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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 issued in 2000.

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I. INTRODUCTION

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

Several countries have implemented monitoring programmes for antimicrobial resistance and antimicrobial usage in recent years. Some programmes focus primarily on human consumption and resistance in human pathogens, whereas others also include data concerning veterinary medicine and food production. The EU supported this broad approach in 1998 through the Copenhagen recommendations and at later follow-up conferences in Sweden, Belgium, Luxembourg and Italy. The World Health Organization (WHO), the Food and Agriculture Organization (FAO) and the World Animal Health Organization (OIE) have through several expert consultations emphasised the importance of monitoring antimicrobial usage and resistance in both human and veterinary medicine and have published several reports and recommendations in this regard.
In response to the growing concern about antimicrobial resistance, the Norwegian Ministry of Health and Social Affairs issued a national action plan against antimicrobial resistance (2000–2004) in March 2000. Again, the importance of monitoring both the human and veterinary sectors, including food production, was emphasized. The action plan recognized the need for ongoing surveillance as a fundamental component of the strategy for containment of antimicrobial resistance. The NORM and NORM-VET programmes were consequently established in order to provide and present microbiologically and epidemiologically valid data on the occurrence and distribution of antimicrobial resistance over time.

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

This report, which is the fourth annual joint report from NORM and NORM-VET, presents data for 2003. In addition to resistance data, the NORM/NORM-VET reports present data on the usage of antimicrobial agents in humans and animals in Norway. The present report, together with earlier reports and reports from the years to come form a basis for the detection, interpretation and evaluation of trends regarding antimicrobial usage and occurrence of resistance in Norway. The NORM and NORM-VET programmes are valuable tools for setting policies, assessing risks and evaluating interventions.

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Tromsø / Oslo, September 2004

II. SAMMENDRAG

NORM og NORM-VET programmene er en del av regjeringens tiltaksplan mot antibiotikaresistens som ble offentliggjort i 2000. Norsk overvåkingssystem for antibiotikaresistens hos mikrober (NORM) overvåker forekomsten av antibiotikaresistens blant sykdomsfremkallende bakterier fra mennesker. NORM ble etablert i 1999 og koordineres av Mikrobiologisk Avdeling, Universitetssykehuset i Tromsø. NORM-VET har tilsvarende oppgaver innen veterinærmedisin og matproduksjon og ble etablert i 2000. Norsk zoonosesenter ved Veterinærinstituttet koordinerer NORM-VET. De to programmene har et godt samarbeid og utgir blant annet en felles årsrapport. Den foreliggende rapporten presenterer data for året 2003 og er den fjerde årsrapporten fra NORM/NORM-VET. Årsrapportene gir i tillegg til resistensdata en oversikt over forbruket av antibiotika til mennesker og dyr. Rapportene brukes også til å formidle data fra relevante prosjekter selv om disse ikke er en del av den opprinnelige planen for overvåkingsprogrammene.

Forbruk av antibiotika til dyr

Forbruket av antibiotika i norsk husdyrproduksjon og akvakultur er lavt. Det norske totalsalget av antibiotika godkjent for terapeutisk bruk til landdyr var 5 787 kg i 2003. Fra 1995 til 2001 var det en reduksjon på 40% av slik bruk. Etter dette har forbruket vært relativt konstant. I tillegg er forbruksmønsteret gunstig. β-laktamase-følsomme penicilliner utgjorde 84% av totalsalget av rene penicillinpreparater i 2003.

Forbruket av antibiotika i oppdrettsnæringen har blitt redusert med 98% de siste 15 årene. Denne betydelige reduksjonen kan tilskrives innføringen av effektive vaksiner og bedrede miljøforhold i oppdrettsnæringen. Totalsalget av veterinære antibiotika godkjent for terapeutisk bruk til oppdrettsfisk i Norge var 805 kg i 2003. Kinoloner utgjorde 75% av dette salget.

Avoparcin ble brukt som antibiotisk vekstfremmende fôrtilskudd 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 vekstfremmere brukes fremdeles i norsk fjørfeproduksjon. Salgstallene i kg aktiv substans er på samme nivå som før forbudet mot vekstfremmere, men forbruksmønstret er forandret. Narasin har dominert siden 1996, mens bruken av andre ionofore koksidiostatika har sunket tilsvarende.

Forbruk av antibiotika hos mennesker

Totalsalget av antibiotika til systemisk bruk hos mennesker var 17,1 DDD/1000 innbyggere/dag i 2003 hvilket er uendret sammenliknet med 2002. Det samlede forbruk av antimikrobielle midler har vært stabilt over mange år, men det har skjedd endringer i fordelingen mellom undergrupper. Salget av cefalosporiner, makrolider og kinoloner har økt, salget av penicilliner er stabilt, mens forbruket av tetracykliner og sulfonamider og trimetoprim har sunket. Disse trendene har vært observert gjennom flere år og fortsatte også i 2003.

I 2003 utgjorde penicilliner 42% av det totale antibiotikaforbruket i Norge. Salget av penicilliner har vært stabilt over mange år. Det har imidlertid skjedd en forskyvning mot mer bredspektrede penicilliner. Forbruket av β-laktamase følsomme penicilliner gikk ned 2% i 2003. Salget av benzylpenicillin, som hovedsakelig brukes i sykehus, var stabilt. Nedgangen er følgelig størst for fenoxymethylpenicillin, et peroralt penicillinpreparat som hovedsakelig brukes i primærhelsetjenesten. Forbruket innen subgruppene av bredspektrede penicilliner og β-laktamase stabile penicilliner ble redusert med henholdsvis 2,7% og 18%.

Tetracyklinene utgjorde 18% av totalforbruket. Salget har vært synkende siden 1993 da det høyste forbruket noensinne ble registrert. Makrolidene utgjorde 10% av totalforbruket i 2003. Salget var forholdsvis stabilt gjennom nittitallet, men for 2001 og 2002 ble det registrert en økning på 24% sammenliknet med 2000. Denne trenden fortsatte ikke i 2003. Fordelingen mellom ulike makrolider er heller ikke blitt endret. Forbruket av linkosamidene, som i Norge er representert av klindamycin, har økt med 90% siden 1997 og 19% siden 2002. Salget av cefalosporiner, monobaktamer og karbapenemer er begrenset men økende. Det utgjorde 4,4% av det totale antibiotikasalget i 2003. Fordelingen mellom undergruppene har endret seg siden 1996. Førstegenerasjons cefalosporiner (cefalexin og cefalotin) utgjorde 67% av salget i 1996 og 60% i 2003. Det har skjedd en forskyvning mot tredjegenerasjons cefalosporiner og karbapenemer. Salget av sulfonamider og trimetoprim har sunket med 26% siden 1997. Det har vært en liten men stabil økning i forbruket av kinoloner. Gruppen utgjør fortsatt kun 2,8% av det samlede antibiotikaforbruket, men dette er en økning på 71% siden 1997.

Forskrivningsmønsteret for antibiotika er forskjellig på sykehus og i primærhelsetjenesten. Antibiotikasalget på sykehus utgjorde 8% av totalforbruket målt i definerte døgndoser (DDD). Penicilliner utgjorde rundt 48% av forbruket på sykehus og 41% i primærhelsetjenetsen. De andre viktige antibiotikagruppene i primærhelsetjenetsen var tetracykliner (18%), og makrolider og linkosamider (12%). På sykehus var cefalosporiner (17%) den viktigste gruppen etter penicilliner, etterfulgt av peroralt og parenteralt metronidazol (6%) og kinoloner (5%).

Forbruket av antibiotika utenfor sykehus utgjorde 92% av det samlede forbruket i 2003. I Norge kan antibiotika kun utleveres mot resept. Fra 1 januar 2004 er det etablert en nasjonal reseptdatabase. Data fra dette registeret vil bli tilgjengelige fra 2005.

Resistens hos kliniske isolater fra dyr

De kliniske isolatene inkludert i 2003 var fra diagnostiske prøver fra mastitt hos storfe, sau og geit (*Staphylococcus aureus* og koagulase negative stafyloklokker (KNS)) og er sannsynligvis vinklet mot problematiske tilfeller særlig når det gjelder storfe. Forekomsten av resistens blant *Staphylococcus aureus* og KNS fra mastitt hos ku har vært stabil siden begynnelsen av 1990-tallet. I 2003 var henholdsvis 91,5% og 28%, av slike isolater følsomme for

alle antibiotika som inngikk. Forekomsten av resistens blant *S. aureus* fra mastitt hos sau og geit var på samme lave nivå som for *S. aureus* fra mastitt hos ku. Henholdsvis 95,1% og 91,7% var følsomme for alle antibiotika som inngikk. Resistens ble hyppigst observert mot antibiotika som brukes terapeutisk hos disse produksjonsdyrene; penicillin, trimetoprim og streptomycin. Resultatene av resistenstesting av totalt 100 isolater av *Moritella viscosa* fra vintersår hos atlantisk laks (*Salmo salar*) (før første gang inkludert i NORM-VET i 2003) indikerer full følsomhet ovenfor alle antibiotika som inngikk.

Resistens hos indikatorbakterier

Forekomsten av ervervet antibiotikaresistens blant 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 2003 ble indikatorbakterier fra fecesprøver fra storfe og sau samt kjøttprøver fra storfe inkludert.

Resultatene fra 2003 indikerer en relativ moderat forekomst av resistens blant *E. coli* og *Enterococcus* spp. fra storfe og blant *E. coli* fra villrein, mens forekomsten av resistens blant *E. coli* og enterokokker fra småfe er lav. Forekomsten av resistens blant indikatorbakterier fra storfe og sau er på samme nivå som i 2001, og resistens ble kun observert for vanlig brukte antibiotika til storfe og sau.

Resistens hos zoonosebakterier og andre enteropatogene bakterier

Salmonella spp., med unntak av *S. enterica* subsp. *diarizonae* fra sau, påvises sjelden hos matproduserende dyr i Norge. I 2003 ble i alt 14 *Salmonella*-isolater resistenstestet. Alle utenom ett isolat, et multiresistent isolat fra hest (*S.* Typhimurium), var følsomme for alle antibiotika som inngikk.

Av de humane salmonellosetilfellene som ble rapportert i 2003, var 80% oppgitt å ha blitt smittet i utlandet. Forekomsten av multiresistent DT104-infeksjon, spesielt antall tilfeller smittet innenlands, ble redusert markant i 2003 sammenlignet med 2002.

Andelen *S.* Typhimurium isolater (unntatt DT104) som var følsomme for alle antibiotika, var høyere for kategorien "smittet i Norge" (88,2%) enn for kategorien "smittet i utlandet" (59,4%). Multiresistens ble hyppigere påvist i sistnevnte kategori (19,8%) enn førstnevnte (5,3%).

De aller fleste av *S.* Enteritidis-isolatene var fra pasienter smittet utenlands. Nalidiksinsyreresistens var mer utbredt blant *S.* Enteridis (26,6%) enn *S.* Typhimurum (10,1% i kategorien "smittet i utlandet" og 2,5% i kategorien "smittet i Norge"). Av de nalidixinsyre-resistente *S.* Enteritidis isolatene viste 6,9% redusert følsomhet for ciprofloxacin, hvilket indikerer mulig utvikling av fluorokinolonresistens.

Resultatene fra 2003 viser at forekomsten av resistens hos *Campylobacter jejuni* fra norske broilere er lav. Totalt 95% av isolatene var følsomme for alle antibiotika som inngikk, og kun ett isolat (1,4%) var resistent mot fluorokinoloner. Tilsammen 3,6% var resistente mot ett antibiotikum (ampicillin eller oxytetracyklin) og 1,4% var resistent mot oxytetracyklin, nalidixinsyre og fluorokinolonet enrofloxacin. Resultatene stemmer overens med det som er blitt presentert i tidligere NORM/NORM-VET-rapporter.

