1	> 2	95.4	0.2	4.4
Clindamycin	≤ 0.25	> 0.5	97.4	0.1	2.5
Fusidic acid	≤ 1	> 1	95.4	-	4.6
Ciprofloxacin	≤ 1	> 1	97.2	-	2.8
Gentamicin	≤ 1	> 1	98.6	-	1.4
Linezolid	≤ 4	> 4	100.0	-	0.0
Rifampicin	≤ 0.06	> 0.5	99.7	0.2	0.1
Tetracycline	≤ 1	> 2	96.2	0.1	3.7
Trimethoprim-sulfamethoxazole*	≤ 2	> 4	99.3	0.2	0.5
Beta-lactamase	Negative	Positive	28.4	-	71.5
Cefoxitin screen	Negative	Positive	98.6	-	1.4
MRSA (*mecA*)	Negative	Positive	99.3	-	0.7
Vancomycin screen	Negative	Positive	100.0	-	0.0

*Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

Six methicillin resistant *S. aureus* (MRSA) isolates were detected in the NORM surveillance system in 2008 (Table 35) corresponding to a prevalence of 0.7%. This is a slight increase from 0.2% in 2007 and should be carefully monitored. The resistance phenotype was confirmed by *mecA* PCR in all cases.

Laboratory screening for MRSA in NORM is performed using cefoxitin disks. All MRSA isolates had cefoxitin zone diameters below the screening breakpoint. Cross resistance was detected towards ciprofloxacin (4/6), gentamicin (2/6) and fusidic acid (1/6), and two isolates were constitutively resistant to erythromycin and clindamycin. All MRSA isolates were fully susceptible to linezolid, rifampicin and trimethoprim-sulfamethoxazole. Six methicillin susceptible *S. aureus* (MSSA) isolates (0.7%) displayed reduced cefoxitin zone diameters but were not confirmed as MRSA by genotypic analysis. All these isolates had cefoxitin zone diameters within 2 millimeters of the screening breakpoint.

The findings are in accordance with reports from the databases of the participating laboratories where 10 out of 1,359 (0.7%) *S. aureus* blood culture isolates were MRSA. None of the six *S. aureus* isolates recovered from cerebrospinal fluids were methicillin resistant, thus bringing the total number of systemic MRSA isolates to 10/1,365 (0.7%). The Norwegian Surveillance System for Communicable Diseases (MSIS) reported a stable number of MRSA infections in Norway with 342 cases in 2007 and 348 in 2008. However, the cases reported to MSIS are predominantly skin and soft tissue infections (n=306) and colonizations (n=304). The proportion of MRSA among systemic (0.7%) and localized (0.7%) *S. aureus* infections was comparable in 2008. A total of 652 cases of MRSA infections and colonizations were reported to MSIS in 2008. The 10% increase from 594 cases in 2007 was due to a 20% increase in the number of reported MRSA colonizations from 252 cases in 2007 to 304 in 2008. Further information about MRSA cases in MSIS is presented on page 64.

A total of 40 isolates (4.6%) were non-susceptible to erythromycin. This is a minor increase from 2007 (4.0%) and 2006 (3.3%). The macrolide resistance phenotypes were determined by the double disk diffusion (DDD) test. Twelve (30%) were constitutively MLS_B resistant, 19 (47.5%) were inducibly MLS_B resistant and nine (22.5%) displayed efflux mediated M type resistance. These figures represent 1.4%, 2.2% and 1.0% of all *S. aureus* isolates from blood cultures, respectively. The distribution of macrolide resistance phenotypes is similar to the results from previous years.

The prevalence of resistance to fusidic acid remained unchanged in 2008 (4.6%) compared to 2007 (4.2%). This may indicate that the epidemiology of fusidic resistant *S. aureus* has now stabilized in Norway. There were no significant changes for ciprofloxacin, gentamicin, rifampicin or trimethoprim-sulfamethoxazole. No isolates displayed growth on the vancomycin agar screen, and all isolates were fully susceptible to linezolid. Figure 36 shows the prevalences of non-susceptibility to various antimicrobials.

A total of 71.5% of the isolates were beta-lactamase positive which is unchanged from previous years. A subgroup analysis revealed that beta-lactamase positive isolates were more often resistant to ciprofloxacin (3.2% vs 1.6%), fusidic acid (5.0% vs 3.6%), erythromycin (5.2% vs 3.2%), clindamycin (3.4% vs 0.8%), gentamicin (1.9% vs 0.0%) and tetracycline (4.4% vs 2.4%) than beta-lactamase negative isolates.

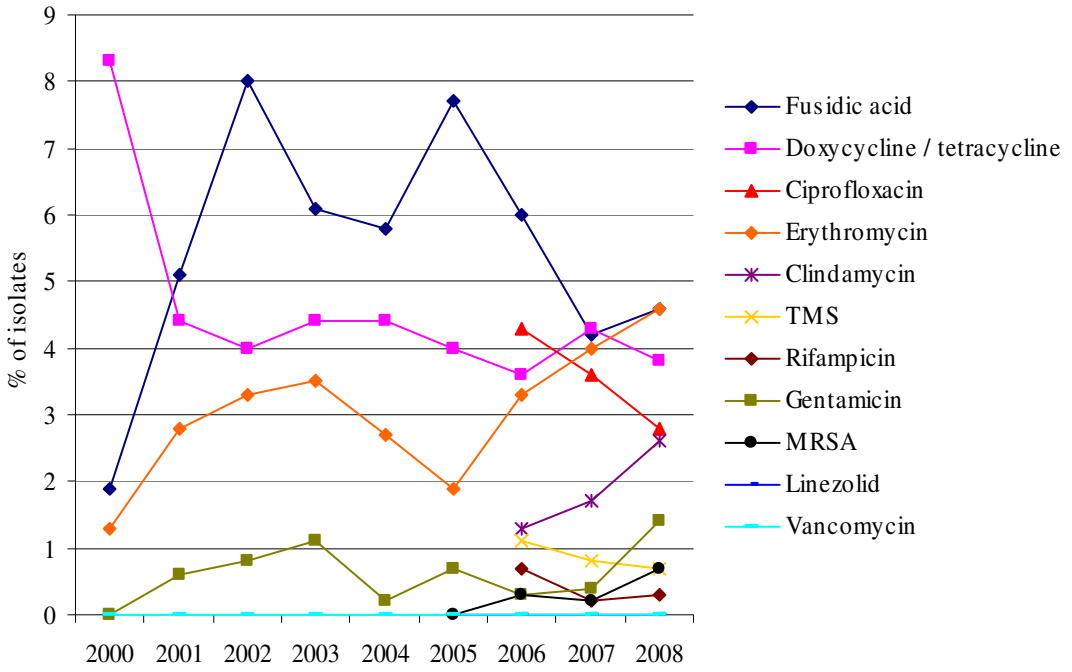

FIGURE 36. Prevalences of non-susceptibility to various antimicrobial agents among *Staphylococcus aureus* blood culture isolates 2000-2008. The breakpoint for susceptibility to gentamicin was decreased from S ≤ 2 mg/L to S ≤ 1 mg/L in 2006. Doxycycline was replaced by tetracycline in 2006. The breakpoints for clindamycin were reduced from S ≤ 1 mg/L and R > 2 mg/L to S ≤ 0.25 mg/L and R > 0.5 mg/L in 2007. The breakpoints for fusidic acid were increased from S ≤ 0.5 mg/L and R > 0.5 mg/L to S ≤ 1 mg/L and R > 1 mg/L in 2009. The breakpoint for resistance to trimethoprim-sulfamethoxazole was decreased from R > 8 mg/L to R > 4 mg/L in 2009. The breakpoints for rifampicin were decreased from S ≤ 1 mg/L and R > 1 mg/L to S ≤ 0.06 mg/L and R > 0.5 mg/L in 2009.

Staphylococcus aureus in wound specimens

TABLE 36. *Staphylococcus aureus* isolates from wound specimens (n=1,061). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Erythromycin	≤ 1	> 2	95.0	0.3	4.7
Clindamycin	≤ 0.25	> 0.5	98.1	0.3	1.6
Fusidic acid	≤ 1	> 1	89.7	-	10.3
Ciprofloxacin	≤ 1	> 1	97.1	-	2.9
Gentamicin	≤ 1	> 1	99.7	-	0.3
Linezolid	≤ 4	> 4	100.0	-	0.0
Rifampicin	≤ 0.06	> 0.5	99.9	0.0	0.1
Tetracycline	≤ 1	> 2	94.0	0.2	5.8
Trimethoprim-sulfamethoxazole*	≤ 2	> 4	99.0	0.4	0.7
Beta-lactamase	Negative	Positive	26.2	-	73.8
Cefoxitin screen	Negative	Positive	99.1	-	0.9
MRSA (*mecA*)	Negative	Positive	99.3	-	0.7
Vancomycin screen	Negative	Positive	100.0	-	0.0

*Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

S. aureus from wound specimens were screened for methicillin resistance by the cefoxitin disk method in the same way as blood culture isolates. Seven out of 1,061 (0.7%) isolates were confirmed as MRSA by *mecA* PCR. This is the same prevalence as in 2007 and at the same level as in blood cultures (see above). The seven MRSA isolates all displayed cefoxitin zone diameters below the screening breakpoints for the respective test systems. One MRSA isolate was resistant to ciprofloxacin and erythromycin and another was resistant to erythromycin, clindamycin and fusidic acid. The remaining five isolates were susceptible to all non-beta-lactams tested. Only three out of 1,061 MSSA isolates were false positive by the cefoxitin test, and none of these isolates had zone diameters more than two millimeters below the sceening breakpoint.

The breakpoints for a number of non-beta-lactam antimicrobial agents including fusidic acid, rifampicin and trimethoprim-sulfamethoxazole were adjusted in 2009, but the historical data have not been recalculated as very few isolates would have been categorized differently by the new protocol. Further details are given in the figure legends to Figure 37.

The high prevalence of resistance to fusidic acid in *S. aureus* wound isolates continued to decline from 25.0% in 2004, 14.5% in 2006 and 11.1% in 2007, to 10.3% in 2008, see Table 36 and Figure 37. One may speculate that this is due to herd immunity to the fusidic acid resistant clone which has caused a high incidence of bullous impetigo over the last years. The prevalence of resistance to fusidic acid is still much lower in blood culture isolates (4.6%).

For other antimicrobial agents such as tetracyclines and macrolides there were only minor changes from 2007 to 2008, and the prevalences of non-susceptibility were similar for blood culture isolates and isolates from wound specimens. A total of 53 (5.0%) isolates were non-susceptible to erythromycin, and 49 of these were further examined for determination of resistance phenotype. The majority (25/49, 51% of macrolide resistant isolates) were inducibly resistant to clindamycin, thus representing the $iMLS_B$ phenotype. Only a few isolates were either constitutively resistant to clindamycin (n=10) or low-level resistant to erythromycin (n=14) expressing efflux mediated M type resistance. The findings are in accordance with the results from blood culture isolates.

A total of 73.8% of the isolates were beta-lactamase positive which is unchanged from 2007. Resistance to fusidic acid was significantly more common among the 783 beta-lactamase positive isolates (11.7%) than among the 278 beta-lactamase negative ones (6.1%). A similar trend was seen for ciprofloxacin (3.7% vs 0.7%) and tetracycline (6.5% vs 4.0%).