Nivået av resistens og resistensmønstrene for *C. jejuni* fra norske broilere samsvarer med *C. jejuni* fra mennesker smittet i Norge med unntak av en høyere forekomst av kinolonresistens, særlig nalidiksinsyre, blant humanisolatene. Dette forholdet ble også påvist i NORM/NORM-VET 2001 og 2002.

Resistens var betydelig mer utbredt blant *C. jejuni* fra pasienter smittet i utlandet (86% resistente mot minst ett antibiotikum) enn pasienter smittet i Norge (16%). Fluorokinolonresistens var mer vanlig blant isolater fra pasienter smittet utenlands enn innenlands smittede; 70,8% mot 7,9%.

De aller fleste *Shigella*-isolatene var fra pasienter smittet utenlands. I likhet med hva som rapporteres fra andre land, var resistens utbredt. Forekomsten av resistens var særlig utbredt for trimetoprim/sulfa (>65%) og for tetracyklin (>58%). Fluorokinolonresistens var lite utbredt og ble kun observert blant *S. flexneri* (2,3%). Imidlertid var en betydelig andel av isolatene av *S. flexneri* (14%) og *S. sonnei* (18,9%) nalidiksinsyreresistente, hvorav mange også viste redusert følsomhet for ciprofloxacin, noe som indikerer mulig utvikling av fluorokinolonresistens.

Resistens hos kliniske isolater fra mennesker

Forekomsten av antibiotikaresistens i kliniske isolater fra mennesker i Norge var fortsatt meget lav i 2003, og det ble kun påvist mindre endringer fra 2002 til 2003. Fire av 637 *Staphylococcus aureus* blodkulturisolater (0,6%) og tre av 1144 *S. aureus* isolater fra sårprøver (0,3%) ble verifisert som methicillinresistente (MRSA) ved *mecA* og *nuc* PCR. Dette står i kontrast til den betydelige økningen i antall kliniske tilfeller av MRSA-infeksjon som ble rapportert til Meldesystemet for infeksjonssykdommer (MSIS) fra 143 tilfeller i 2002 til 216 tilfeller i 2003. Uoverensstemmelsen har ingen åpenbar forklaring. Kun en liten andel av tilfellene er invasiv sykdom med bakteriemi, og det vil derfor kunne ta noe tid før en økende tendens vil kunne påvises i blodkulturisolater. Man kan også tenke seg at MRSA-stammene som sirkulerer i Norge har virulensegenskaper for etablering av bløtdelsinfeksjoner uten å forårsake systemisk sykdom. Både den epidemiologiske og den mikrobiologiske overvåkingen av MRSA vil bli videre utviklet i 2004 og forhåpentligvis gi svar på disse spørsmålene. Forekomsten av fusidinresistens i *S. aureus* blodkulturisolater sank fra 8,0% i 2002 til 6,1% i 2003. Selv om forekomsten av fusidinresistens fortsatt er høy i isolater fra sårprøver (23%) var dette kun en beskjeden økning fra 2002 (20,8%). Man kan derfor mistenke at spredningen av denne resistenstypen nå er avtagende.

Blodkulturisolater av *E. coli* og *Klebsiella* spp. var generelt følsomme for bredspektrede antibiotika inkludert ceftazidim, cefpirom, meropenem, ciprofloxacin og piperacillin/tazobactam. Tre av 966 *E. coli* isolater (0,3%) og ett av 299 *Klebsiella* spp. isolater (0,3%) produserte β-laktamaser med utvidet spektrum (ESBL). Forekomsten av nedsatt følsomhet for og resistens mot kinoloner er lav men muligens økende. Prevalensen av nedsatt følsomhet for og resistens mot ciprofloxacin var 5,1% i *E. coli* og 8,3% i *Klebsiella* spp. Den reduserte følsomheten for kinoloner i disse bakterieartene gjenspeiler økningen i

forbruket av ciprofloxacin fra 0,29 DDD/1000 innbyggere/dag i 2000 til 0,42 DDD/1000 innbyggere/dag i 2003.

Det ble bare funnet en enkelt *Enterococcus faecium* stamme med VanB resistens (VRE) blant 252 blodkulturisolater av enterokokker (0,4%). Det ble imidlertid påvist en økning i prevalensen av høygradig gentamicinresistens fra 4,5% i 2001 til 9,6% i 2002 og videre til 14,2% i 2003. En liknende utvikling ble ikke observert i *E. faecium*. Den økende forekomsten av høygradig gentamicinresistens i enterokokker er bekymringsverdig da den setter i fare det tradisjonelle β-laktam/aminoglykosid kombinasjonsregimet som brukes ved sepsisbehandling i mange norske sykehus.

Streptococcus pneumoniae isolater fra blodkulturer var generelt følsomme for alle relevante antibiotika. Bare tre av 514 blodkulturisolater (0,6%) og 21 av 752 luftveisisolater (2,8%) hadde nedsatt følsomhet for penicillin G. Ingen pneumokokkisolater var resistente mot penicillin G eller cefalosporiner. Forekomsten av nedsatt følsomhet for og resistens mot erytromycin økte imidlertid til 6,0% i blodkulturisolater og 5,7% i luftveisisolater. Dette skyldes tilsynelatende videre spredning av kloner med lavgradig resistens mot erytromycin og følsomhet for klindamycin.

Moraxella catarrhalis ble inkludert for første gang i NORM i 2003, og dette er såvidt vi vet den første systematiske undersøkelsen av resistensforhold i denne bakteriearten i Norge. I samsvar med funnene fra andre land var det store flertallet av isolatene β-laktamaseproduserende (92,3%). Resultatene viste videre utbredt følsomhet for tetracykliner, makrolider, trimetoprim/sulfamethoxazol, kinoloner og bredspektrede β-laktamer. I tillegg ble det samlet verdifull informasjon om analysemetoder og egnede brytningspunkter.

Det ble ialt rapportert 339 tilfeller av tuberkulose til MSIS i 2003, og 320 var ikke tidligere blitt behandlet med tuberkulostatika. Det ble utført resistensundersøkelse av 272 *Mycobacterium tuberculosis* isolater. Ett enkelt isolat hadde kombinert resistens mot rifampicin (R) og isoniazid (H) og var følgelig multiresistent etter vanlige definisjoner. Isolatet var imidlertid følsomt for andre førstelinjepreparater. Resistensundersøkelse ble også utført på bakteriestammene fra elleve pasienter som tidligere hadde blitt behandlet med tuberkulostatika. To isolater fra Øst-Europa var multiresistente (MDR).

Konklusjon

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

III. SUMMARY

The NORM and NORM-VET programmes are a part of the Norwegian Government's Action Plan against Antimicrobial Resistance issued in 2000. The NORM surveillance programme for antimicrobial resistance in human pathogens was established in 1999. It is coordinated by the Department of Microbiology at the University Hospital of North Norway in Tromsø. The NORM-VET monitoring programme for antimicrobial resistance in the veterinary and food production sectors was established in 2000. The Norwegian Zoonosis Centre in Oslo is the coordinator of NORM-VET. Successful collaboration has been established between NORM and NORM-VET and a joint report is issued annually. The current report, which is the fourth joint report, presents data for the year 2003. In addition to data on antimicrobial resistance, the NORM/NORM-VET reports include data on usage of antimicrobial agents in humans and animals. The joint report also presents data from specific surveys or projects that are not part of the continuous monitoring through NORM or NORM-VET.

Use of antimicrobial agents in animals

The use of antimicrobials in Norwegian animal production and aquaculture is low. In 2003, the total sale of antimicrobial drugs approved for therapeutic use in animals (excluding fish) in Norway was 5,787 kg. The annual use of veterinary antimicrobial drugs decreased gradually by 40% from 1995 to 2001. This figure has remained on a relatively constant level since. Furthermore, the patterns of use are favourable. β-lactamase sensitive penicillins accounted for 84% of the veterinary penicillin preparations sold in 2003.

The usage of antimicrobials in aquaculture has decreased by 98% over the last 15 years. This significant decrease in antimicrobial use is mainly attributed to the introduction of effective vaccines and improved health management in Norwegian aquaculture. In 2003, the total sale of antimicrobial drugs for therapeutic use in farmed fish was 805 kg of active substance. Quinolones accounted for 75% of this.

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

Coccidiostatic growth promoters are still used in Norwegian poultry production. The total sales of coccidiostats, in kilograms of active substance, are at the same level as before the ban on antimicrobial growth promoters was implemented. However, the pattern of usage has changed. The usage of coccidiostats has been dominated by narasin since 1996, while the usage of other ionophores has decreased correspondingly.

Use of antimicrobials in humans

In 2003, the overall sales of antimicrobials for systemic use in humans represented 17.1 DDD/1,000 inhabitants/day, the same as in 2002. Total sales of antimicrobials have remained relatively unchanged for many years. However, within subgroups of antimicrobials, usage trends have changed. The subgroups of cephalosporins, macrolides and quinolones are steadily increasing, the penicillin group is stable, while the subgroups of tetracyclines and the sulphonamides and trimethoprim are decreasing. These are trends that have been observed over years and the sales in 2003 follow the same pattern.

In 2003, the penicillins represented 42% of the total antimicrobial use in Norway. The sales of penicillins have been stable over years. It has, however, been a shift towards more broad-spectrum penicillins. β-lactamase sensitive penicillins decreased by 2% in 2003. The sales of benzylpenicillin, mainly used in hospitals, were stable; hence, the decrease was due to phenoxy-methylpenicillin, a peroral penicillin formulation mainly used in ambulatory care. The subgroups of penicillins with extended spectrum and β-lactamase-resistant penicillins are both increasing, by 2.7% and 18%, respectively.

The tetracyclines represent 18% of total use. The sales have been decreasing since 1993, when the highest sale ever was registered. The macrolides represents 10% of total use in 2003. Sales have been fairly stable through the nineties. In 2001 and 2002 an increase of 24% compared to 2000 was found. However, this trend was not continued in 2003. The internal macrolide pattern has remained unchanged. The lincosamides, in Norway represented by clindamycin, is increasing, by 90% since 1997 and by 19% since 2002. Sales of cephalosporins, monobactams and carbapenems, although limited, have also been increasing, now representing 4.4% of the total sales of antimicrobials. The internal subgroup pattern has changed since 1996. Mainly 1st generation cephalosporins are used i.e. cefalexin and cefalotin, representing 67% of the cephalosporins in 1996 and 60% in 2003. Over the years, there has been a shift towards using more 3rd generation cephalosporins and carbapenems. The sales of sulfonamides and trimethoprim have decreased by 26% since 1997. There has been a small, but stable increase in quinolone use. It still represents only a minor fraction (2.8%) of total antimicrobials sales, but the increase has been 71% since 1997.

The therapy pattern of antimicrobials in hospitals differs from the sale in ambulatory care. The antimicrobials sales in DDDs to hospitals represented, in 2003, eight percent of total sales in the country. Penicillins represent around 48% and 41% of the use in hospitals and in ambulatory care, respectively. The most important other groups in ambulatory care are tetracyclins (18%) and macrolides and lincosamides (12%). In hospitals cephalosporins (17%) is the most used group after the penicillins, followed by metronidazole - oral and parenteral (6%) and the quinolones (5%).

The use of antimicrobials outside hospital represents 92% of the total human sale of antimicrobials. In Norway antibiotics are prescription-only drugs. From 1 January 2004, a national prescription database has been established. Data from this register will be available in 2005.

Resistance in animal clinical isolates

The clinical isolates included in 2003 were from diagnostic samples from mastitis in cattle, sheep and goat (*Staphylococcus aureus* and coagulase negative staphylococci (CoNS)) and are probably biased towards problematic cases, at least for cattle.

The prevalence of resistance in *S. aureus* and in CoNS from mastitis in cattle has remained at the same level since the 1990s. In 2003, 91.5% and 28% of the isolates, respectively, were susceptible to all antimicrobials included. The prevalence of resistance in *S. aureus* from sheep and goat were at the same low level as for *S. aureus* from mastitis in cattle, 95.1% and 91.7%, respectively, being susceptible to all antimicrobials included. Resistance was most frequently observed for antimicrobials commonly used for treatment of infections in these production animals; penicillin, trimethoprim and streptomycin.

A total of 100 isolates of *Moritella viscosa* from diagnostic samples in winter ulcer in Atlantic salmon (*Salmo salar*) were included for the first time in NORM-VET. The results indicate susceptibility to all antimicrobials included.

Resistance in indicator bacteria

The prevalence of acquired antimicrobial resistance among certain bacteria of the normal enteric microflora can serve as an indicator of the selective antimicrobial pressure in the various populations.

In NORM-VET, *Escherichia coli* and *Enterococcus* spp. serve as indicator bacteria. In 2003, indicator bacteria from faecal samples from cattle and sheep and from cattle meat samples were included. In addition, data from a survey involving reindeer are presented.

The results obtained in 2003 indicate a moderate occurrence of antimicrobial resistance in generic *E. coli* and *Enterococcus* spp. isolated from cattle and in *E. coli* from wild reindeer, whereas the prevalences in *E. coli* and *Enterococcus* spp. from sheep were low. The occurrence of resistance in indicator bacteria from cattle and sheep is at the same level as was observed in 2001. Resistance was only observed to those antimicrobials commonly used for therapy in cattle and sheep.

Resistance in zoonotic and other enteropathogenic bacteria

Salmonella spp., apart from *S. enterica* subsp. *diarizonae* in sheep, is rarely isolated from food producing animals in Norway. In 2003, 14 isolates of *Salmonella* spp. were susceptibility tested. All but one of the isolates, which was multiresistant (*S.* Typhimurium from a horse), were susceptible to all antimicrobials included.

I 2003, 80% of the human cases of salmonellosis was reported as being infected abroad. The incidence of multiresistant *S.* Typhimurium DT104 infection, especially domestically acquired cases was reduced markedly in 2003 as compared to 2002. The proportion of *S.* Typhimurium isolates (excluding DT104) susceptible to all antimicrobials was higher for the category "infected in Norway" (88.2%) than for the "infected abroad" category (59.4%). Multiresistance was more common in the latter category (19.8%) as compared to the former (5.3%). The vast majority of *S.* Enteritidis isolates came from patients infected abroad. Resistance to nalidixic acid was more widespread among *S.* Enteritidis (26.6%) as compared to *S.* Typhimurium (10.1% in the category "infected abroad" and 2.5% in the category "infected in Norway"). Of the nalidixic acid resistant *S.* Enteritidis isolates, 6.9% showed reduced susceptibility to ciprofloxacin indicating that fluoroquinolone resistance could be developing.

The results obtained in 2003 show that the prevalence of resistance in *Campylobacter jejuni* from Norwegian broilers is low. A total of 95% of the isolates were susceptible to all antimicrobials included and only 1.4% of the isolates were fluoroquinolone resistant. Altogether 3.6% were resistant to one antimicrobial (ampicillin or oxytetracycline) and 1.4% to oxytetracycline, nalidixic acid and the fluoroquinolone enrofloxacin. The results are similar to those presented in previous NORM/NORM-VET reports.

The level of resistance and the resistance patterns for *C. jejuni* isolated from Norwegian broilers correspond quite well with what was observed for *C. jejuni* isolated from humans infected within Norway, except for a higher prevalence of resistance to quinolones among the human isolates. This relationship was also observed in NORM/NORM-VET 2001 and 2002. Resistance was significantly more widespread in the *C. jejuni* isolates derived from patients infected abroad (86% resistant to at least one antimicrobial) than patients infected in Norway (16%). Fluoroquinolone resistance was more common in isolates from infections acquired abroad than from domestic cases; 70.8% versus 7.9%.

The vast majority of the *Shigella* isolates tested came from patients infected abroad. As is the case in reports from other countries, resistance was widespread. The resistance prevalences were particularly high for trimethoprim/sulfonamides (>65%), and for tetracycline (>58%). Resistance to fluoroquinolones was less prevalent and only observed in *S. flexneri* (2.3%). However, a considerable proportion of *S. flexneri* (14%) and *S. sonnei* (18.9%) were resistant to nalidixic acid, many of which also expressed reduced susceptibility to ciprofloxacin, indicating that fluoroquinolone resistance could be developing.

Resistance in clinical isolates from humans

The prevalence of antimicrobial resistance in human clinical isolates was still very low in Norway in 2003, and only minor changes were observed from 2002 to 2003. Four of 637 *Staphylococcus aureus* blood culture isolates (0.6%) and three of 1,144 *S. aureus* wound isolates (0.3%) were verified as methicillin resistant (MRSA) by *mecA* and *nuc* PCRs. This is in contrast to the significant increase in the number of clinical MRSA cases reported to the Norwegian Surveillance System for Communicable Diseases (MSIS) from 143 cases in 2002 to 216 cases in 2003. This discrepancy cannot be readily explained. The proportion of invasive isolates is obviously small, and it may therefore take some time before an upward trend will be noted among blood culture isolates. Alternatively, the MRSA strains circulating in Norway may have virulence factors needed to establish soft tissue infections without causing systemic disease. Both the epidemiological and the microbiological surveillance systems for MRSA will be further improved in 2004 and hopefully answer some of these questions. The prevalence of resistance to fusidic acid in *S. aureus* blood culture isolates declined from 8.0% in 2002 to 6.1% in 2003. Even though the

prevalence of resistance to fusidic acid remained high in wound specimens (23.0%), it was only marginally increased from 2002 (20.8%). One may therefore suspect that the epidemic spread of this phenotype has now decreased.

Blood culture isolates of *E. coli* and *Klebsiella* spp. were generally susceptible to broad-spectrum antimicrobials including ceftazidime, cefpirome, meropenem, ciprofloxacin and piperacillin/tazobactam. Three out of 966 *E. coli* isolates (0.3%) and one out of 299 *Klebsiella* spp. isolates (0.3%) produced extended spectrum β-lactamases (ESBL). The prevalence of intermediate susceptibility and resistance to quinolones was low but may be increasing. The overall prevalence of ciprofloxacin non-susceptibility was 5.1% in *E. coli* and 8.3% in *Klebsiella* spp. The reduced susceptibility to quinolones in these species parallels the increasing use of ciprofloxacin from 0.29 DDD/1,000 inhabitants/day in 2000 to 0.42 DDD/1,000 inhabitants/day in 2003.

Only a single VanB vanomycin resistant *Enterococcus faecium* strain (VRE) was detected among 252 enterococcal blood culture isolates (0.4%). However, the prevalence of high-level resistance to gentamicin in *E. faecalis* has increased from 4.5% in 2001 to 9.6% in 2002 and further to 14.2% in 2003. A similar trend has not been observed for *E. faecium*. The increasing prevalence of high-level gentamicin resistance in enterococci is worrisome as it endangers the traditional β-lactam/aminoglycoside combination regimen used for empirical treatment of septicaemiae in Norwegian hospitals.

Streptococcus pneumoniae from blood cultures were generally susceptible to all relevant antimicrobials. Only three of 514 blood culture isolates (0.6%) and 21 of 752 respiratory tract isolates (2.8%) displayed reduced susceptibility to penicillin G. No pneumococcal isolates were resistant to penicillin G or cephalosporins. However, the prevalence of non-susceptibility to erythromycin continued to increase and reached 6.0% in blood culture isolates and 5.7% in respiratory tract isolates. This was apparently due to further dissemination of strains with low-level resistance to erythromycin and susceptibility to clindamycin.

Moraxella catarrhalis was included in NORM for the first time in 2003, and to our knowledge this is the first systematic survey of antimicrobial resistance in this species in Norway. As reported from other countries, the vast majority of isolates (92.3%) produced β-lactamase. The results further demonstrated widespread susceptibility to tetracyclines, macrolides, trimethoprim/sulfamethoxazole, quinolones and broad-spectrum β-lactams in this species. In addition, valuable technical information concerning methodology and determination of breakpoints was collected.

A total of 339 cases of tuberculosis were reported to MSIS in 2003, and 320 had not previously been treated with antituberculosis drugs. Susceptibility tests were performed on 272 *Mycobacterium tuberculosis* isolates. One isolate showed combined resistance to rifampicin (R) and isoniazid (H) and was thus multidrug resistant by usual definitions. However, the isolate was susceptible to other first-line drugs. Susceptibility tests were also performed on *Mycobacterium tuberculosis* isolates from 11 patients who had previously received antituberculosis drug treatment. Two isolates from East Europe were multidrug resistant (MDR).

Conclusion

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

IV. POPULATION STATISTICS

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

TABLE 1. Human population in Norway as of January 1, 2004.
(Data provided by Statistics Norway).

Age group	All	Males	Females
0 to 4 years	292 594	149 678	142 916
5 to 14 years	617 734	317 382	300 352
15 to 24 years	546 993	279 072	267 921
25 to 44 years	1 323 730	673 481	650 249
45 to 64 years	1 097 625	555 165	542 460
65 years and older	673 576	281 329	392 247
All age groups	4 552 252	2 256 107	2 296 145

TABLE 2. Livestock population in Norway and the number of slaughtered animals in 2003.

Animal category	Herds[1]	Number* of Animals[1]	Slaughtered animals[2]
Cattle	23 600	949 600	333 700
Dairy cow**	16 100	252 300	-
Suckling cow**	4 100	42 600	-
Combined production (cow)**	1 400	33 900	-
Goat	1 100	49 500	19 500
Dairy goat**	600	45 800	-
Sheep	-	2 415 400	1 214 300
Breeding sheep > 1 year**	18 400	927 800	-
Swine	4 000	781 000	1 339 500
Breeding animal > 6 months**	2 300	60 000	-
Fattening pigs for slaughter	3 600	419 000	-
Poultry			
Egg laying hen (> 20 weeks of age)	2 900	3 214 600	2 156 700
Flocks > 250 birds**	980	-	-
Broiler	500	-	40 372 400
Turkey, duck and goose for slaughter	200	311 600	904 500
Flocks > 25 birds**	79	-	-
Ostrich	26	330	-
Horse			2 200
Farmed salmon			532 400 [3]
Farmed trout			71 500 [3]

Data from: [1)] Register of Production Subsidies as of July 31, 2003; [2)] Register of Slaughtered Animals; [3)] Directorate of Fisheries, amount in metric tons, ungutted fish, preliminary data. * Numbers > 100 rounded to the nearest ten, numbers > 1000 rounded to the nearest hundred. ** Included in above total.

TABLE 3. Animals (excluding fish) imported to Norway in 2003. *(Data provided by the Norwegian Food Safety Authority).*

	Live animals*		Semen doses		Embryos	
Animal species	Individuals	Consignments	Doses	Consignments	Numbers	Consignments
Cattle	17	1	< 180 000	47	< 100	20
Goat	92	7				
Pig	6	2	< 200	21		
Fur animals	59	3				
Gallus gallus – day old chicks	Approx. 8 300	7				
Gallus gallus – consignments < 20 birds	Approx. 210	12				

*All live animals were imported from Finland, Denmark or Sweden

V. USAGE OF ANTIMICROBIAL AGENTS

A. USAGE IN ANIMALS

Antimicrobial and coccidiostatic growth promoters

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

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. The measures resulted in an immediate reduction in the use of these substances (Table 4). In 1998, the streptogramin virginiamycin was officially prohibited due to reports from other countries of an association between its use and the occurrence of enterococci that were resistant to quinupristin-dalfopristin, a streptogramin combination preparation used in human medicine. Antimicrobial growth promoters have not been used in animals in Norway since 1998.