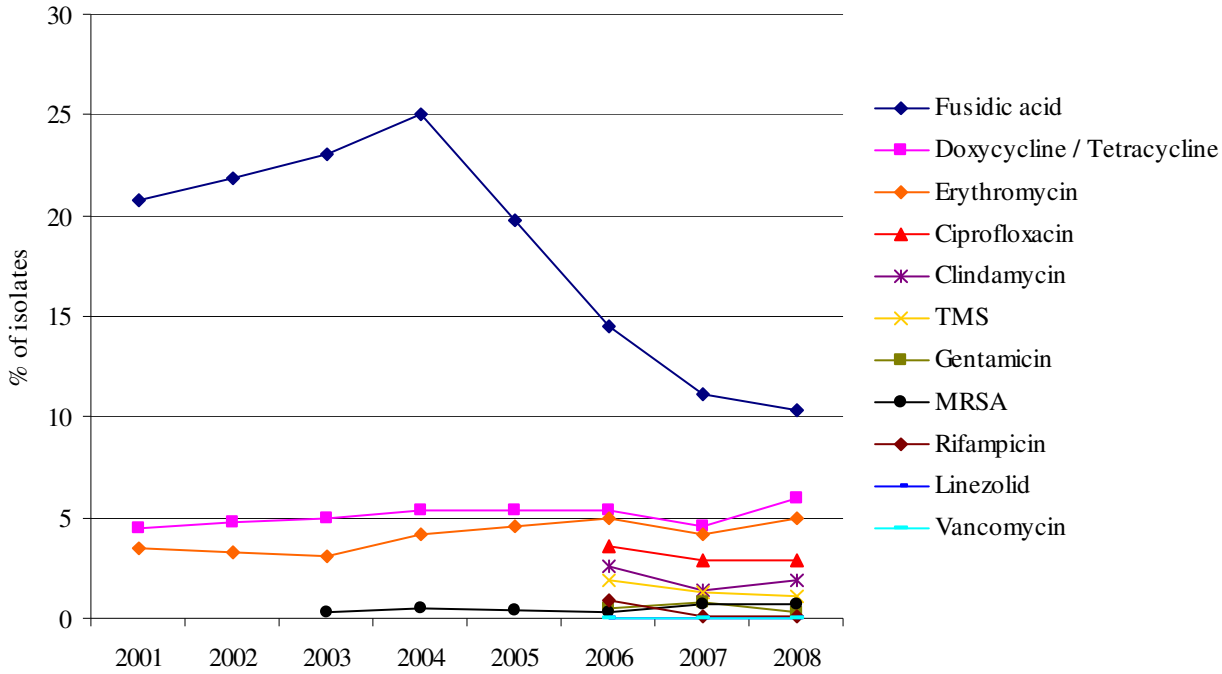

FIGURE 37. Prevalences of non-susceptibility to various antimicrobial agents among *Staphylococcus aureus* wound isolates 2000 – 2008. The breakpoint for susceptibility to gentamicin was decreased from S ≤ 2 mg/L to S ≤ 1 mg/L in 2006. Doxycycline was replaced by tetracycline in 2006. The breakpoints for clindamycin were reduced from S ≤ 1 mg/L and R > 2 mg/L to S ≤ 0.25 mg/L and R > 0.5 mg/L in 2007. The breakpoints for fusidic acid were increased from S ≤ 0.5 mg/L and R > 0.5 mg/L to S ≤ 1 mg/L and R > 1 mg/L in 2009. The breakpoint for resistance to trimethoprim-sulfamethoxazole was decreased from R > 8 mg/L to R > 4 mg/L in 2009. The breakpoints for rifampicin were decreased from S ≤ 1 mg/L and R > 1 mg/L to S ≤ 0.06 mg/L and R > 0.5 mg/L in 2009.

MRSA infections in humans in Norway 2008

Infections with methicillin resistant *Staphylococcus aureus* (MRSA) have been notifiable in Norway since 1995, and colonization without infection was made notifiable in 2005. Discrimination between colonization and infection can be difficult. A total of 652 cases of MRSA were notified in 2008. A total of 348 (53%) cases were reported as infections and 304 as colonizations (Figure 39). Males and females were equally affected. At the time of diagnosis, 148 (23%) were hospitalised, 61 (9%) were residents in nursing homes, and 443 (68%) were diagnosed outside health care institutions. 40 persons were reported being health care worker, of whom 36 were colonized with MRSA and four had MRSA infections.
The majority of 348 cases notified with MRSA infections were reported with a clinical picture of wound infections or abscesses (88% of reported infections). The number of reported severe infections was low, 17 infections were classified as systemic infections or organ-specific infections: urinary tract infections (9), septicaemia (4), lower respiratory tract infections (3) and arthritis (1). MRSA was detected in blood cultures in ten patients in 2008. From the start of systematic surveillance in 1995 the annual number of MRSA detections reported from blood or spinal fluids have been ten cases or less.
The measured increase in MRSA detections has to be interpreted with caution. The increase is mainly seen in non-hospitalised patients with minor infections or colonisations. This may indicate increased testing of non-hospitalised patients.

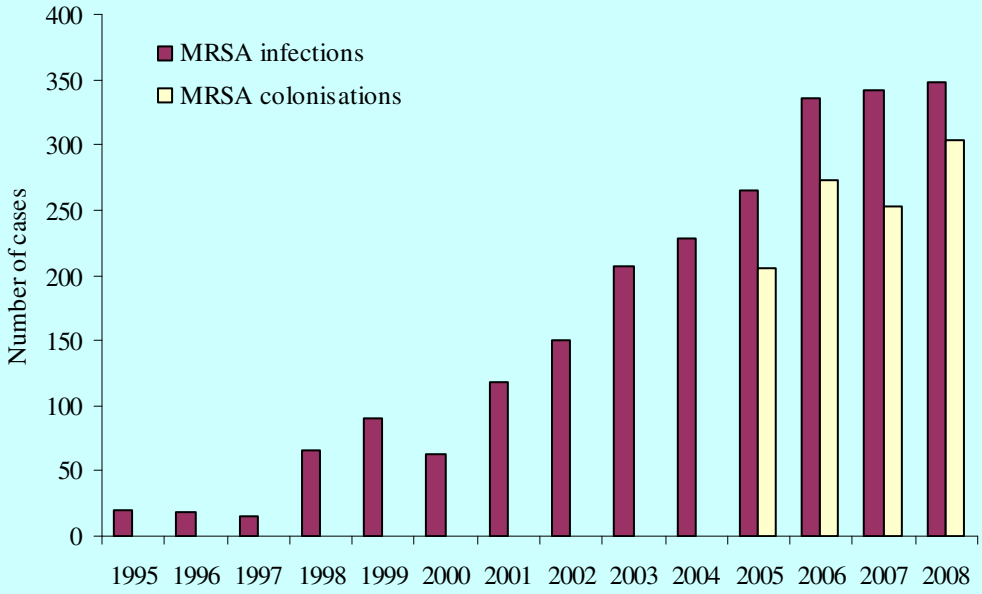

FIGURE 38. Reported cases of MRSA infection 1995-2008 and MRSA colonisation 2005-2008 in Norway.

Among 348 cases reported as MRSA infections in 2008, we found 91 different spa-types. The five most frequent spa-types were t019 (14.3%), t008 (12.9%), t002 (10.0%), t044 (9.1%), and t437 (5.0%) (Table 37). The ten most frequent spa-types reported as infections represented 60% of all the isolates reported as MRSA infections. In total, their share was 58% of all strains. Compared to 2006 (51.9%) and 2007 (53.1%) the diversity among strains was decreasing. 56% of all strains were Panton-Valentine Leukocidin (PVL) positive. The PVL positive rate was 82% among the five most common spa-types causing infections.

TABLE 37. Most frequent spa-types causing infection distributed on clinical diagnosis.

	Spa-types (n)					
Clinical diagnosis	Total	t019	t008	t002	t044	t437
Skin and soft tissue infection	297	46	40	29	30	17
Urinary tract infection	8		2	2		
Blood stream infection	10					
Respiratory tract infection	3			1		
Bone and joint infection	1	1				
Meningitis	1		1			
Others	20	2	1	2	1	
Total	337*	49	44	34	31	17

*11 of the strains causing infections were not received by the Reference Laboratory.

Among 304 cases reported as colonization in 2008, we found 127 different spa-types. The five most frequent spa-types were t002 (13.4%), t223 (8.7%), t304 (7.7%), t008 (5.7%) and t044 (4.7%) (Table 38). The ten most frequent spa-types reported as colonization, represent 56.7% of all the isolates in this group. 20.1% of the strains were PVL positive.

TABLE 38. Most frequent spa-types associated with colonisation.

	Spa-types (n)					
	Total	t002	t223	t304	t008	t044
Carriers	298*	40	26	23	17	14

*6 of the strains associated with colonisation were not received by the Reference Laboratory.

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Enterococcus spp. in blood cultures

TABLE 39. *Enterococcus* spp. blood culture isolates (n=474). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 4	> 8	80.0	0.0	20.0
Gentamicin	≤ 128	> 128	63.3	-	36.7
Linezolid	≤ 4	> 4	100.0	-	0.0
Vankomycin	≤ 4	> 8	98.7	-	1.3

TABLE 40. *Enterococcus faecalis* blood culture isolates (n=323). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 4	> 8	100.0	0.0	0.0
Gentamicin	≤ 128	> 128	66.6	-	33.4
Linezolid	≤ 4	> 4	100.0	-	0.0
Vankomycin	≤ 4	> 8	100.0	-	0.0

TABLE 41. *Enterococcus faecium* blood culture isolates (n=110). Sampling, laboratory methods, and data handling are described in Appendix 5. Distributions of zone diameters are available at www.antibiotikaresistens.no.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Ampicillin	≤ 4	> 8	21.8	0.0	78.2
Gentamicin	≤ 128	> 128	46.4	-	53.6
Linezolid	≤ 4	> 4	100.0	-	0.0
Vankomycin	≤ 4	> 8	100.0	-	0.0

RESULTS AND COMMENTS

As in previous years, enterococci were analysed both as a group and separately for *E. faecalis* and *E. faecium*. 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 infections with each enterococcal species. The overall results for enterococci are presented in Table 39. The surveillance in NORM 2008 included 323 (68.1%) *E. faecalis* isolates, 110 (23.2%) *E. faecium* isolates and 41 (8.6%) unspeciated enterococcal isolates. The proportion of isolates not speciated to the genus level or identified as *E. faecalis* or *E. faecium* has decreased over the last three years.

The panel of antimicrobial agents examined was unchanged from 2007. Streptomycin is not included in the printed tables as one of the disk diffusion systems does not provide breakpoints for this substance. Distributions of zone diameters for both systems are available at www.antibiotikaresistens.no. The breakpoint for susceptibility to ampicillin was increased from S ≤ 2 mg/L to S ≤ 4 mg/L in 2009. The results from previous years have been recalculated according to the new breakpoint.

E. faecalis was universally susceptible to ampicillin (Table 40). The prevalence of non-susceptibility to ampicillin in *E. faecium* remained relatively stable at 78.2% compared to 80.3% in 2007 and 82.7% in 2006 (Table 41 and Figure 39). The prevalence of high-level gentamicin resistance (HLGR) continued to increase in *E. faecalis* from 30.4% in 2007 to 34.4% in 2008 (Figure 40). In *E. faecium*, the prevalence of HLGR remained unchanged at 53.6% compared to 55.1% in 2007. Virtually all (56/59, 94.9%) HLGR *E. faecium* isolates were concomitantly non-susceptible to ampicillin. Conversely, 56 out of 86 (65.1%) ampicillin non-susceptible *E. faecium* also displayed HLGR. These findings are identical to the results from previous years.