Coccidiostatic growth promoters are still used in Norwegian poultry production. The total sales of coccidiostats, in kilograms of active substance, are at the same level as before the ban on antimicrobial growth promoters was implemented. However, the pattern of usage has changed. The use of coccidiostats has been dominated by narasin since 1996, while the usage of other ionophores has decreased correspondingly.

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

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

*Prohibited part of the year

Therapeutic usage of veterinary antimicrobial drugs

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

Table 5 summarizes the sales (in kg of active substance) in 2003 of veterinary antimicrobial drugs approved for therapeutic use in domestic animals in Norway. The data are organized according to the main groups of substances (ATCvet) and show the total use for the various routes of administration. The total sale of veterinary antimicrobial drugs is given in Figure 1. Figure 2 illustrates the proportion of the total sale of the various main groups of antimicrobial substances. Both figures present annual sales data for the period 1995–2003. In 2003, the sales of veterinary antimicrobials approved for therapeutic use in animals in Norway amounted to 5,787 kg of active substance (Table 5). The annual use of veterinary antimicrobial drugs decreased gradually by 40% from 1995 to 2001. This figure has remained on a relatively constant level since. The proportion accounted for by pure penicillin preparations rose from 25% in 1995 to 40% in 2003. Altogether 84% of the veterinary penicillin preparations sold in 2003 were β-lactamase-sensitive penicillins. From 1995 to 2002, the sale of sulfonamides in combination with trimethoprim or baquiloprim increased from 11% to 24% of the total sales, this figure declined by 2% from 2002 to 2003 mainly due to a reduced sale of formulations for dogs and cats. The proportion of sale of the combination preparations of penicillins and amino-glycosides decreased from 35% to 24% from 1995-2003. The corresponding figure for the sulfonamides decreased gradually from 14% in 1995 to 0.4% and 0.3% in 2002

and 2003, resepectively. The proportion of tetracyclines declined from 5% to 3% during the same period.

TABLE 5. Sales in 2003 (in kilograms of active substance) of veterinary antimicrobial drugs approved in Norway for therapeutic use in animals, excluding fish. The data were obtained from Norwegian Institute of Public Health and represent sales from drug wholesalers to Norwegian pharmacies.

Groups of substances	ATCvet code	Active substance or combinations of substances	Gastro-intestinal (QA07)	Uterine (QG01)	Systemic indiv. (QJ01)	Systemic herds (QJ01)	Intra-mammary (QJ51)
Tetracyclines	QG01AA07	Oxytetracycline		2			
	QJ01AA02	Doxycycline			< 0.1		
	QJ01AA06	Oxytetracycline			88	87	
Beta-lactams	QJ01CA01	Ampicillin			8		
	QJ01CA04	Amoxicillin			87	105	
	QJ01CE09	Benzylpenicillinprocain*			1 963		
	QJ01CE90/QJ51CE90	Penethamate hydroiodide*			9		
	QJ01CR02/QJ51RV01	Amoxicillin+clavulanic acid			159		6
	QJ51CA51	Ampicillin+cloxacillin					1
Sulfonamides	QJ01EQ06	Sulfanilamid			19		
Sulfonamides and	QJ01EQ10	Sulfadiazine+trimethoprim			1 161		
trimethoprim**	QJ01EQ13	Sulfadoxine+trimethoprim			112		
Lincosamides	QJ01FF01	Clindamycin			9		
	QJ01FF02	Lincomycin			7		
Aminoglycosides	QA07AA01	Neomycin	35				
	QA07AA90	Dihydrostreptomycin (DHS)	139				
Quinolones	QJ01MA90	Enrofloxacin			27		
Others	QJ01XX92	Tiamulin			11	181	
Combinations	QG01AE99	Sulfadimidine+procaine penicillin*+DHS		193			
	QJ01RA01	Benzylpenicillinprocain *+DHS			586		764
	QJ01RA01	Spiramycin+metronidazole			4		
	QJ51RC25	Penethamate hydroiodide*+ DHS					24
Total per route of administration			**174**	**195**	**4 250**	**373**	**795**
Total							**5 787**

*Calculated as benzylpenicillin
**Includes baquiloprim

In Norway, medicated feeds and premixes for farmed fish are approved by the drug authorities and classified as pharmaceutical specialities. Sales figures, in kg of active substance, of such products and premixes containing antimicrobial drugs are presented in Table 6. Quinolones is the antimicrobial class most commonly used in farmed fish. In 2003, quinolones accounted for 75% (in kg) of the total antimicrobial drug use in fish.

Altogether, 805 kg of veterinary antimicrobial drugs for therapeutic use in farmed fish were sold in 2003. Annual use of antimicrobial drugs for fish declined by 98% during the period 1987-2003. In the same period, the total production of farmed fish increased massively. This significant decrease in the use of antimicrobial drugs in aquaculture is mainly attributed to the introduction of effective vaccines and to improved health management.

TABLE 6. Total sales (in kilograms of active substance) of veterinary antimicrobial drugs for therapeutic use in farmed fish in Norway 1995-2003. The data were obtained from Norwegian Institute of Public Health and represent sales from drug wholesalers to Norwegian pharmacies and sales by feed mills.

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

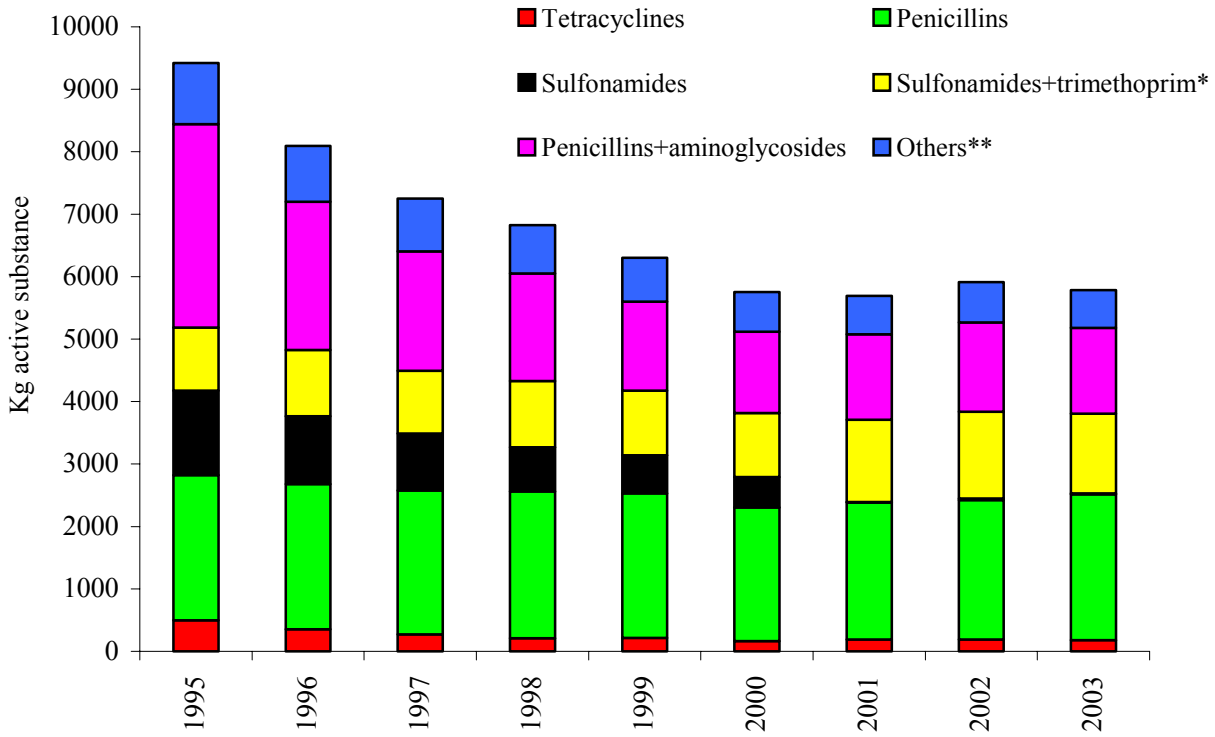

FIGURE 1. Sales (in kilograms of active substance) of veterinary antimicrobial drugs (QA07AA, QG01AA, QG01AE, QJ01, QJ51) for therapeutic use in Norway in 1995–2003, fish not included.

*Includes small amounts of baquiloprim.
**Includes ATCvet codes: QA07AA01; QA07AA51; QA07AA90; QG01AE99; QJ01EQ06; QJ01FA02; QJ01FF01; QJ01FF02; QJ01MA90; QJ01RA91; QJ01XX92.

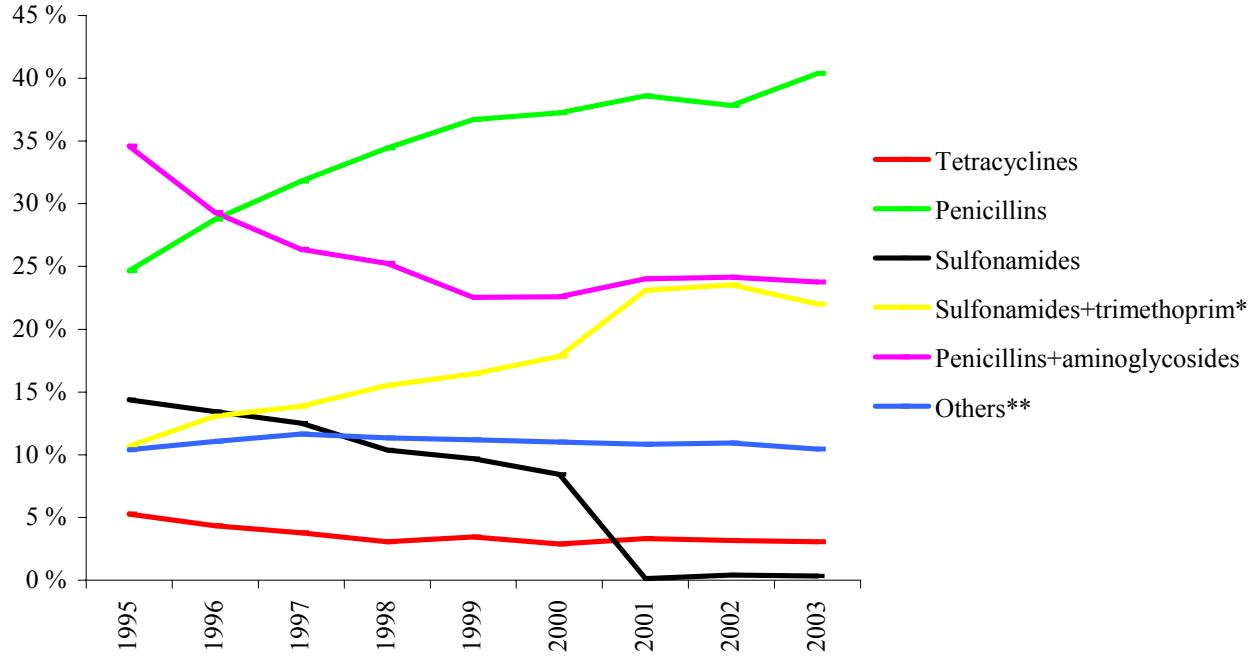

FIGURE 2. Sales (as percentage of total sales) of veterinary antimicrobial drugs (QA07AA, QG01AA, QG01AE, QJ01, QJ51) in Norway in 1995–2003, fish not included.

*Includes small amounts of baquiloprim.
**Includes ATCvet codes: QA07AA01; QA07AA51; QA07AA90; QG01AE99; QJ01EQ06; QJ01FA02; QJ01FF01; QJ01FF02; QJ01MA90; QJ01RA91; QJ01XX92.