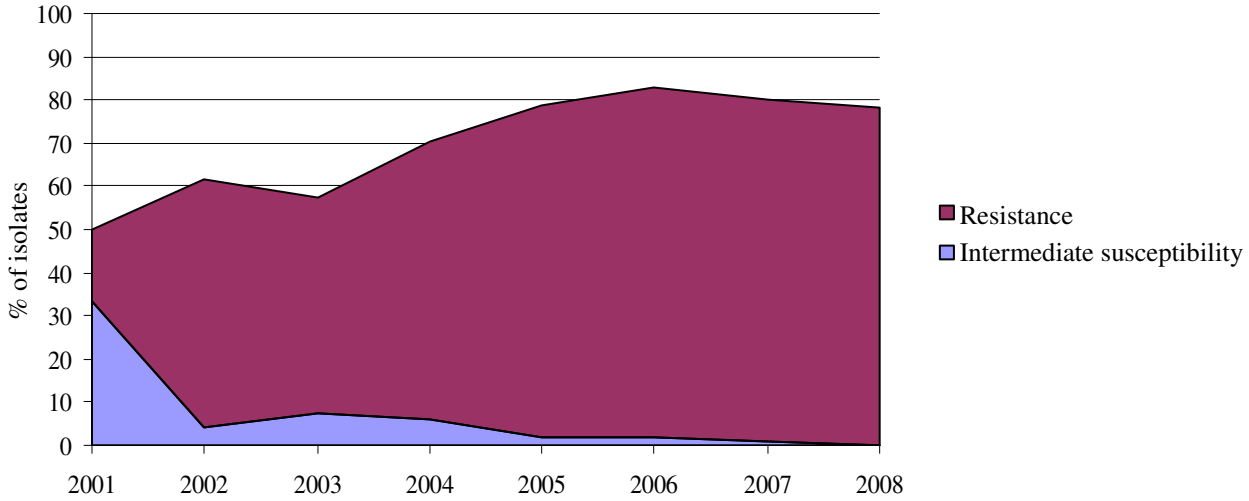

FIGURE 39. Prevalence of intermediate susceptibility and resistance to ampicillin in *E. facium* blood culture isolates 2001-2008. The results are interpreted according to the 2009 breakpoint protocol of S ≤ 4 mg/L and R > 8 mg/L.

The strong linkage between ampicillin resistance and HLGR may indicate the continuing presence of the internationally disseminated *E. faecium* clonal complex (CC) 17 which is non-susceptible to ampicillin and often harbors high-level resistance to aminoglycosides and vancomycin. The wide dissemination of high-level gentamicin resistance in enterococci is of great concern as it abolishes the bactericidal synergy between aminoglycosides and beta-lactams often used for treatment of severe enterococcal infections.

Transferable vancomycin resistance has not yet been established in clinical enterococcal isolates in Norway. Six isolates were reported as vancomycin resistant (1.3%), but they were all registered as either *E. gallinarum* (n=4) or *E. casseliflavus* (n=2) which are inherently low-level resistant to vancomycin due to expression of the VanC ligase. There were no isolates with transferable high-level vancomycin resistance, and all the six isolates with low-level resistance were fully susceptible to linezolid.

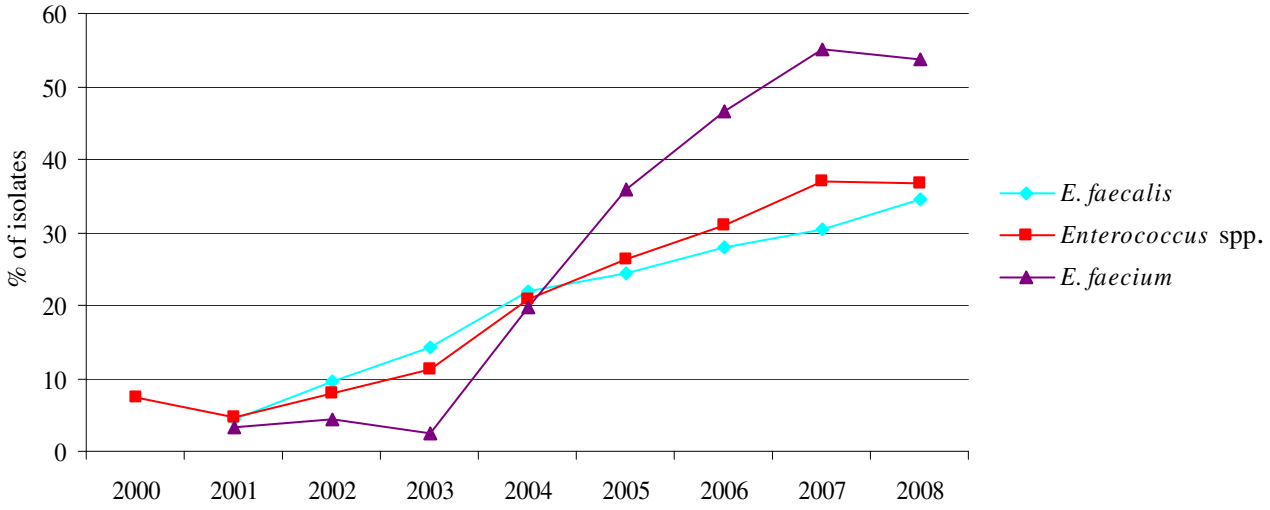

FIGURE 40. Prevalence of high-level resistance to gentamicin in blood culture isolates of *E. faecalis*, *E. faecium* and all enterococci combined during 2000-2008. The breakpoint for high-level resistance was decreased from R ≥ 1,024 mg/L to R > 128 mg/L in 2004.

Clostridium difficile associated disease

Since *Clostridium difficile* became established as the primary identifiable pathogen of antibiotic-associated diarrhoea and colitis in 1978, these iatrogenic complications of antibiotic use have been extensively studied. *Clostridium difficile* associated disease (CDAD) is a toxin-mediated disease in which two main toxins, A and B, are responsible for the clinical picture. For a long time toxin A was thought to be the key component in triggering the disease pathogenesis, but toxin B now appears to play an equally important role. *Clostridium difficile* infection is an important and frequent iatrogenic complication. For surveillance purposes, CDAD is classified as either hospital onset-healthcare facility-associated, community onset-healthcare facility-associated or community-associated. Thus, cases not associated with hospitals have also been identified.

Risk factors

Clinical disease and *C. difficile* toxin are present almost exclusively in patients with antibiotic exposure. Any antibacterial agent may be the cause, but the risk depends both on the type of agent and the extent of use. In the first years, clindamycin followed by ampicillin or amoxicillin were the most common causes. However, during the 1980s cephalosporins became the prominent antibiotic group causing CDAD as a result of extensive use of this class of antibiotics. During later years, the fluoroquinolones have also been added to the list. In addition to antibiotic exposure, advanced age and hospitalization are major risk factors.

Diagnosis

The cell culture cytotoxicity assay was the first diagnostic test for *C. difficile* toxin. It is a two-step test including neutralization with anti-toxin and detects primarily toxin B with high sensitivity, but is labour intensive and slow (24-48 h). Most laboratories nowadays therefore use an enzyme immunoassay (EIA), which either detects toxin A or both toxin A and B. The EIA tests are, however, less sensitive (65-85%) than the cytotoxicity assay. Stool culture for *C. difficile* is the most sensitive test available when performed appropriately, but is labour intensive and requires 48-96 hours for results, and due to the presence of non-toxigenic strains the isolates must be tested for toxin to avoid false positive results. Culture is, however, essential for epidemiological purposes and susceptibility testing. Nevertheless, the majority of laboratories do not perform culture routinely. Another option for detection of *C. difficile* is the use of EIA for glutamate dehydrogenase (GDH), also called *C. difficile* common antigen. The test is rapid (< 1 hour) and very sensitive (> 99%), but less specific (89%) with positive predictive values ~ 60% and toxin testing is required to verify the diagnosis. The optimal diagnostic testing strategy is therefore a two-step method with EIA for GDH as screen and cell cytotoxicity assay or culture with cytotoxicity assay as confirmatory test (1).

New epidemic

In most cases, the disease used to be relatively easy to manage, with the exception of occasional institutional outbreaks and a nagging problem of relapsing disease. However, since the turn of the century increasing incidence and severity of CDAD have been reported from many countries (2-4). The increased incidence of CDAD and *C. difficile* associated mortality appears to be due to the spread of an epidemic strain, known variously as North American Pulsed-field 1(NAP1), restriction analysis type "BI" or PCR ribotype 027. This strain, which is historically uncommon, has a 16-fold increased toxin A production and 23-fold increased toxin B production documented in vitro. It also has an extra toxin, known as binary toxin, of unknown significance. Coincidentally, NAP1/BI/027 has become more resistant to the fluoroquinolones.

Clostridium difficile in Norway

Except for outbreaks in hospitals, *Clostridium difficile* infection is not a notifiable disease in Norway, and the insight in the epidemiology of this disease is therefore scanty. In September 2007 a questionnaire was sent to all Norwegian microbiological laboratories enquiring about the diagnostic tests used in 2007 and the numbers of positive specimens during 2006. Twenty one of 23 laboratories responded. Eighteen laboratories used a commercial EIA for toxin A and B as their primary diagnostic method, and 3 laboratories tested for toxin A only. Six laboratories also cultured for *C. difficile*, either as a secondary test for toxin positive specimens (5 laboratories) or in selected cases only (1 laboratory). A total of 19,080 toxin tests were done in the 21 laboratories in 2006 (not excluding repeat samples from the same patients). On average, 13% of the tests were toxin positive.

The laboratories performing culture detected a total of 687 strains of *C. difficile* in 2006. However, PCR ribotyping or other molecular typing methods were not done by any laboratory at that time. Ribotyping was established at Rikshospitalet University Hospital, Department of Infection Prevention, in November 2007. During 2007 and 2008 a total of 123 strains from several hospitals were tested, identifying 25 different PCR ribotypes. The most frequent types were ribotype 014 (21%), ribotype 012 (11%) and ribotype 001 (10%). Three strains (2.4%) were ribotype 027 and these were all resistant to moxifloxacin. Ribotype 078, another strain that may be associated with more serious disease, was identified in 8 strains (6.5%). During 2008 and 2009 PCR ribotyping was established at two other laboratories.

Susceptibility and treatment

In the first years after 1978 oral vancomycin was the most common treatment. During the 1980s metronidazole replaced vancomycin as standard treatment due to the concern associated with the increasing prevalence of vancomycin resistance among enterococci. However, for serious cases, vancomycin may offer an advantage over metronidazole (5, 6).

Antimicrobial therapy plays a central role in the pathogenesis of CDAD because of its disruption of indigenous intestinal microflora, which allows *C. difficile*, if present, to grow and produce toxin. Inhibitory activity against *C. difficile* may also influence the likelihood that particular drugs may cause CDAD. For example, the emergence of fluoroquinolones as high-risk agents for CDAD has coincided with increasing fluoroquinolone resistance.

In most studies, isolates of *C. difficile* have generally been found to be susceptible to vancomycin and metronidazole (7, 8). However, a few studies have reported strains resistant to metronidazole or with reduced susceptibility to vancomycin (9, 10). Also recent studies from several countries have reported increasing prevalence of resistance and a high proportion of isolates resistant to clindamycin, moxifloxacin, ceftriaxone and erythromycin (11, 12). Plasmids as a source for antibiotic resistance genes have not been found in *C. difficile*. However, conjugative transposons are now recognized to be as important for the dissemination of antibiotic resistance genes as conjugative plasmids.

Due to the need for better treatment of severe or fulminant *Clostridium difficile* infection and the problem with relapsing disease, a number of alternative therapeutic options have been tried, and several are presently under investigation. Targeted antimicrobials (i.e. bactericidal) undergoing clinical trials are rifaximin, OPT-80, ramoplanin and nitazoxanide, but no major breakthrough has so far been reported. Also, toxin binding drugs, active (i.e. vaccination) and passive immunotherapy (i.e. immunoglobulins) as well as novel molecules and approaches like furylidene thiosemicarbazones, cinnamon zylanicum (cinnamon) bark oil, replidyne (REP3123), orally administered lipopeptides and lanthionine (amino-acid)-containing antibiotics as well as agents that inhibit germination of spores ("blockers" of glycine and cholate) and bacteriophages are all under investigation. Fecal flora replenishment and probiotics are also options, but the latter has not been demonstrated to be effective (13).

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Streptococcus pneumoniae in blood cultures

TABLE 42. *Streptococcus pneumoniae* blood culture isolates (n=507). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Penicillin G	≤ 0.064	> 2	97.0	2.8	0.2
Cefuroxime	≤ 0.5	> 1	98.6	0.4	1.0
Cefotaxime	≤ 0.5	> 2	99.4	0.6	0.0
Erythromycin	≤ 0.25	> 0.5	91.5	0.2	8.3
Clindamycin	≤ 0.5	> 0.5	98.2	-	1.8
Tetracycline	≤ 1	> 2	95.7	1.0	3.4
Trimethoprim-sulfamethoxazole*	≤ 1	> 2	94.7	2.0	3.4
Oxacillin screen (mm)	≥ 20	< 20	95.7	-	4.3

*Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 43. *Streptococcus pneumoniae* blood culture isolates (n=507). Distribution (%) of MICs (mg/L) and zone diameters for oxacillin (mm).*

	≤0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	≥ 128
Penicillin G	3.4	14.8	55.0	21.9	2.0	1.2	0.2	0.6	0.8		0.2					
Cefuroxime			34.3	45.2	14.8	2.8	1.6		0.4	0.4	0.6					
Cefotaxime	1.4	8.1	43.6	36.7	6.7	1.4	1.0	0.6	0.6							
Erythromycin		0.2	0.4	4.1	22.1	56.2	8.5	0.2		0.4	0.6	1.6	3.2	1.0	0.2	1.4
Clindamycin			1.0	4.3	35.3	44.0	13.4	0.2			0.2		0.2			1.4
Tetracycline				1.0	17.4	63.5	12.6	1.0	0.2	1.0	0.6	0.4	1.6	0.6	0.2	
TMS**					1.4	19.7	57.0	14.2	2.4	2.0	1.6	1.0	0.2	0.6		
Norfloxacin										4.5	27.4	50.7	15.6	1.6	0.2	

	< 20	20	21	22	23	24	25	26	27	28	29	30	31	32	33	≥ 34
Oxacillin disk	4.3	0.6	0.2	0.8	1.2	3.2	4.7	6.5	9.5	14.2	10.5	15.2	8.3	5.9	2.4	12.6

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

RESULTS AND COMMENTS

The results are summarized in Tables 42-43 and Figures 41-42. All *S. pneumoniae* isolates collected during the first nine months of 2008 were included, and this sampling strategy was unchanged from 2007. It is therefore noteworthy that the sample size was reduced from 609 isolates in 2007 to 507 isolates in 2008. This trend was also seen in the number of systemic *S. pneumoniae* infections reported to the Norwegian Surveillance System for Communicable Diseases (MSIS): 1,126 in 2004, 1,083 in 2005, 1,015 in 2006, 958 in 2007, and 856 in 2008. The proportion of *S. pneumoniae* among blood culture isolates in the laboratory databases was reduced from 13.2% in 2004 to 8.7% in 2008 (skin flora excluded). A major shift in the epidemiology of *S. pneumoniae* is thus apparently underway, possibly in part related to introduction of the conjugated pneumococcal vaccine after 2006.

The general breakpoint for resistance to penicillin G was increased from R > 1 mg/L to R > 2 mg/L in 2009. In cases of meningitis, a breakpoint for resistance of R > 0.06 mg/L should be used. The breakpoint for susceptibility to tetracycline was decreased from S ≤ 2 mg/L to S ≤ 1 mg/L, whereas the breakpoint for susceptibility to trimethoprim-sulfamethoxazole was increased from S ≤ 0.5 mg/L to S ≤ 1 mg/L. The breakpoints for resistance remained unchanged for both substances. In the following, all historical data are recategorized according to the new breakpoints.

A total of 3.0% (15/507) *S. pneumoniae* isolates were non-susceptible to penicillin G. This is a slight decrease from 3.3% in 2007. Fourteen isolates (2.8%) were intermediately susceptible (MIC 0.125-1 mg/L) whereas a single isolate was reported with an MIC of 4 mg/L. Seven of the penicillin G non-susceptible isolates were either intermediately susceptible (MIC 1 mg/L, n=2) or resistant (MIC 2-4 mg/L, n=5) to cefuroxime. Three of the cefuroxime resistant isolates were also intermediately susceptible to cefotaxime (MIC 1 mg/L). No penicillin G susceptible isolates displayed reduced susceptibility to cephalosporins. The oxacillin screening disk is often used to discriminate between penicillin susceptible and non-susceptible isolates. Thirteen of the 15 penicillin G non-susceptible isolates were resistant to oxacillin. Conversely, nine penicillin G susceptible isolates were oxacillin resistant. The sensitivity and specificity of the screening test were thus 86.7% and 98.2%, respectively. The oxacillin screening test identified all cephalosporin non-

susceptible isolates. Many of the penicillin non-susceptible *S. pneumoniae* isolates were concomitantly non-susceptible to erythromycin (10/15), trimethoprim-sulfamethoxazole (9/15) and tetracycline (5/15).

The decrease in the prevalence of macrolide resistance seen in 2007 continued in 2008 (Figure 41). A total of 8.3% of the isolates were erythromycin resistant compared to 9.4% in 2007. In addition, 0.2% displayed reduced susceptibility to this agent in 2008 compared to 0.5% in 2007. As the surveillance protocol was not changed during this time period, the results support the hypothesis that the epidemiology of systemic *S. pneumoniae* infections is changing with decreasing absolute numbers and proportions of resistant serotypes included in the 7-valent conjugated pneumococcal vaccine (PCV-7). Among the 43 erythromycin non-susceptible isolates, 41 were subjected to double disk diffusion (DDD) tests for characterization of MLS phenotypes. A majority of isolates (n=25, 61.0% of erythromycin non-susceptible isolates, 4.9% of all isolates) displayed a phenotype compatible with efflux-based M-type resistance to erythromycin only. The remaining isolates were either inducibly (n=8, 19.5% of erythromycin non-susceptible isolates, 1.6% of all isolates) or constitutively (n=8, 19.5% of erythromycin non-susceptible isolates, 1.6% of all isolates) resistant to clindamycin, thus indicating the presence of *erm*-encoded MLS resistance to both macrolides and lincosamides. The distribution of MLS phenotypes was not significantly altered from 2007. Further studies are needed to explore the relationship between vaccination, incidence of systemic pneumococcal infections, serotype distribution and burden of resistance in different age groups.

There was a minor decrease in the prevalence of non-susceptibility to trimethoprim-sulfamethoxazole from 6.6% in 2007 to 5.4% in 2008. Similarly, the prevalence of non-susceptibility to tetracycline decreased from 5.2% in 2007 to 4.4% in 2008 (Figure 42). The low prevalence of high-level norfloxacin resistance (Table 43) reflects that levofloxacin and other "respiratory fluoroquinolones" are not marketed in Norway.

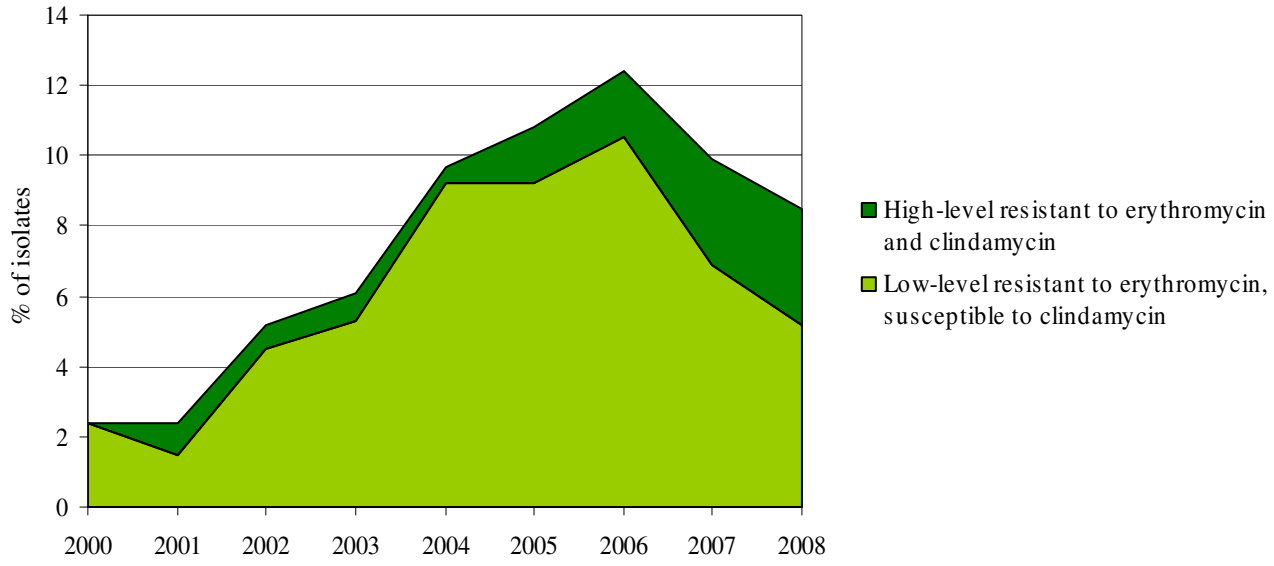

FIGURE 41. Prevalence (%) of macrolide non-susceptible *Streptococcus pneumoniae* blood culture isolates with constitutive or inducible MLS_B phenotype (high-level resistance to erythromycin and clindamycin) and M phenotype resistance (low-level resistance to erythromycin, susceptibility to clindamycin) 2000-2008. A breakpoint for susceptibility of S ≤ 0.25 mg/L was used for inclusion of isolates which were subsequently categorized on the basis of the double disk diffusion (DDD) test.

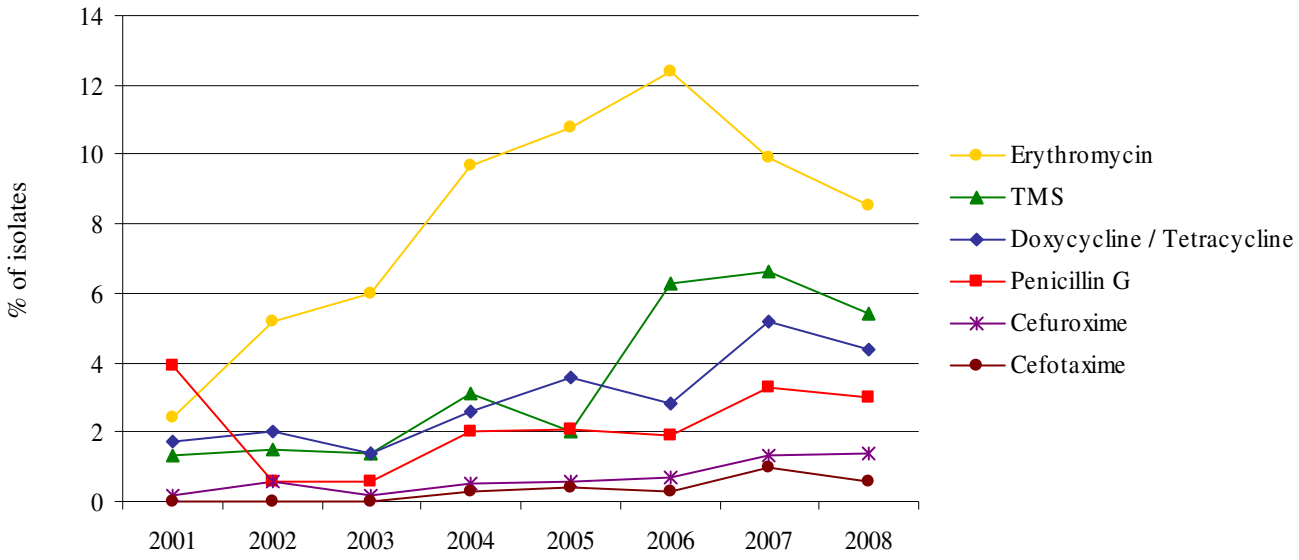

FIGURE 42. Prevalences (%) of non-susceptibility to various antimicrobial agents in *Streptococcus pneumoniae* blood culture isolates during 2000-2008. Doxycycline was substituted by tetracycline in 2005. The breakpoints for erythromycin were changed from S ≤ 0.5 mg/L and R > 0.5 mg/L to S ≤ 0.25 mg/L and R > 0.5 mg/L in 2007. The breakpoints for susceptibility were changed from S ≤ 2 mg/L to S ≤ 1 mg/L for tetracycline and from S ≤ 0.5 mg/L to S ≤ 1 mg/L for trimethoprim-sulfamethoxazole in 2009. The genereal breakpoint for resistance to penicillin G was increased from R > 1 mg/L to R > 2 mg/L in 2009. All results are categorized according to the 2009 breakpoint protocol.