The effect of discontinuing use of antimicrobial groth promoters

When Sweden voluntarily banned all use of antimicrobial growth promoters (AGPs) in 1986 it was feared that a rise in the use of therapeutic antimicrobials would occur. However, overall usage data on veterinary antimicrobial agents in Sweden have shown a reduced consumption following the ban (1, 2). Usage data on veterinary antimicrobials published in the present report show that this is also the case in Norway after avoparcin was banned and the use of other AGPs voluntarily abandoned in 1995.
In 1999, Switzerland introduced a ban on AGPs. To assess whether this ban resulted in an increase in the therapeutic use of antimicrobials in piglets and fattening pigs the usage of antimicrobials for the period 1996 to 2001 was investigated (3). A total of 6,427 prescriptions on medicated feedstuff delivered to pig farms in a Swiss canton were investigated and the overall annual amounts of antimicrobials delivered (kg active substance) were calculated. To correct for differences in dosages between the various substances and for the population liable to be treated, prescribed daily dosages (PDD)/population were estimated. The usage of antimicrobials in pigs decreased from 1,200 to 709 kg in the period 1996-1999, while this figure increased gradually to 936 kg in 2001. When expressed as number of PDDs/population the usage decreased from 6.1 in 1996 to 3.6 in 1999 and thereafter remained low (3.3 in 2000 and 3.4 in 2001). It was concluded that the ban of AGPs did not result in increased therapeutic use of antimicrobials in pig farming in Switzerland.
When avoparcin was banned in Norway on May 31, 1995 it was feared that this would result in an increased incidence of *Clostridium perfringens*-associated necrotic enteritis (NE) and subsequently an increase in the therapeutic use of antimicrobials in meat type poultry. To evaluate this hypothesis, the annual treatment frequency of NE in poultry before and after the ban of avoparcin was estimated by use of national sales statistics of antimicrobial drugs indicated for the treatment of NE in poultry (4). This assessment showed that the ban led to a temporary increase in the treatment rates in the last quarter of 1995. However, since 1996, the treatment rates of NE in poultry have been approximately at the same level as before the ban. A preferential use of narasin instead of other ionophores following the avoparcin ban may explain the low treatment rates of NE in broilers since 1996.
In Denmark, avoparcin was banned in May 1995, whereas virginiamycin was banned in January 1998. In February 1998, the Danish poultry industry voluntarily decided to discontinue the use of all AGPs. To investigate how the removal of AGPs influenced the broiler productivity in Denmark, data from 6,815 flocks collected from January 1996 to July 1999 were analysed (5). The three flock parameters investigated were kilogram broilers produced per square meter, feed-conversion ratio and total percent dead broilers. The study showed that kilogram broilers produced per square meter and percent dead broilers in total were not affected by the discontinued use of AGPs. However, the feed-conversion ratio increased marginally by 0.016 kg/kg.
In 2002, a WHO International Review Panel evaluated the impacts of AGP termination in Denmark (6). It was found that the overall therapeutic use in pigs in 2000 and 2002 was similar to that in 1994, the peak year before any AGPs were terminated. Therapeutic use in poultry appeared to be unaffected by the AGP termination.
The data and studies described above show that it is possible, at least for some animal production systems, to abandon the use of AGPs in animal production without any significant increase in therapeutic use or any considerable loss of productivity.

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B. USAGE IN HUMANS

In 2003, the overall sales of antimicrobials for systemic use (ATC group J01) in humans represented 17.1 DDD/1,000 inhabitants/day, the same as in 2002. Total sales of antimicrobials have remained relatively unchanged for many years. However, within subgroups of antimicrobials, usage trends have changed. The subgroups of cephalosporins, macrolides and quinolones are steadily increasing, the penicillin group is stable, while the subgroups of tetracyclines and the sulphonamides and trimethoprim are decreasing (Table 7, Figure 3). These are trends that have been observed over years and the sales in 2003 follow the same pattern.

TABLE 7. Human usage of antimicrobial agents in Norway 1997-2003 by ATC groups. The usage is presented as DDD (Defined Daily Doses)/1,000 inhabitants/day and in % change 1997-2003. Collection of data on human usage of antimicrobial agents is presented in Appendix 2.

ATC	Groups of substances	1997	1998	1999	2000	2001	2002	2003	Change (%) 1997-2003
J01A	Tetracyclines	3.55	3.37	3.19	3.17	3.11	3.13	3.03	- 15
J01B	Amphenicols	0.005	0.004	0.005	0.004	0.003	0.002	0.002	
J01CA	Penicillins with extended spectrum	1.87	1.90	1.96	2.01	2.1	2.23	2.29	+ 22
J01CE	β-lactamase sensitive penicillins	5.32	5.12	5.01	4.66	4.68	4.48	4.38	- 18
J01CF	β-lactamase resistant penicillins	0.24	0.27	0.32	0.35	0.41	0.50	0.59	+146
J01CR	Combination of penicillins	0.02	0.01	0.01	0.01	0.01	0.01	0.01	
J01D	Cephalosporins, monobactams, carbapenems	0.42	0.44	0.47	0.52	0.55	0.58	0.62	+ 48
J01E	Sulfonamides and trimethoprim	1.45	1.34	1.26	1.17	1.16	1.15	1.08	- 26
J01F	Macrolides, lincosamides and streptogramins	1.58	1.61	1.59	1.59	1.8	1.98	1.92	+ 22
J01G	Aminoglycosides	0.05	0.05	0.05	0.04	0.06	0.06	0.07	
J01M	Quinolones	0.28	0.30	0.33	0.35	0.40	0.44	0.48	+ 71
J01X	Other antimicrobials	2.06	2.2	2.34	2.39	2.55	2.57	2.63	+ 27
	Total	**16.8**	**16.6**	**16.6**	**16.3**	**16.8**	**17.1**	**17.1**	

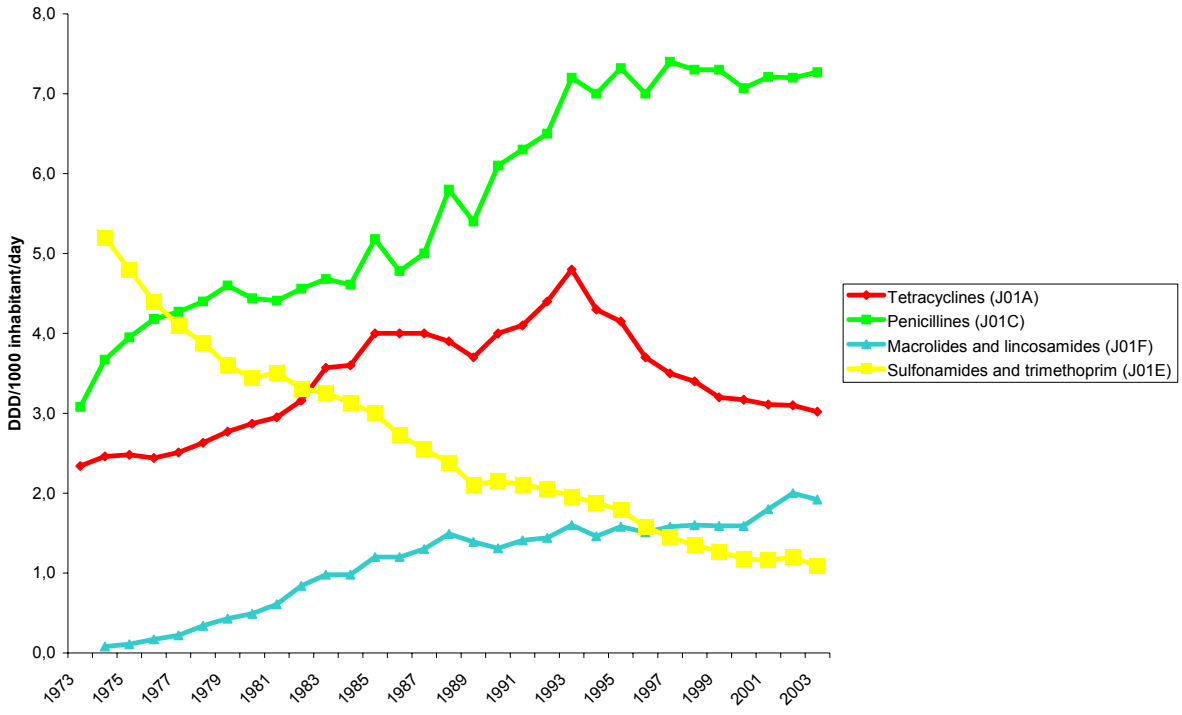

FIGURE 3. Sales of penicillins (J01C), tetracyclines (J01A), macrolides, lincosamides and streptogramines (J01F) and sulfonamides and trimethoprim (J01E) in Norway 1973-2003.

In 2003, the penicillins (ATC group J01C) represented 42% of the total antimicrobial use in Norway (Figure 4). The sales of penicillins have been stable over years. It has, however, looking at ATC 4th levels, been a shift towards more broad-spectered penicillins (Figure 5). β-lactamase sensitive penicillins (J01CE) decreased - by 2% in 2003. The sales of benzylpenicillin, mainly used in hospitals, were stable; hence, the decrease was due to phenoxy-methylpenicillin (Table 8), a peroral penicillin formulation mainly used in ambulatory care. The subgroups of penicillins with extended specter and β-lactamase-resistant penicillins are both increasing, by 2,7% and 18%, respectively.

The tetracyclines (J01A) represent 18% of total use. The sales have been decreasing since 1993, when the highest sale ever was registered.

The macrolides (J01FA) represents 10% of total use in 2003. Sales have been fairly stable through the nineties. In 2001 and 2002 an increase of 24% compared to 2000 was found. However, this trend was not continued in 2003. The internal macrolide pattern has remained unchanged. The lincosamides, in Norway represented by clindamycin, is increasing, by 90% since 1997 and by 19% since 2002 (Table 8). Sales of cephalosporins, monobactams and carbapenems, although limited, have also been increasing, now representing 4.4% of the total sales of antimicrobials. The internal subgroup pattern has changed since 1996 (Figure 6). Mainly 1st generation cephalosporins are used i.e. cefalexin and cefalotin, representing 67% of the cephalosporins in 1996 and 60% in 2003. Over the years, there has been a shift towards using more 3rd generation cephalosporins and carbapenems. The sales of sulfonamides and trimethoprim are decreasing - by 26% since 1997. There has been a small, but stable increase in quinolone use. Still it represents only a minor fraction (2.8%) of total antimicrobials sales, but the increase has been 71% since 1997. The increase of ATC group J01X is mainly due to the urinary prophylactic agent methenamine representing 12.8% of total use. The sales have increased by 35% since 1997.

The therapy pattern of antimicrobials in hospitals differs from the sale in ambulatory care (Figure 7). The antimicrobials sales in DDDs to hospitals represented, in 2003, eight percent of total sale in the country. Penicillins (J01C) represent around 48% and 41% of the use in hospitals and in ambulatory care, respectively. The most important other groups in ambulatory care are tetracyclines, J01A (18%) and macrolides and lincosamides, J01F (12%). In hospitals cephalosporins, J01DA (17%) is the most used group after the penicillins, followed by metronidazole - oral and parenteral (6%) and the quinolones, J01M (5%).

The use of antimicrobials outside hospital represents 92% of the total human sale of antimicrobials. Therapy traditions in ambulatory care therefore have much greater impact on the total burden of antimicrobials and furthermore to the development of bacterial resistance. Hence, surveillance of use in ambulatory care may be regarded as very important. In Norway antibiotics are prescription-only drugs. From 1 January 2004, a national prescription database has been established. Data from this register will be available in 2005. Furthermore, for overall use, the slow, but steady shift towards use of more broad-spectered antimicrobials in Norway is of concern and deserves close surveillance.