Streptococcus pyogenes in specimens from wounds and the respiratory tract

TABLE 44. *Streptococcus pyogenes* isolates from wound specimens (n=342). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Penicillin G	≤ 0.25	> 0.25	100.0	-	0.0
Erythromycin	≤ 0.25	> 0.5	98.8	0.0	1.2
Clindamycin	≤ 0.5	> 0.5	99.7	-	0.3
Tetracycline	≤ 1	> 2	88.6	0.0	11.4
Trimethoprim-sulfamethoxazole*	≤ 1	> 2	98.0	1.2	0.9

*Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 45. *Streptococcus pneumoniae* isolates from wound specimens (n=342). Distribution (%) of MICs (mg/L) and zone diameters for oxacillin (mm).*

	≤0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	≥ 128
Penicillin G	0.9	30.1	68.1	0.3	0.3	0.3										
Erythromycin				3.5	36.5	51.5	7.3						0.3			0.9
Clindamycin			1.2	8.8	39.2	48.8	1.5	0.3								0.3
Tetracycline				0.3	23.1	51.5	13.5	0.3					4.4	4.4	2.0	0.6
TMS**			0.3	0.9	7.0	28.4	32.7	22.8	5.8	1.2	0.3			0.6		

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

TABLE 46. *Streptococcus pyogenes* respiratory tract isolates (n=262). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Penicillin G	≤ 0.25	> 0.25	100.0	-	0.0
Erythromycin	≤ 0.25	> 0.5	96.9	0.0	3.1
Clindamycin	≤ 0.5	> 0.5	99.6	-	0.4
Tetracycline	≤ 1	> 2	92.0	0.0	8.0
Trimethoprim-sulfamethoxazole*	≤ 1	> 2	94.7	2.3	3.1

*Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.

TABLE 47. *Streptococcus pneumoniae* respiratory tract isolates (n=262). Distribution (%) of MICs (mg/L) and zone diameters for oxacillin (mm).*

	≤0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	≥ 128
Penicillin G	1.1	40.8	54.2	2.7	0.4	0.4	0.4									
Erythromycin				5.3	40.8	45.8	5.0		0.4	0.4	0.8	0.4	0.8	0.4		
Clindamycin			0.4	5.0	51.9	39.7	2.7		0.4							
Tetracycline				1.1	27.9	48.1	14.9				1.1	0.8	2.7	2.3	1.1	
TMS**				0.4	4.2	21.4	38.2	23.7	6.9	2.3	0.4		0.4	2.3		

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

RESULTS AND COMMENTS

Streptococcus pyogenes (beta-haemolytic streptococci group A - GAS) has previously been surveyed in NORM in 2002, 2004 and 2006. Blood cultures were included in the surveillance protocol in 2006, for the other years only respiratory tract isolates and isolates from wound specimens have been examined. The results for 2008 are presented in Tables 44-47 and the trends during 2002-2008 in Figure 43. The breakpoints for penicllin G were increased from S ≤ 0.125 mg/L and R > 0.125 mg/L to S ≤ 0.25 mg/L and R > 0.25 mg/L in 2009. Similarly, the breakpoint for susceptibility to tetracycline was decreased from S ≤ 2 mg/L to S ≤ 1 mg/L, and the breakpoint for susceptibility to trimethoprim-sulfamethoxazole was increased from S ≤ 0.5 mg/L to S ≤ 1 mg/L. All comparisons with historical data are based on recategorization of isolates using the 2009 breakpoint protocol.

As expected, all isolates were fully susceptible to penicillin G. Penicillin G non-susceptibility has never been detected in group A streptococci, and the highest MIC values in this study were 0.25 mg/L which is equivalent to the breakpoint for susceptibility. Most isolates displayed MICs of 0.016 mg/L.

As seen in Figur 43, the prevalence of resistance to tertacycline in isolates from wound specimens has steadily decreased over the years. A similar tendency was seen among resipratory tract isolates until 2006, but the trend has now been broken. Consequently, the significant difference in tetacycline resistance between isolates from wounds (11.4%) and the resipratory tract (8.0%) has now been reduced. Differences in resistance rates between isolates from different clinical conditions may be caused by clonal variation, but further studies are needed to draw any firm conclusions.

Macrolide resistant group A streptococci has been a problem in many countries, including Finland and Italy. In NORM, the prevalence of erythromycin resistance has remained stable below 5% and was unchanged from 2006 to 2008. There was no major difference between isolates from wound specimens (1.2%) and the respiratory tract (3.1%). Nine out of 12 erythromycin resistant isolates were further investigated using the double disk diffusion (DDD) assay for determination of resistance phenotype. Five isolates displayed *erm*-encoded MLS_B resistance either inducibly (n=3) or constitutively (n=2). The remaining four isolates displayed low-level resistance to erythromycin seen in the *mef*-encoded M-phenotype. It is remarkable that macrolide resistance has remained so rare in Norwegian group A streptococci when both *mef* and *erm* resistance determinants have disseminated widely in *Streptococcus pneumoniae* in the same time period. This observation indicates that herd immunity and other selective forces are acting in addition to the effect of antibiotic exposure.

In Norway, trimethoprim-sulfamethoxazole (TMS) is rarely used for treatment of infections caused by group A streptococci. The increase in non-susceptibility to TMS noted in earlier reports is less obvious after adjustment of the breakpoints, and the trend from previous years was apparently reversed in 2008.

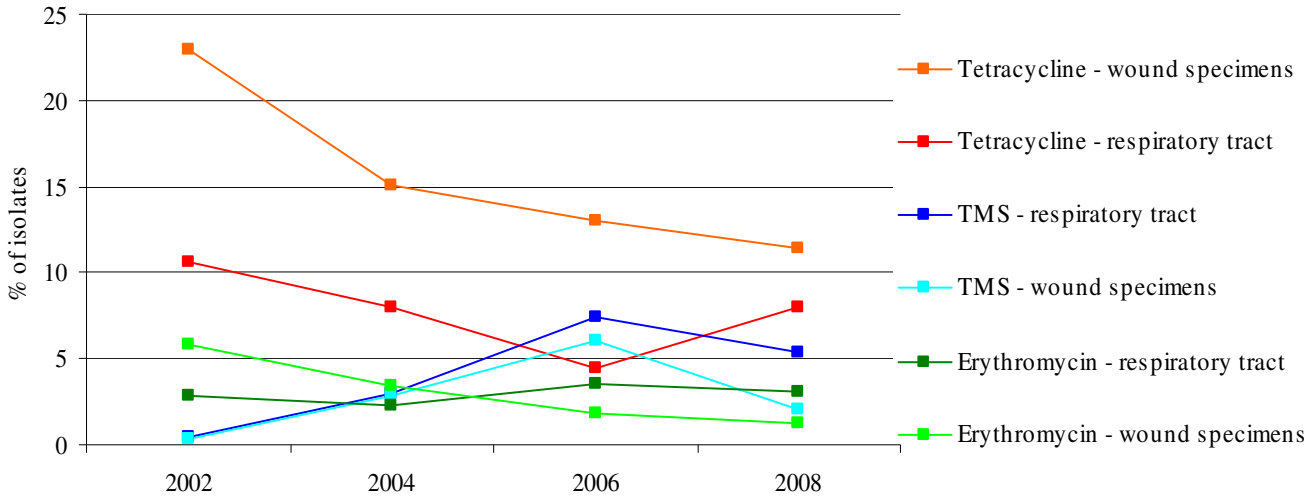

FIGURE 43. Prevalences of non-susceptibility to various antimicrobial agents in *Streptococcus pyogenes* from wound specimens and the respiratory tract in 2002, 2004, 2006 and 2008. Doxycycline was used in 2002 and 2006 but replaced by tetracycline in 2006 and 2008. The breakpoint for susceptibility to erythromycin was reduces from S ≤ 0.5 mg/L to S ≤ 0.25 mg/L in 2008. The breakpoint for susceptibility to tetracycline was decreased from S ≤ 2 mg/L to S ≤ 1 mg/L, and the breakpoint for susceptibility to trimethoprim-sulfamethoxazole (TMS) was increased from S ≤ 0.5 mg/L to S ≤ 1 mg/L in 2009. All data are categorized according to the 2009 breakpoint protocol.

Mycobacterium tuberculosis

A total of 324 cases of infection with *M. tuberculosis* were reported to the Norwegian Surveillance System for Communicable Diseases (MSIS) in 2008. Among these, 27 cases had been previously treated with drugs against tuberculosis, 13 from Africa, 7 from Europe and 7 from Asia. Fifteen had an earlier history of tuberculosis without drug treatment. A total of 225 cases were confirmed by culture and the strain susceptibility tested. The results are presented in Table 48.

TABLE 48. Antimicrobial susceptibility of 225 isolates of *M. tuberculosis* complex isolated from human infections in 2008.

Origin of birth	No. of cases	No. of isolates	Resistance to antimicrobial agents (No. of isolates)					
			Isoniazid	Rifampicin	Ethambutol	Streptomycin	Pyrazinamid	MDR TB*
Norway	56	34	1			1	1	
Europe outside Norway	28	22	6	1	3	6	1	1
Asia	109	82	9	2	2	4	3	1
Africa	126	84	18	3	1	14	2	2
America	3	2						
No information	2	1						
Total	324	225	34	6	6	25	7	4
Proportion of resistant isolates (%)			15.1	2.7	2.7	11.1	3.1	1.8

*MDR TB: Multi drug resistant tuberculosis, resistant to at least rifampicin and isoniazid.

RESULTS AND COMMENTS

Susceptibility tests were also performed on *M. tuberculosis* isolates from 27 patients who had previously received antituberculosis drug treatment.

Candida spp. in blood cultures

TABLE 49. Antimicrobial susceptibility of *Candida albicans* blood culture isolates (n=140)*. Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Amphotericin B*	≤ 1	> 1	100.0	-	0.0
Fluconazole**	≤ 2	> 2	99.3	-	0.7
Voriconazole***	≤ 0.125	> 0.125	100.0	-	0.0
Caspofungin****	≤ 1	> 1	100.0	-	0.0
Anidulafungin	≤ 1	> 1	100.0	-	0.0

* 96 isolates were susceptibility tested for anidulafungin. ** Recommended breakpoints by the Norwegian Reference group on Antibiotic Susceptibility testing – AFA. *** Recommended breakpoints by the European Commitee on antimicrobial susceptibility testing – EUCAST. **** There are no European breakpoints for caspofungin and anidulafungin. Strains with MICs ≤ 1 mg/l are considered susceptible.

TABLE 50. *Candida albicans* blood culture isolates (n=140)*. 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
Ampho. B					1.4	10.7	41.5	45.7	0.7								
Fluconazole	0.7				1.4	18.6	61.5	15.7	1.4			0.7					
Voriconazole	15.7	69.4	11.4	1.4	2.1												
Caspofungin			1.4	8.6	35.8	40.7	10.7	2.8									
Anidulafungin	75.1	20.8	3.1	1													

* 96 isolates were susceptibility tested for anidulafungin. ** Shaded areas in each row indicate susceptibility (light) and resistance (dark).

TABLE 51. Antimicrobial susceptibility of *Candida glabrata* blood culture isolates (n=37)*. Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Amphotericin B*	≤ 1	> 1	100.0	-	0.0
Fluconazole**	≤ 2	> 2	13.5	-	86.5
Voriconazole***	≤ 0.125	> 0.125	59.5	-	40.5
Caspofungin****	≤ 1	> 1	100.0	-	0.0
Anidulafungin	≤ 1	> 1	100.0	-	0.0

* 25 isolates were susceptibility tested for anidulafungin. ** Recommended breakpoints by the Norwegian Reference group on Antibiotic Susceptibility testing – AFA. *** According to EUCAST, there is insufficient evidence that fluconazole and voriconazole are appropriate drugs for *C. glabrata*. Breakpoints for *C. albicans* are included in the table. **** There are no European breakpoints for caspofungin and anidulafungin. Strains with MICs ≤ 1 mg/l are considered susceptible.

TABLE 52. *Candida glabrata* blood culture isolates (n=22)*. 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
Ampho. B					5.4	5.4	18.9	67.6	2.7								
Fluconazole										13.5	54.1			2.7	8.1	2.7	18.9
Voriconazole					21.6	37.9	8.1		5.4	8.1		2.7		16.2			
Caspofungin						21.6	75.7	2.7									
Anidulafungin	8	12	72	8													

* 25 isolates were susceptibility tested for anidulafungin. ** Shaded areas in each row indicate susceptibility (light) and resistance (dark).

TABLE 53. Antimicrobial susceptibility of *Candida tropicalis* blood culture isolates (n=13)*. Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)		
	Susceptible	Resistant	Susceptible	Intermediately susceptible	Resistant
Amphotericin B*	≤ 1	> 1	100.0	-	0.0
Fluconazole**	≤ 2	> 2	100.0	-	0.0
Voriconazole***	≤ 0.125	> 0.125	100.0	-	0.0
Caspofungin****	≤ 1	> 1	100.0	-	0.0
Anidulafungin	≤ 1	> 1	83.3	-	16.7

* 6 isolates for anidulafungin susceptibility testing. ** Recommended breakpoints by the Norwegian Reference group on Antibiotic Susceptibility testing – AFA. *** According to EUCAST, there is insufficient evidence that fluconazole and voriconazole are appropriate drugs for *C. glabrata*. Breakpoints for *C. albicans* are included in the table. **** There are no European breakpoints for caspofungin and anidulafungin. Strains with MICs ≤ 1 mg/l are considered susceptible.

TABLE 54. *Candida tropicalis* blood culture isolates (n=13)*. 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
Ampho. B							15.4	69.2	15.4								
Fluconazole						7.7	23.1	61.5	7.7								
Voriconazole	7.7		15.3	38.5	38.5												
Caspofungin						23.1	69.2	7.7									
Anidulafungin			50	33.3						16.7							

* 6 isolates were susceptibility tested for anidulafungin. ** Shaded areas in each row indicate susceptibility (light) and resistance (dark).

RESULTS AND COMMENTS

In 2008, 214 isolates of nine different *Candida* species were isolated from blood stream infections and received at the National Mycology Reference Laboratory. In 2007, 207 isolates of eight different *Candida* species were received. All isolates were susceptibility tested for amphotericin B, fluconazole, voriconazole and caspofungin by E-test according to the manufacturers` instructions. From ultimo July 2008, anidulafungin was included in the test panel. *Candida albicans* is still the most common *Candida* species observed (n=140, 65.4%), followed by *Candida glabrata* (n=37, 17.3%) and *Candida tropicalis* (n=13, 6.1%).

All *C. albicans* isolates, with one exception, were susceptible to all antifungal drugs tested. The exception (0.7%) had an MIC of fluconazole of 8 mg/L. Likewise, all *C. tropicalis* isolates, with one exception (16.7%), were susceptible to all drugs tested. The exception had an MIC of 2 mg/L for anidulafungin. However, the number of *C. tropicalis* isolates is small, and calculations may therefore be misleading.

C. glabrata was found to be susceptible to amphotericin B, caspofungin and anidulafungin without exception. However, only five isolates (13.5%) had an MIC ≤ 2 mg/L for fluconazole, while 32 isolates had an MIC > 2 mg/L. Seven isolates (18.9%) had an MIC ≥ 256 mg/L. When testing voriconazole susceptibility, 22 of the isolates (59.5%) had an MIC ≤ 0.125 mg/L. Fifteen isolates (48.5%) had an MIC ≥ 0.25 mg/L and six of these isolates (16.2%) had an MIC of ≥ 32 mg/L. There is no decrease in the number of isolates with low MICs for fluconazole and voriconazole compared to 2007, but among the isolates with higher MICs, there are more isolates with MICs above the highest measurable level. The occurrence of heteroresistent *C. glabrata* increased from two isolates in 2007 to eight isolates in 2008. The European Committee on Antimicrobial Susceptibility Testing (EUCAST) has not determined fluconazole or voriconazole breakpoints for *C. glabrata* due to insufficient data to support the use of the drugs to this yeast. Our findings of consistently higher MIC values to these drugs and the increase of heteroresistant strains do support the notion that fluconazole and voriconazole should not be recommended when treating serious *C. glabrata* infections.

The breakpoints used in yeast susceptibility testing are still under consideration. EUCAST is expected to introduce breakpoints for caspofungin in 2009. In this report the breakpoints recommended by the Norwegian Reference Group on Antibiotic Susceptibility Testing (AFA) are used for amphotericin B. The EUCAST breakpoints have been introduced for fluconazole and voriconazole for some species. No European breakpoint recommendations have been made for caspofungin or anidulafungin, but strains with MICs ≤ 1 mg/l are presumably susceptible.

Resistance in influenza viruses

Background

Two classes of antiviral drugs are being used against influenza virus infection. Whereas M2 blockers inhibit replication of influenza type A viruses, the more recently developed neuraminidase inhibitors (NIs) inhibit the replication of both type A and B. In Norway, only the NIs oseltamivir and zanamivir are approved for the prophylaxis and treatment of influenza; however, both the NI oseltamivir and the M2 blocker rimantadine form parts of the government's stockpile for use against pandemic influenza.

Usage of influenza antivirals in Norway is very sparse, but effective treatment initiated within the first 48 hours is imperative for severe influenza. Type of antiviral must be chosen on empirical grounds and knowledge on the occurrence of resistance at population level is therefore needed. The Department of Virology at the Norwegian Institute of Public Health (NIPH) functions as a WHO National Influenza Centre (NIC) and has been designated by the Ministry of Health (MoH) as national reference laboratory for influenza. In the latter function lies also the obligation to monitor and assess the occurrence of resistance. Historically, resistance has been known to develop quite easily against the M2 blockers Over the last decade, increasing proportions of resistant viruses have been observed, particularly of subtype A(H3N2) (1). During the first years of use, the more recently developed NIs seemed to be much less affected by resistance development and resistant mutants in general have seemed less viable. Locally, the influenza viruses die out at the end of each winter season, and subsequently are reintroduced at the beginning of the next season through global spread of new variants. Consequently, resistance can change abruptly and the monitoring needs to have a strong international dimension. In addition to national monitoring, a selection of influenza viruses shipped by European NICs to the WHO Collaborating Centre in the United Kingdom are routinely passed on to Health Protection Agency (HPA) for antiviral susceptibility testing.

Strengthening of resistance monitoring in Norway

Because stockpiled antivirals form a major part of the Norwegian national preparedness against pandemic influenza, the MoH has tasked the NIPH to closely monitor resistance. For this purpose, genotypic and phenotypic methods have been implemented.

Surveillance findings

Findings from the first years of surveillance are summarised in Table 55. Since the influenza viruses at the end of the year are invariably more closely related to viruses occurring early next year than to the viruses from the preceding spring, it is meaningful to summarise according to winter seasons rather than by calendar years. The findings for M2 blocker resistance is largely in accordance with the global patterns, with high and increasing proportion of resistant viruses. For NIs, resistant viruses have been very rare in most countries; this is also reflected in the Norwegian data up to 2007. However, during the 2007-08 season, an unprecedented proportion of high-level resistance to oseltamivir (but not zanamivir) was found (2,3). This global emergence of resistance was discovered first through analysis of viruses from Norwegian influenza surveillance, and it took place with no association to recorded usage of drug. During the 2008-09 winter season, A(H3N2) viruses that were susceptible to neuraminidase inhibitors but resistant to M2 blockers, predominated. Fortunately, viruses resistant to both NIs and M2 blockers have been extremely rare. Currently, the world is seeing pandemic emergence of a novel influenza A(H1N1) virus that appears to have originated from influenza viruses circulating in pigs. To date, these viruses are uniformly resistant to M2 blockers but with very few exceptions they remain fully susceptible to the neuraminidase inhibitors oseltamivir and zanamivir. Their resistance pattern is being monitored very closely.

TABLE 55. Norwegian influenza viruses resistant to M2 blockers (adamantanes) and the NI oseltamivir, during the influenza seasons 2005/06 to 2008/09.

	Adamantane resistance		Oseltamivir resistance**			Zanamivir resistance		
	A(H1N1)	A(H3N2)	A(H1N1)	A(H3N2)	B	A(H1N1)	A(H3N2)	B
2005/06	nd	75% (n=4)	0% (n=6)	0% (n=13)	0% (n=21)	0% (n=6)	0% (n=13)	0% (n=21)
2006/07	0% (n=6)	90% (n=10)	0% (n=5)	0% (n=10)	nd	0% (n=5)	0% (n=10)	nd
2007/08	0% (n=112)	100% (n=2)	68% (n=272)	0% (n=2)	0% (n=59)	0% (n=114)	0% (n=2)	0% (n=59)
2008/09*	0% (n=5)	100% (n=65)	100% (n=33)	0% (n=13)	0% (n=1)	0% (n=5)	0% (n=12)	0% (n=1)
2009-swH1	100% (n=28)		0% (n=72)			0% (n=20)		

*For the 2008/09 season, also viruses from early 2009 are included. **Two screening tools were used to determine oseltamivir resistance: sequence analysis of viral genes or a neuraminidase inhibition assay.

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Primary HIV resistance in Norway 2006-2008

Background

Internationally there is concern that HIV with primary resistance to antiretroviral drugs may reduce the chances of treatment success. In Norway surveillance of HIV drug resistance among persons newly diagnosed with HIV infection was started in January 2006. The main purpose of this programme is to study transmitted resistance at the population level over time in untreated individuals. We here present preliminary results for the first three years of HIV resistance surveillance in Norway.

Material and methods

Physicians diagnosing patients with previously undiagnosed (and therefore untreated) infection are asked to send plasma samples for HIV resistance testing to one of three laboratories performing this analysis in Norway (Haukeland University Hospital, Oslo University Hospital, Ullevål and Rikshospitalet). After DNA sequencing of relevant areas of the genome coding for HIV resistance, the electronic sequences are sent anonymously to the Norwegian Institute of Public Health. There the sequences are analysed for mutations using the Stanford University HIV Drug Resistance Database (spring 2009), where mutations are classified as coding for high-level, intermediate-level, low-level or potentially low-level resistance. These mutations are cross-checked with International AIDS Society-USA resistance mutations (1), and only mutations listed there are included.