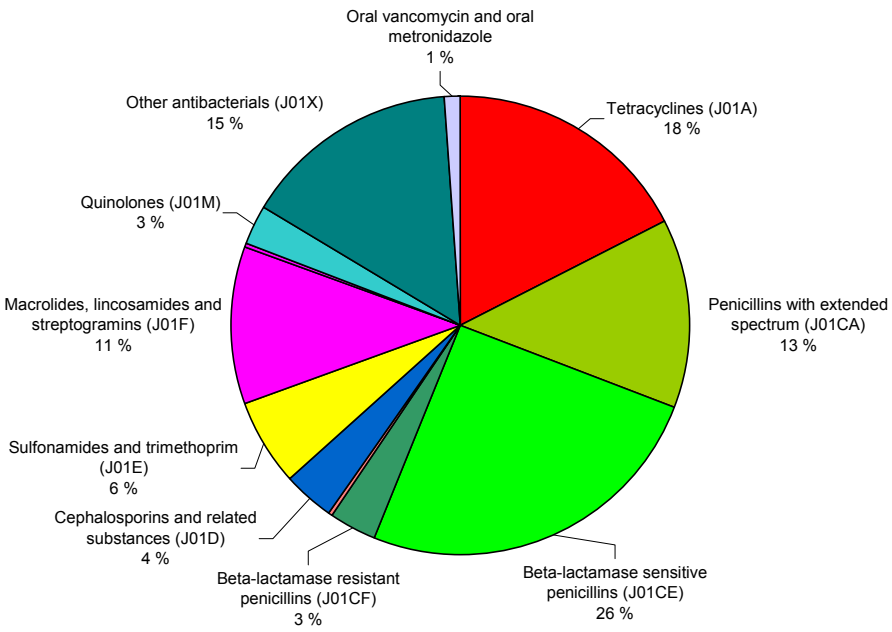

FIGURE 4. Relative amount of antimicrobial agents for systemic use in 2003 in Defined Daily Doses (DDD) (total sale in the country). Groups of antimicrobials are represented by ATC numbers.

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

ATC	Substance	1997	1998	1999	2000	2001	2002	2003
A07AA09	Vancomycin	0.001	0.001	0.001	0.001	0.001	0.001	0.001
J01A A02	Doxycycline	2.47	2.34	2.20	2.10	2.1	2.03	1.93
J01A A04	Lymecycline	0.10	0.09	0.09	0.14	0.19	0.26	0.30
J01A A06	Oxytetracycline	0.30	0.27	0.25	0.24	0.22	0.21	0.19
J01A A07	Tetracycline	0.68	0.67	0.65	0.69	0.64	0.62	0.60
J01B A01	Chloramphenicol	0.005	0.004	0.005	0.004	0.003	0.002	0.002
J01C A01	Ampicillin	0.09	0.09	0.09	0.09	0.08	0.09	0.1
J01C A02	Pivampicillin	0.17	0.15	0.14	0.13	0.11	0.11	0.09
J01C A04	Amoxicillin	0.85	0.85	0.87	0.83	0.89	0.94	0.95
J01C A08	Pivmecillinam	0.75	0.81	0.86	0.96	1	1.09	1.14
J01C A11	Mecillinam	0.003	0.003	0.004	0.004	0.005	0.005	0.005
J01C E01	Benzylpenicillin	0.19	0.21	0.23	0.21	0.23	0.24	0.25
J01C E02	Phenoxymethylpenicillin	5.13	4.91	4.78	4.45	4.45	4.24	4.13
J01CE08*	Benzathine benzylpenicillin	<0.0001	<0.0001	<0.0001	0.0001	<0.0001	0.0001	0.0001
J01C F01	Dicloxacillin	0.16	0.19	0.22	0.25	0.31	0.39	0.48
J01C F02	Cloxacillin	0.08	0.08	0.10	0.10	0.09	0.11	0.11
J01C F05*	Flucloxacillin						0.0001	0.0002
J01C R02	Amoxicillin and enzyme inhibitor	0.02	0.01	0.01	0.01	0.01	0.01	0.01
J01C R05	Piperacillin and enzyme inhibitor				0.0001	0.0006	0.0014	0.0024
J01D A01	Cefalexin	0.22	0.22	0.22	0.26	0.27	0.29	0.3
J01D A03	Cefalotin	0.04	0.04	0.05	0.05	0.05	0.05	0.06
J01D A05	Cefoxitin	0.0004	0.0004	0.0004	0.0004	0.0003	0.0002	0.0001
J01D A06	Cefuroxim	0.11	0.12	0.13	0.13	0.14	0.15	0.15
J01D A10	Cefotaxim	0.02	0.03	0.04	0.04	0.05	0.05	0.07
J01D A11	Ceftazidim	0.01	0.01	0.01	0.01	0.01	0.01	0.01
J01D A13	Ceftriaxone	0.004	0.007	0.008	0.011	0.01	0.01	0.01
J01D A63	Ceftriaxone, combinations	0.0001	0.0001	0.0001				
J01D F01	Aztreonam	0.0007	0.0005	0.0008	0.001	0.001	0.001	0.001
J01D H02	Meropenem	0.002	0.004	0.008	0.012	0.014	0.017	0.02
J01D H51	Imipenem and enzyme inhibitor	0.007	0.007	0.006	0.006	0.005	0.005	0.006
J01E A01	Trimethoprim	0.90	0.87	0.84	0.79	0.8	0.8	0.74
J01E B02	Sulfamethizole		0.0002	0.001	0.002	0.002	0.0001	
J01E C20	Sulfonamides, combinations	0.003	0.003	0.0004				
J01E E01	Sulfamethoxazol and trimethoprim	0.55	0.47	0.42	0.38	0.36	0.36	0.34
J01F A01	Erythromycin	1.04	1.06	1.01	1.00	1.13	1.2	1.09
J01F A02	Spiramycin	0.05	0.04	0.03	0.02	0.02	0.02	0.02
J01F A09	Clarithromycin	0.22	0.24	0.26	0.26	0.3	0.36	0.37
J01F A10	Azithromycin	0.17	0.17	0.18	0.19	0.21	0.24	0.26
J01FA15	Telithromycin						0.0001	0.0003
J01F F01	Clindamycin	0.10	0.11	0.11	0.12	0.14	0.16	0.19
J01GA01*	Streptomycin						0.0015	0.0004
J01G B01	Tobramycin	0.03	0.03	0.03	0.02	0.03	0.04	0.04
J01G B03	Gentamicin	0.006	0.006	0.006	0.006	0.008		0.03
J01G B06*	Amikacin						0.0009	0.0008
J01G B07	Netilmicin	0.02	0.02	0.02	0.02	0.02	0.007	

ATC	Substance	1997	1998	1999	2000	2001	2002	2003
J01M A01	Ofloxacin	0.07	0.06	0.06	0.05	0.05	0.05	0.05
J01M A02	Ciprofloxacin	0.20	0.23	0.26	0.29	0.34	0.38	0.42
J01MA12*	Levofloxacin						0.001	0.0003
J01M B02	Nalidixic acid	0.01	0.01	0.01	0.01	0.01		
J01X A01	Vancomycin	0.005	0.005	0.004	0.005	0.005	0.006	0.006
J01X A02	Teicoplanin	0.0009	0.001	0.0007	0.0012	0.0013	0.0013	0.0009
J01X B01	Colistin	0.004	0.003	0.003	0.003	0.003	0.003	0.002
J01X C01	Fusidic acid	0.003	0.003	0.003	0.003	0.01	0.01	0.007
J01X D01	Metronidazole	0.056	0.056	0.060	0.063	0.065	0.069	0.073
J01X E01	Nitrofurantoin	0.38	0.38	0.37	0.37	0.36	0.35	0.35
J01X X05	Methenamin	1.61	1.75	1.91	1.95	2.08	2.13	2.18
J01XX08	Linezolid						0.002	0.004
P01AB01	Metronidazole	0.18	0.18	0.18	0.18	0.18	0.19	0.19
J04AB**	Rifampicin	-	-	0.052	0.046	0.054	0.043	0.049

* Drugs not licenced for the Norwegian market.
** Given as the amount of Rifampicin in plain and combination products. Data for 1997 and 1998 are not available.

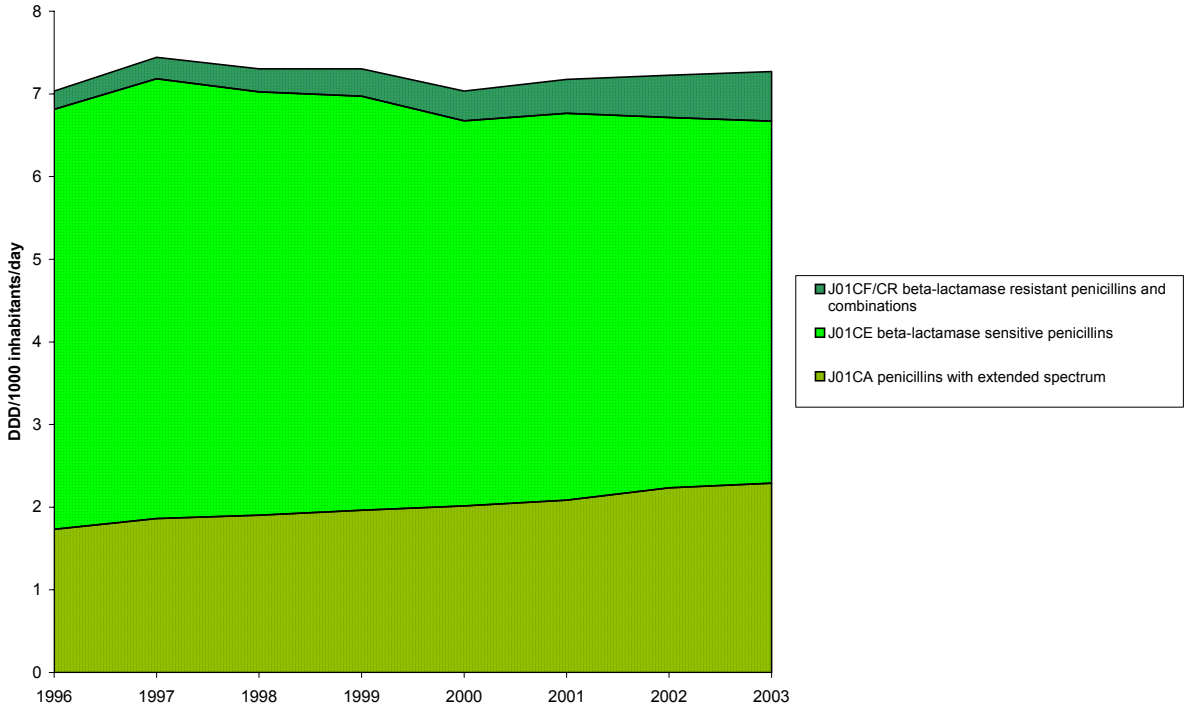

FIGURE 5. Sales of penicillins (J01C) in Norway 1996-2003 and changes within groups of penicillins.