Results

During the period 2006-2008, DNA sequences were received from 332 of 823 (40.3%) patients who could be identified in the anonymous database as newly diagnosed with HIV infection. Preliminary analysis shows that 28 cases (8.4%) had virus with one or more mutations coding for resistance. Six patients (1.8%) had high-level, three (0.9%) had intermediate-level, three (0.9%) had low-level and 16 (4.8%) had potential low-level resistance. Of these, four (1.2%) had resistance mutations for protease inhibitors, four (1.2%) for nucleoside and nucleotide analogue reverse transcriptase inhibitors and 20 (6.0%) for non-nucleoside analogue (NNRTI) reverse transcriptase inhibitors (of which 16 were potential low-level resistant). No isolates with double class resistance were identified.

Discussion and conclusion

The prevalence of primary HIV resistance in Norway was 8.4%. Reports on primary HIV resistance are difficult to compare due to differences in study periods and different definitions of resistance. We identified nine patients (2.7%) with high or intermediate level resistance, and this is in agreement with 3.3 % found in a similar cohort study from the UK (2). In our study, resistance testing was only performed in 40.3 % of newly diagnosed cases, and selection bias is possible. There is a need to improve the surveillance system for primary HIV resistance in order to include a higher proportion of patients with newly diagnosed infection.

Reference List

1. Johnson VA, Brun-Vezinet F, Clotet B, et al. Update of the Drug Resistance Mutations in HIV-1. Top HIV Med 2008; 16: 138-45.
2. Payne BA, Nsutebu EF, Hunter ER, et al. Low prevalence of transmitted antiretroviral drug resistance in a large UK HIV-1 cohort. J Antimicrob Chemother 2008; 62: 464-8.

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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 drugs 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. Number of sold items in 2007 for the various veterinary drug formulations, strengths and package sizes were obtained from the Norwegian Institute of Public Health and calculated to express kg active substance.

Veterinarians have since 1989 been obliged by regulation to submit copies of all prescriptions to farmed fish to the Norwegian Directorate of Fisheries (NDF), and since 2004 to the Norwegian Food Safety Authority (NFSA). NFSA (and formerly NDF) compiles all relevant information from the prescriptions into a prescription database such as the drug substance and the amounts prescribed, fish species to be treated and the date of prescribing. Data on annual usage of antimicrobials per fish species was obtained from this prescription database. These data has since 1996 been regularly validated against overall national sales statistics of drugs sold for use in farmed fish, and this validation shows that the data from these two sources are highly correlated.

Drug classification system

The Anatomical Therapeutic Chemical vet (ATCvet) classification system was used to categorize veterinary medicinal products (http://www.whocc.no/atcvet).

Unit of measurement

The amount of active substance denoted in kilograms was chosen as the unit of measurement. Veterinary antimicrobial drug 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

The veterinary drugs included for terrestrial animals were all the approved veterinary antimicrobial (AM) specialities belonging to the following ATCvet groups: QA07AA (gastrointestinal infections), QG01AA+AX (uterine infections) and, QJ [AM agents for systemic use that includes intramammary dose applicators (QJ51)]. Additionally, a few AMs preparations sold on special exemption from market authorization have been included following a case by case assessment (se footnotes for the various tables and figures). Sales of AMs as medicated feeds and as premixes, both intended for use in farmed fish, belonging to QJ are presented separately. An exemption has been made for an AMs premix approved for farmed fish only (trimethoprim+sulfadiazine 1:5) but sold solely for use in terrestrial animals since 1995 (unpublished data). In the present report, the sales of this premix has for the first time been included in Table 5 that presents detailed sales figures for AMs for terrestrial animals for the latest year; for Fig. 1 and 2 this premix has been included for the whole period. Consequently, the sales of the AM drugs in terrestrial animals reported for the years 1995-2005 were underestimated, although only slightly. However, the updated usage figures for 1995-2005 correlated highly positively (r=0.998) with the data reported previously for these years confirming the formerly reported reduction in the usage of AMs in terrestrial animals. Dermatological preparations (QD) and preparations intended for sensory organs (QS) are not included in the material. Human antimicrobial preparations are used in small animal practice. However, data on the use of such antimicrobial preparations in animals are not included in this report as such sales cannot be separated from sales intended for use in humans.

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

Data sources
In Norway, antibacterials are prescription only medicines, and only allowed sold through pharmacies. This data are collected from three large databases: the Norwegian drug wholesales statistics database, the hospital pharmacies drug statistics database and the Norwegian prescription database, NorPD.

The wholesales database covers total sales of antibacterials in Norway and is based on sales of medicaments from drug wholesalers to pharmacies and hospitals in Norway. The figures presented should be regarded as maximum figures based on the assumption that all medicaments sold are actually consumed. The actual drug consumption will probably be somewhat lower. The Norwegian Institute of Public Health collects the data. Data on drug use from wholesalers has been collected since the beginning of the seventies.

Data on antibacterial use in hospitals are retrieved from *Sykehusapotekenes Legemiddelstatistikk* (the hospital pharmacies drug statistics database): a cooperation of the Norwegian pharmacies delivering drugs to hospitals and LIS (Drug Purchasing Cooperation - Legemiddel Innkjøp Samarbeid). *Sykehusapotekenes Legemiddelstatistikk* collects sales data from each hospital pharmacy. Data are collected as sales to wards/hospitals from the pharmacy.

Data on the use in ambulatory care are retrieved from NorPD, a national prescription database. This database includes all prescriptions being prescribed to out-patients in Norway. These data gives us the exact population prevalence of antibacterials in ambulatory care. The Norwegian Institute of Public Health collects the data.

Drug Classification
The data is categorized according to the ATC classification system. Defined Daily Doses (DDDs) are employed as units of measurement. The ATC/DDD index of 2009 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 antibacterials are as a main rule based on the use in infections of moderate severity. Some antibacterials are only used in severe infections and their DDDs are assigned accordingly. The DDDs assigned are based on daily treatment.

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

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

Sampling strategy
Clinical isolates of beta-haemolysin-producing *Staphylococcus* spp. from dogs included in the NORM-VET monitoring programme 2008 were collected from diagnostic submissions.
The isolates of indicator bacteria, *Escherichia coli*, *Enterococcus* spp. included in 2008 were collected from faecal samples from dogs (*E. coli*) and faecal samples from swine (*E. coli* and *Enterococcus* spp). Faecal samples from healthy dogs were collected from five small animals clinics geographically spread throughout Norway. Each clinic was asked to submit ten samples, four times during the whole year.
The faecal samples from swine were collected within the frame of other surveillance programs. Only one sample from each individual or per production unit was subjected to NORM-VET.

Isolation and identification of bacteria

Escherichia coli
The *E. coli* strains included in NORM-VET 2008 were isolated and identified at the National Veterinary Institute. The samples were plated directly onto the surface of lactose-saccarose-bromthymol blue agar without broth enrichment, incubated at 35-37°C for 24 h, and on MacConkey agar with 1 mg/L cefotaxime, incubated at 35-37°C for 48 h. (Screening for ESBL producing isolates.) Following incubation of the agar plates, 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 strains included in NORM-VET 2008 were isolated and identified at the National Veterinary Institute. The sample was plated directly onto the surface of Slanetz & Bartley agar (Oxoid) without broth enrichment. Following incubation of the agar plates at 44°C for 48h, typical colonies were plated onto blood agar (Heart infusion agar (Difco) with 5% bovine blood). Typical colonies were tested by catalase reaction and *E. faecium* and *E. faecalis* were identified by *ddl*-PCR (Dutka-Malen et al., 1995).

beta-haemolysin-producing *Staphylococcus* spp.
The beta-haemolysin-producing *Staphylococcus* spp. isolates included in NORM-VET 2008 were isolated and identified at the National Veterinary Institute. The swab samples were plated directly onto the surface of blood agar (heart infusion agar (DIFCO) with 5% bovine blood). The plates were incubated at 35-37°C for 16-24 h. Greyish white typical colonies with a beta-haemolytic zone on blood agar were isolated and tested for production of catalase, β-galactosidase, arginindihydrolase and growth on P-agar with acriflavin.

Susceptibility testing
Only one isolate per production unit were tested for antimicrobial susceptibility. Bacterial isolates were tested for antimicrobial susceptibility at the National Veterinary Institute. A broth microdilution method; VetMIC™ (Dept. of Antibiotics, National Veterinary Institute, Sweden) was used for susceptibility testing of all isolates.
Epidemiological cut-off values recommended by the European Committee on Antimicrobial Susceptibility Testing (EUCAST) were used for the substances recommended by EUCAST with the exception of ciprofloxacin for *E. coli.* For the additional antimicrobial agents, included in our national monitoring programme, a cut-off value was defined on the basis of the actual MIC distributions obtained in the NORM-VET programme (see also appendix 6).

Quality assurance systems
The following susceptible bacteria were included as quality controls on a weekly basis, *E. coli* ATCC 25922, *E. faecalis* ATCC 29212, *S. aureus* ATCC 29213. The following resistant bacteria were tested on a regular basis: *E. faecium* CCUG 33829, CCUG 36804, *S. aureus* CCUG 35603. The results were approved according to reference values given by CLSI when available. Additional control strains were included when necessary.
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 (VLQAS Veterinary Laboratories Agency Quality Assurance Unit, Loughborough, England and Community Reference Laboratory, Denmark).

Data processing
Susceptibility data were recorded and processed in WHONET 5.4, 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 MIC-values.

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
Caecal samples from positive flocks in the Norwegian action plan against *Campylobacter* in broilers (www.vetinst.no) were collected. One isolate from each flock were included in the analysis.

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

Campylobacter
A total of 250 human isolates were obtained from clinical specimens. Five regional laboratories submitted the first five independent isolates each month to the National Reference Laboratory for Enteropathogenic Bacteria at the Norwegian Institute of Public Health.

Isolation and identification of bacteria
Isolation and identification of *Salmonella* spp. from animals was carried out at the National Veterinary Institute according to ISO 6579:2002/Amd.1:2007: Annex D: Detection of *Salmonella* spp. in animal faeces and in environmental samples from the primary production stage. Isolation of *Campylobacter* spp. from broiler was carried out by local laboratories. The samples from turkeys and broiler meat were analysed at the National Veterinary Institute. All *Campylobacter* spp. were isolated according to the Nordic Committee on Food Analyses (NMKL) method number 119, with minor modifications. Identification of the isolates was carried out by the National Veterinary Institute.
Isolation and identification of bacteria from humans was performed according to conventional methods described in standard reference literature (e.g. Murray PR & al.: Manual of Clinical Microbiology, 8th edition ASM-Press, Washington 2003 and Ewing WH: Edwards and Ewing's Identification of *Enterobacteriaceae*, 4. edition, Elsevier, New York 1986). The identification of all isolates from animals and humans was verified at the National Reference Laboratory for Enteropathogenic Bacteria at the Norwegian Institute of Public Health.

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

Salmonella spp., *Yersinia* spp. and *Shigella* spp. isolates from humans were susceptibility tested at the Norwegian Institute of Public Health by an agar disk diffusion test using BD Sensi-Disc and Mueller-Hinton II-medium. The *Campylobacter* isolates from humans were tested for antimicrobial susceptibility at the Norwegian Institute of Public Health using Etest (AB Biodisk).

For animal isolates, epidemiological cut-off values recommended by the European Committee on Antimicrobial Susceptibility Testing (EUCAST) were used (http://www.escmid.org). When no cut-off value was available, or the range of concentrations tested was inappropriate for the recommended value, a cut-off value was defined on the basis of the actual MIC distributions obtained in the NORM-VET programme. The same approach was used when recommended cut-off values would have divided MIC distributions in a manner not in agreement with the concept of wild type distributions and thereby causing an erroneously high frequency of resistance in (a) single year(s).

For human isolates, MIC breakpoints defined by AFA (Norwegian Reference Group on Antibiotic Susceptibility Testing) were applied. For disk diffusion results, population based breakpoints were used. Breakpoints for *Campylobacter* spp. are based on MIC distributions.