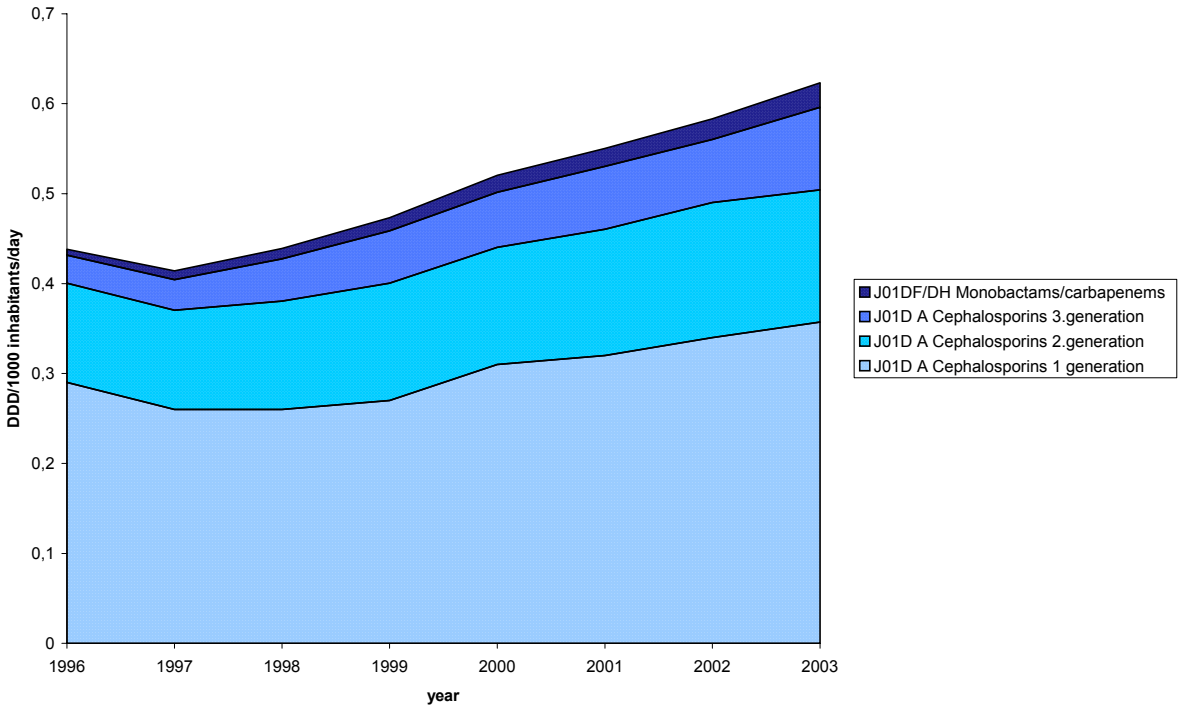

FIGURE 6. Sales of cephalosporins, monobactams and carbapenems (J01D) in Norway 1996-2003 and changes between generations of cephalosporins and monobactams/carbapenems.

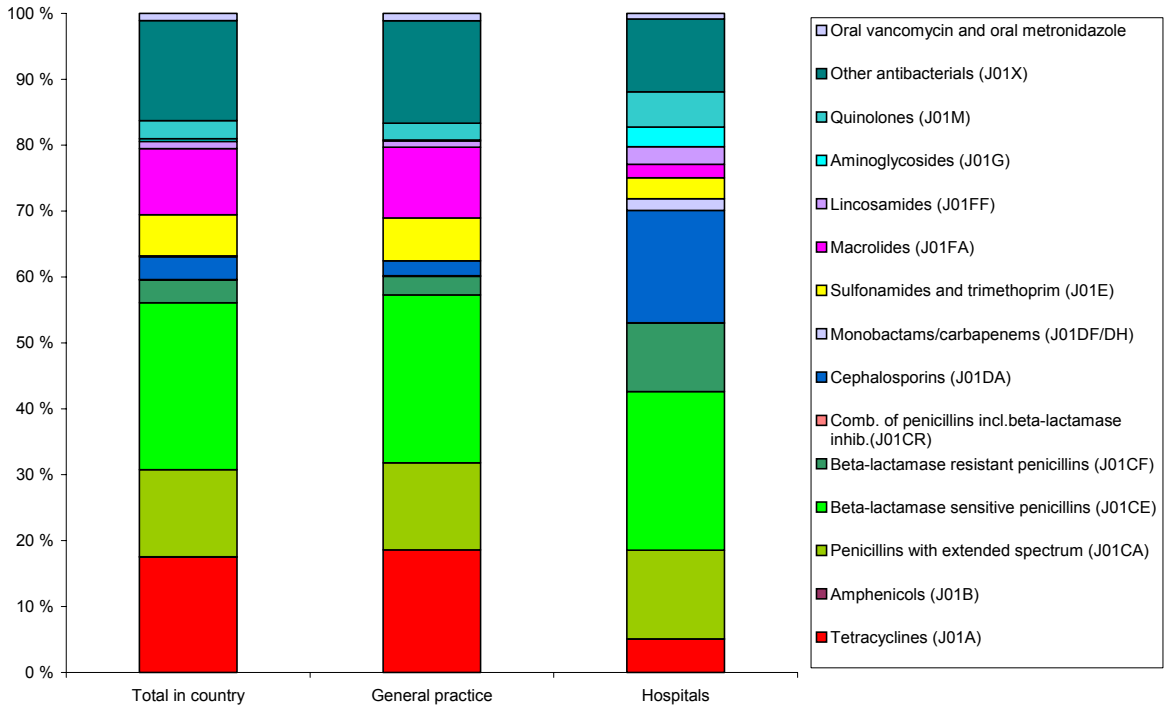

FIGURE 7. Proportions of antimicrobial agents for systemic use in Norway 2003 measured in DDD. Shown as total use, in general practice and in hospitals.

VI. OCCURRENCE OF ANTIMICROBIAL RESISTANCE

A. ANIMAL CLINICAL ISOLATES

As proposed in the NORM-VET plan, the clinical isolates in 2003 included *Staphylococcus aureus* and coagulase negative staphylococci from mastitis in cattle and *S. aureus* from mastitis in sheep and goat. In addition, isolates of *Moritella viscosa* from diagnostic samples in wound infections in Atlantic salmon (*Salmo salar*) were included for the first time in NORM-VET. Sampling, laboratory methods and data processing are described in Appendix 3.

Staphylococcus spp. from cattle, sheep and goats

A total of 117 isolates of *Staphylococcus aureus* from mastitis in cattle, 107 isolates of coagulase negative staphylococci (CoNS) from mastitis in cattle, 82 isolates of *S. aureus* from mastitis in sheep and 60 isolates of *S. aureus* from mastitis in goats were susceptibility tested. The results are presented in Tables 9, 10, 11 and Figure 8.

TABLE 9. Antimicrobial resistance in *Staphylococcus aureus* from mastitis in cattle (n=117), sheep (n=82) and goat (n=60).

Substance	Animal species	Resistance (%) [95% CI*]		Distribution (%) of MIC values (mg/L)													
				0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	Cattle	0	[0.0-3.1]					88.9	11.1									
	Sheep	1	[0.0-6.6]					90.2	8.5							1.2		
	Goat	0	[0.0-6.0]					98.3	1.7									
Chloramphenicol	Cattle	0	[0.0-3.1]					0.9			8.5	87.2	3.4					
	Sheep	0	[0.0-4.4]								8.5	91.5						
	Goat	0	[0.0-6.0]								18.3	81.7						
Penicillin G**	Cattle	5.1	[1.9-10.8]	63.2	29.1	2.6	0.9	0.9				3.4						
	Sheep	2.4	[0.0-8.5]	69.5	25.6	2.4				1.2		1.2						
	Goat	6.7	[1.9-16.2]	68.3	23.3	1.7	1.7					5.0						
Oxacillin	Cattle	0	[0.0-3.1]				21.4	47.0	29.1	2.6								
	Sheep	0	[0.0-4.4]			1.2	19.5	45.1	29.3	4.9								
	Goat	0	[0.0-6.0]				25.0	36.7	31.7	6.7								
Cephalothin	Cattle	0	[0.0-3.1]			11.1	82.1	6.8										
	Sheep	0	[0.0-4.4]			11.0	85.4	3.7										
	Goat	0	[0.0-6.0]		1.7	21.7	70.0	6.7										
Trimethoprim	Cattle	0	[0.0-3.1]						3.4	17.9	70.9	7.7						
	Sheep	1	[0.0-6.6]					1.2	1.2	22.0	73.2	1.2			1.2			
	Goat	0	[0.0-6.0]						3.3	33.3	56.7	6.7						
Erythromycin	Cattle	< 1	[0.0-3.1]				1.7	79.5	17.1		0.9							
	Sheep	0	[0.0-4.4]				2.4	76.8	20.7									
	Goat	0	[0.0-6.0]					80.0	20.0									
Clindamycin	Cattle	0	[0.0-3.1]				77.8	19.7	1.7	0.9								
	Sheep	0	[0.0-4.4]				81.7	17.1		1.2								
	Goat	0	[0.0-6.0]				70.0	28.3	1.7									
Streptomycin	Cattle	2	[0.0-6.0]								29.9	46.2	20.5	1.7	0.9			0.9
	Sheep	1	[0.0-6.6]							1.2	29.3	52.4	14.6	1.2				1.2
	Goat	5	[1.0-13.9]								26.7	35.0	30.0	3.3	1.7			3.3
Gentamicin	Cattle	0	[0.0-3.1]					53.0	42.7	4.3								
	Sheep	0	[0.0-4.4]					64.6	29.3	6.1								
	Goat	0	[0.0-6.0]					43.3	48.3	8.3								
Neomycin	Cattle	< 1	[0.0-4.7]						85.5	13.7	0.9							
	Sheep	0	[0.0-4.4]						85.4	14.6								
	Goat	0	[0.0-6.0]						63.3	36.7								
Enrofloxacin	Cattle	0	[0.0-3.1]			58.1	41.9											
	Sheep	0	[0.0-4.4]			63.4	35.4	1.2										
	Goat	0	[0.0-6.0]			65.0	35.0											
Vancomycin	Cattle	0	[0.0-3.1]						92.3	7.7								
	Sheep	0	[0.0-4.4]						82.9	17.1								
	Goat	0	[0.0-6.0]						86.7	13.3								
Fusidic acid	Cattle	0	[0.0-3.1]		96.6	3.4												
	Sheep	0	[0.0-4.4]		96.3	2.4	1.2											
	Goat	0	[0.0-6.0]		98.3	1.7												
Avilamycin	Cattle	< 1	[0.0-4.7]							2.6	62.4	33.3	0.9	0.9				
	Sheep	0	[0.0-4.4]								51.2	46.3	2.4					
	Goat	0	[0.0-6.0]							1.7	63.3	33.3	1.7					
Virginiamycin	Cattle	0	[0.0-3.1]					6.8	89.7	3.4								
	Sheep	0	[0.0-4.4]					4.9	93.9	1.2								
	Goat	0	[0.0-6.0]					10.0	88.3	1.7								

Bold vertical lines denote microbiological cut-off values for resistance. White fields denote range of dilutions tested for each antimicrobial agent. MIC-values higher than the highest concentration tested for are given as the lowest MIC-value above the range. MIC-values equal to or lower than the lowest concentration tested are given as the lowest concentration tested. *CI = Confidence interval. ** Resistance to penicillin G was based on β-lactamase production. All isolates with a positive β-lactamase test had a MIC-value > 0.125 mg/L, and all β-lactamase negative isolates had a MIC-value ≤ 0.125 mg/L.

TABLE 10. Antimicrobial resistance in coagulase negative staphylococci (CoNS) from mastitis in cattle (n=107).

	Resistance (%)		Distribution (%) of MIC values (mg/L)													
Substance	[95% CI*]		0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	0	[0.0-3.4]					72.9	25.2	1.9								
Chloramphenicol	0	[0.0-3.4]								57.0	43.0						
Penicillin G**	26.2	[18.2-35.1]	51.4	20.6	1.9	5.6	2.8	3.7	4.7	2.8	6.5						
Oxacillin***	< 1	[0.0-5.1]			2.8	47.7	35.5	6.5	3.7	2.8			0.9				
Cephalothin	0	[0.0-3.4]			9.3	72.9	14.0	3.7									
Trimethoprim	37	[28.2-47.3]					1.9	8.4	18.7	15.0	18.7	26.2	8.4	2.8			
Erythromycin	2	[0.0-6.6]				16.8	76.6	4.7		1.9							
Clindamycin	0	[0.0-3.4]				63.6	29.9	3.7	2.8								
Streptomycin	16	[9.5-24.2]							24.3	36.4	15.0	4.7	3.7	1.9	3.7	6.5	3.7
Gentamicin	0	[0.0-3.4]					95.3	4.7									
Neomycin	0	[0.0-3.4]						98.1	1.9								
Enrofloxacin	< 1	[0.0-5.1]			42.1	53.3	3.7	0.9									
Vancomycin	0	[0.0-3.4]						70.1	29.0	0.9							
Fusidic acid	2	[0.0-6.6]		68.2	20.6	3.7	5.6	0.9	0.9								
Avilamycin	2	[0.0-6.6]						0.9	14.0	41.1	20.6	21.5	0.9	0.9			
Virginiamycin	< 1	[0.0-5.1]					30.8	60.7	7.5		0.9						

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.
*CI = Confidence interval.
** Resistance to penicillin G was based on β-lactamase production. All isolates with a positive β-lactamase test had a MIC-value > 0.125 mg/L, and all β-lactamase negative isolates had a MIC-value ≤ 0.125 mg/L.
*** Resistance to oxacillin was based on *mecA* detection.