Quality assurance systems
The National Veterinary Institute and the Reference Laboratory for Enteropathogenic Bacteria/NIPH 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 strains at the National Veterinary Institute on a weekly basis, at the Reference Laboratory at NIPH *Campylobacter jejuni* subsp. *jejuni* CCGU 11284 is used. The National Veterinary Institute participates in an external quality assurance programme for veterinary pathogens organized by the VLQA (Veterinary Laboratories Agency Quality Assurance Unit, Loughborough, England) and also in the external quality assurance programmes organized by ARBAO-II http://www.dfvf.dk/. The Norwegian Institute of Public Health participates in the external quality assessment programme for *Salmonella* spp. and antimicrobial susceptibility testing, in 2008 still organized by Enter-Net.

Data processing
Susceptibility data were recorded and processed in WHONET 5.4, a program developed by the World Health Organization (WHO) for analysis of resistance data (http://www.who.int/drugresistance/whonetsoftware/). The susceptibility data were stored as discrete values (MIC).

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

General considerations
NORM is based upon periodic sampling and testing in each participating laboratory of microbial isolates from defined clinical conditions such as respiratory tract infections, wound infections, urinary tract infections, or septicaemiae. For enteric infections see Appendix 4. 2008 was the nineth year of surveillance, and all 22 laboratories in Norway participated in the surveillance system in addition to the Norwegian Institute of Public Health. All laboratories followed the same sampling strategy and used identical criteria for the inclusion of microbial strains. Only one isolate per patient and infectious episode was included. All microbes 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 isolates were included for defined time periods for each surveillance category. The surveillance categories and sampling periods in 2008 were as follows: *E. coli* in blood cultures (6 months); *Klebsiella* spp., *Staphylococcus aureus*, *Streptococcus pneumoniae* and *Enterococcus* in blood cultures (9 months); *Candida* spp. from blood cultures (12 months), *S. pyogenes* from wound specimens (3 weeks) and respiratory tract specimens (2 weeks), *Moraxella catarrhalis* from respiratory tract specimens (3 weeks); *S. aureus* from wound samples (1 week); *E. coli* from urinary tract infections (2 days); and *Mycobacterium tuberculosis* from all samples (12 months). All *Enterobacter cloacae* isolates from 2006, 2007 and the first 9 months of 2008 were also included.

Susceptibility testing
E. coli, *Klebsiella* spp., *E. cloacae*, *M. catarrhalis, Enterococcus* spp., *S. aureus* isolates were examined by disk duffusion using either Oxoid disks on Isosensitest agar, or Beckton Dickinson disks on Mueller Hinton II agar with nutritional additives when specified by the manufacturers. Suitable antibiotics were selected for each bacterial species, and the results were interpreted according to the respective manufacturers´ recommendations using the breakpoints of the Norwegian Reference Group on Antibiotic Susceptibility Testing (AFA). The AFA breakpoints are harmonized with EUCAST breakpoints with few exceptions. All *S. aureus* isolates were tested for beta-lactamase production by nitrocefin disks, acidometric agar plates (3.6 mg/L penicillin G and phenol red) or the clover leaf test. All *S. aureus* and *Enterococcus* spp. isolates were screened for glycopeptide resistance using the vancomycin 6 mg/L BHI agar. *S. pneumoniae* and *S. pyogenes* isolates were susceptibility tested using Etest on MH II agar supplemented with 5% lysed sheep blood AB Biodisk (Solna, Sweden). All resistance values were recorded either as mm inhibition zone sizes or MIC values in order to monitor trends in the occurrence of resistance.

Confirmation of resistance phenotypes
E. coli and *Klebsiella* spp. with reduced susceptibility to 3rd generation cephalosporins were examined for ESBL production using the ESBL Etest according to the instructions of the manufacturer. ESBL positive strains were subjected to PCR and DNA sequencing for determination of ESBL genotype. *S. aureus* isolates with reduced susceptibility to cefoxitin were examined by *mecA* PCR for confirmation of methicillin resistance (MRSA). *Enterococcus* spp. isolates displaying growth on the vancomycin screening agar were examined by *van* gene PCRs for confirmation of VRE. Erythromycin resistant *S. pneumoniae* and *S. aureus* isolates were analysed for determination of MLS phenotype using the double disk diffusion (DDD) synergy assay with erythromycin and clindamycin disks.

Quality control
The following strains were used for quality control: *E. coli* ATCC 25922, *K. pneumoniae* ATCC 700603 (ESBL positive), *E. faecalis* ATCC 29212, *E. faecalis* ATCC 51299, *S. pneumoniae* ATCC 49619, *S. aureus* ATCC 29213, *S. aureus* ATCC 25923, *S. aureus* ATCC 43300 (heterogeneous MRSA), *S. aureus* CCUG 35600 (homogeneous MRSA).

Data processing
The specially designed eNORM computer program was used for the registration of patient data, sample data and resistance data. The results were further analysed by WHONET5.3 with the aid of the NORMlink program, both developed by John Stelling. The distribution of microbial 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 were considered duplicates and omitted from the survey. No attempt was made to evaluate the clinical significance of each finding.

Mycobacterium tuberculosis
Susceptibility testing (DST) was performed at the Norwegian Institute of Public Health, Ullevål University Hospital and Rikshospitalet. All isolates were tested using the BACTEC 460 or BACTEC MGIT 960 systems. All three laboratories participate in the WHO external DST quality control program. The same three laboratories and Haukeland University Hospital also perform tests for mutations in *rpoB* gene to detect resistance to rifampicin.

Yeasts
All systemic yeast isolates in Norway are submitted to Rikshospitalet, Oslo. Susceptibility testing on *Candida* spp. isolates was performed by Etest using RPMI agar containing 2% glucose and MOPS. *C. albicans* ATCC 10231 was used for quality control.

Appendix 6: Breakpoints NORM-VET

The substances for the monitored bacterias for which there are recommended epidemiological cut-off values (EUCAST) were mainly applied, marked in the table in bold. For the additional antimicrobial agents included in our national monitoring programme, a cut-off value was defined on the basis of the actual MIC distributions obtained in the NORM-VET programme. The cut-off value recommended by EUCAST for ciprofloxacin in *E. coli* was not applicable to the distributions of MICs in our laboratory and it was therefore decided to use the above approach also for this antimicrobial agent and bacteria.

Antimicrobial	Resistant (MIC values, mg/L)	*Campylobacter jejuni*	*Salmonella* spp.	*Escherichia coli*	*Staphylococcus* spp.	*Enterococcus* spp.
Tetracycline**	**> 2**	■			■	■
	>4					
	> 8		■	■		
Chloramphenicol**	**> 16**		■	■	■	
	>32					■
Florfenicol	> 16		■	■		
Ampicillin	**>4**		■			■
	> 8			■		
Oxacillin	>1				■	
Cephalothin	>1				■	
Ceftiofur	> 1			■		
Cefotaxime	**>0.25**			■		
	>0.5		■			
Trimethoprim**	**> 2**		■	■		
	>8				■	
Sulfonamides	**> 256**		■	■		
Erythromycin**	**> 2**				■	
	> 4	■				■
Clindamycin	>2				■	
Streptomycin	**>2**	■				
	> 16			■		
	> 32		■			
	>128					■ [a]
	> 512					■ [a]
Gentamicin**	**> 1**	■				
	>2		■	■	■	
	> 32					■
Kanamycin	>16		■	■	■	
	> 1,024					■
Ciprofloxacin**	**>0.06**		■	■#		
	>0.5				■	
	>1	■				
Nalidixic acid*	**> 16**	■	■	■		
Vancomycin	> 4					■
Fusidic acid	> 0.5				■	
Bacitracin**	> 32					■
Linezolid	**> 4**					■
Virginiamycin***	> 4					■
Narasin	> 2					■

In bold: Antimicrobial agents and epidemiological cut-off values recommended by EUCAST

[a] >128 mg/L for *E. faecium*, >512 mg/L for *E. faecalis*. * Not included in the recommendation by EUCAST for *Campylobacter jejuni*. ** units, *** applies only for *E. faecium,* **** No recommendation by EUCAST for beta-haemolysin-producing *Staphylococcus* spp.. # for *E. coli* used on the MIC-distribution, not as recommeded by EUCAST for *E. coli*

Appendix 7: Breakpoints NORM

NORM data are categorized according to the breakpoints of the Norwegian Reference Group on Antibiotic Susceptibility Testing – AFA (isolates from humans) which are harmonized with EUCAST breakpoints when available. For details regarding bacteria and antimicrobial panels, see tables in text. AFA breakpoints are available at www.antibiotikaresistens.no.

	MIC values mg/L															
Antimicrobials	S	R	*Escherichia coli*	*Klebsiella* spp.	*Enterobacter cloacae*	*Salmonella* spp.	*Yersinia enterocolitica*	*Shigella* spp.	*Campylobacter* spp.	*Moraxella catarrhalis*	*Staphylococcus aureus*	*Enterococcus* spp.	*Streptococcus pneumoniae*	*Streptococcus pyogenes*	*Candida* spp.
Amphotericin B	≤ 1	> 1													■
Ampicillin	≤ 0.5	> 8	■			■	■	■							
	≤ 4	> 8										■			
Anidulafungin	≤ 1	> 1													■
Caspofungin	≤ 1	> 1													■
Cefotaxime	≤ 0.5	> 2											■		
	≤ 1	> 2	■	■	■										
Ceftazidime	≤ 1	> 8	■	■	■										
Cefuroxime	≤ 0.5	> 1											■		
	≤ 0.5	> 8	■	■	■										
Chloramphenicol	≤ 8	> 8				■	■	■							
Ciprofloxacin	≤ 0.5	> 0.5								■					
	≤ 0.5	> 1	■	■	■	■	■	■	■						
	≤ 1	> 1									■				
Clindamycin	≤ 0.25	> 0.5									■				
	≤ 0.5	> 0.5											■	■	
Ertapenem	≤ 0.5	> 1	■	■	■										
Erythromycin	≤ 0.25	> 0.5								■			■	■	
	≤ 1	> 2									■				
	≤ 4	> 4							■						
Fluconazole	≤ 2	> 2													■
Fusidic acid	≤ 1	> 1									■				

Antimicrobials	MIC values mg/L S	R	*Escherichia coli*	*Klebsiella* spp.	*Enterobacter cloacae*	*Salmonella* spp.	*Yersinia enterocolitica*	*Shigella* spp.	*Campylobacter* spp.	*Moraxella catarrhalis*	*Staphylococcus aureus*	*Enterococcus* spp.	*Streptococcus pneumoniae*	*Streptococcus pyogenes*	*Candida* spp.
Gentamicin	≤ 1	> 1									■				
	≤ 2	> 4	■	■	■				■						
	≤ 128	> 128										■			
Linezolid	≤ 4	> 4									■	■			
Mecillinam	≤ 2	> 8	■												
Nalidixic acid	≤ 16	> 16	■	■	■	■	■	■	■						
Nitrofurantoin	≤ 64	> 64	■												
Penicillin G	≤ 0.064	> 2											■		
	≤ 0.25	> 0.25												■	
Pip./Tazo.*	≤ 8	> 16	■	■	■										
Rifampicin	≤ 0.06	> 0.5									■				
Tetracycline	≤ 1	> 1							■[#]						
	≤ 1	> 2								■	■		■	■	
	≤ 4	> 8				■[#]	■[#]	■[#]							
Tigecycline	≤ 0.5	> 1	■	■	■										
Trimethoprim	≤ 2	> 4	■												
TMS*	≤ 1	> 2											■	■	
	≤ 2	> 4	■	■	■	■	■	■			■				
Vancomycin	≤ 4	> 4									■	■			
Voriconazole	≤ 0.125	> 0.125													■

* TMS Trimethoprim-sulfamethoxazole. Breakpoints for the trimethoprim-sulfamethoxazole combination are given for the trimethoprim component only.
[#] Epidemiological cut-off value based on the wild-type distribution by EUCAST.