Oxacillin resistance in coagulase negative staphylococci (CoNS)

The isolates of coagulase negative staphylococci (CoNS) consist of a heterogenous group of species with species-varying susceptibility against several antimicrobial agents. The number of isolates and species included in NORM-VET 2003 are presented in Table 11.

All isolates of CoNS were subjected to PCR for detection of the *mecA* determinant for oxacillin resistance. *mecA* was detected in one isolate (*Staphylococcus lentus*). The MIC-value for oxacillin was > 16 mg/L. The isolate did not produce β-lactamase. Furthermore, the *blaZ*-gene encoding β-lactamase production was not detected by PCR using specific primers.

TABLE 11. Number of isolates per species of coagulase negative staphylococci (CoNS) from bovine milk samples (n=107).

Species	Number of isolates
Staphylococcus capitis ss. *capitis*	4
Staphylococcus chromogenes	12
Staphylococcus cohnii	1
Staphylococcus epidermidis	10
Staphylococcus haemolyticus	3
Staphylococcus hyicus	8
Staphylococcus lentus	1
Staphylococcus saphrophyticus	1
Staphylococcus sciuri	3
Staphylococcus simulans	48
Staphylococcus warneri	2
Staphylococcus xylosus	2
Other CoNS	12

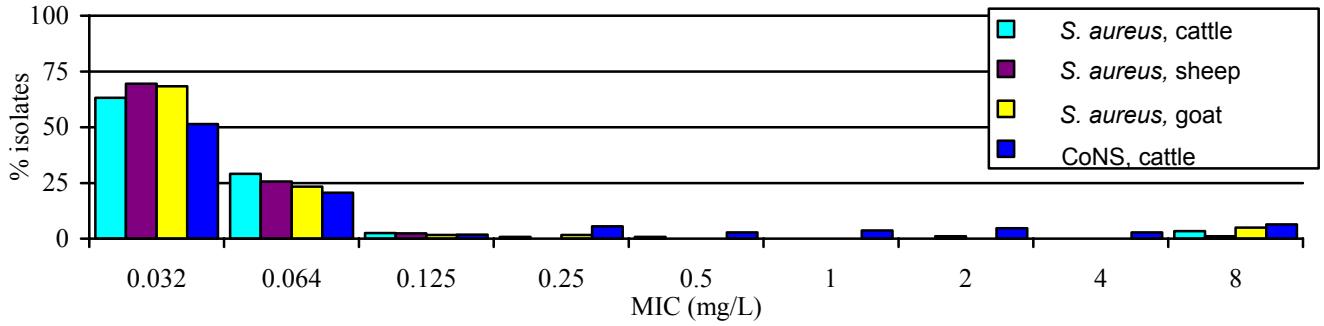

FIGURE 8. Distribution (%) of minimum inhibitory concentrations (MICs) for penicillin G among staphylococci from mastitis in cattle, sheep and goat. All isolates with a positive β-lactamase test had a MIC-value > 0.125 mg/L, and all β-lactamase negative isolates had a MIC-value ≤ 0.125 mg/L.

RESULTS AND COMMENTS

The prevalence of resistance among *S. aureus* isolates has remained at the same level during the 1990s and up to 2003. That is the case even if the inclusion criteria for isolates applied in NORM-VET differ from those applied before NORM-VET´s establishment in 2000. Before 2000, the prevalence of antimicrobial resistance in staphylococci isolated from cases of mastitis was estimated using all isolates submitted to the diagnostic laboratories. Since 2000, only one isolate per herd has been included to avoid the effect of clustering at herd level due to frequent submission of samples from "problem herds".

β-lactamase production was observed in 5.1%, 2.4% and in 6.7% of the *S. aureus* isolates from cattle, sheep and goats, respectively, and in 26.2% of the CoNS isolates. All isolates with a positive β-lactamase test had an MIC-value > 0.125 mg/L for penicillin G, and all β-lactamase negative isolates had a MIC-value ≤ 0.125 mg/L.

In 2003, the occurrence of resistance among *S. aureus* from mastitis in cows was low, 91.5% of the isolates being susceptible to all antimicrobials included. In total, 7.7% were resistant to one antimicrobial (penicillin, streptomycin or erythromycin) and 0.9% to two antimicrobials (penicillin and neomycin). The results obtained in 2003 are similar to the data presented in NORM-VET 2001.

Also the occurrence of resistance among *S. aureus* from mastitis in sheep and goats was low, 95.1% and 91.7% of the isolates, respectively, being susceptible to all antimicrobials included. Five isolates from sheep were resistant to one antimicrobial (penicillin, oxytetracycline, trimethoprim or streptomycin). Of the isolates from goats, one was resistant to one antimicrobial (penicillin), and two were resistant to two antimicrobials (penicillin and streptomycin). The resistance prevalences for the various antimicrobials reflect their usage. Penicillin and streptomycin are among the most commonly used antimicrobials for clinical purposes in cattle, sheep and goat.

Resistance in CoNS from mastitis in cows was considerably more abundant than in isolates of *S. aureus*. Only 28% of the CoNS isolates were susceptible to all the antimicrobials included. Altogether, 58.9% of the isolates were resistant to one, 11.2% to two and 1.9% to three or more antimicrobials. The prevalence of penicillin resistance in CoNS has remained at the same level throughout the 1990s and up to 2003. In 2003, only one isolate (CoNS – *S. lentus*) was resistant to oxacillin.

The use of fluoroquinolones in veterinary practice in Norway has so far been limited, and no cephalosporins are approved for use in animals in Norway. This is reflected by the high prevalence of susceptibility to these classes of antimicrobials in the data presented.

Moritella viscosa from salmon

Moritella viscosa is associated with the so-called winter ulcer in sea-farmed Atlantic salmon (*Salmo salar*). This phenomenon is one of the main reasons for production loss in Norwegian salmon production and a main indication for antimicrobial treatment. *Moritella viscosa* is currently the most commonly isolated bacterial fish pathogen from diagnostic submissions at the National Veterinary Institute. In a survey conducted in 2003, 100 isolates were susceptibility tested, 50% originating from the period 1987-1992 (when antimicrobial use in aquacultures was substantial) and 50 isolates from 2000-2002 (when antimicrobial use in aquacultures was very limited). The results are presented in Table 12.

TABLE 12. Distribution of minimum inhibitory concentrations (MICs) of *Moritella viscosa* from winter ulcer in Atlantic salmon (n=100)**.**

	Distribution (%) of MIC values (mg/L)													
Substance	< 0.002	0.002	0.004	0.008	0.016	0.032	0.064	0.125	0.19	0.25	0.5	1	2	4
Oxytetracycline					1		5	25		46	14	6	3	
Florfenicol						1				9	42	46	1	1
Oxolinic acid	31	5	4	17	20	12	5	4		2				
Flumequine		1	4	21	25	28	9	4	4	3	1			

RESULTS AND COMMENTS

So far, no microbiological cut-off values have been set for classification of *Moritella viscosa* isolates as susceptible or resistant to the various antimicrobials. Nevertheless, the results indicate that none of the isolates tested were resistant to the antimicrobials included. For example, the MIC-values for florfenicol were within the range considered susceptible for several other fish-pathogenic bacteria (Michel et al., J Appl Microbiol, 2003). Furthermore, the MIC-distributions for the various antimicrobials show a unimodal pattern.

The methodology for susceptibility testing of *Moritella viscosa* is still to be standardized. Due to some laboratory difficulties, the MIC-values determined are accompanied with some uncertainty. Because of these uncertainties, it is not possible to compare in detail the MIC-values for the two groups of bacteria included, originating from two different time periods. Nevertheless, the results indicate that there are no significant differences between the two groups as regard susceptibility levels.

B. INDICATOR BACTERIA FROM ANIMALS AND FOOD

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

In NORM-VET, *Escherichia coli* and *Enterococcus* spp. serve as indicator bacteria. In 2003, indicator bacteria from cattle and sheep were included in the monitoring programme. In addition, data from a survey regarding reindeer are presented.

Sampling, laboratory methods and data processing are described in Appendix 3.

Escherichia coli from cattle and sheep

A total of 141 faecal and 157 meat samples from cattle and 137 faecal samples from sheep were collected. *E. coli* was isolated from 120 (85.1%) of the faecal samples from cattle, 90 (57.3%) of the meat samples from cattle and 118 (86.1%) of the faecal samples from sheep. These isolates were susceptibility tested. The results are presented in Table 13 and Figures 9 and 10.

TABLE 13. Antimicrobial resistance in *Escherichia coli* from meat samples from cattle (n=90) and from faecal samples from cattle (n=120) and sheep (n=118).

		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	≥ 512
Oxytetracycline	Cattle^{M}	9	[3.9-16.8]					4.4	31.1	48.9	6.7					8.9		
	Cattle^{F}	5	[1.9-10.6]					0.8	36.7	53.3	4.2					5.0		
	Sheep^{F}	0	[0.0-3.1]					1.7	76.3	22.0								
Chloramphenicol	Cattle^{M}	1	[0.0-6.0]							5.6	56.7	34.4	2.2	1.1				
	Cattle^{F}	< 1	[0.0-4.6]							3.3	50.8	44.2	0.8				0.8	
	Sheep^{F}	0	[0.0-3.1]						0.8	9.3	66.9	22.9						
Florfenicol	Cattle^{M}	0	[0.0-4.0]								30.0	64.4	5.6					
	Cattle^{F}	0	[0.0-3.0]								23.3	74.2	2.5					
	Sheep^{F}	0	[0.0-3.1]								44.1	53.4	2.5					
Ampicillin	Cattle^{M}	8	[3.2-15.4]						8.9	65.6	17.8				7.8			
	Cattle^{F}	3	[0.5-7.1]						8.3	71.7	17.5				2.5			
	Sheep^{F}	< 1	[0.0-4.6]						31.4	58.5	9.3			0.8				
Amoxi/clav***	Cattle^{M}	0	[0.0-4.0]							53.3	36.7	2.2	7.8					
	Cattle^{F}	0	[0.0-3.0]							39.2	56.7	1.7	2.5					
	Sheep^{F}	< 1	[0.0-4.6]							61.9	37.3				0.8			
Ceftiofur	Cattle^{M}	0	[0.0-4.0]			3.3	36.7	57.8	2.2									
	Cattle^{F}	0	[0.0-3.0]			0.8	33.3	63.3	2.5									
	Sheep^{F}	0	[0.0-3.1]			1.7	38.1	57.6	2.5									
Trimethoprim	Cattle^{M}	4	[1.2-11.0]				22.2	57.8	14.4	1.1					4.4			
	Cattle^{F}	0	[0.0-3.0]				23.3	55.0	18.3	3.3								
	Sheep^{F}	0	[0.0-3.1]				61.9	29.7	7.6		0.8							
Sulfamethoxazole	Cattle^{M}	16	[8.8-24.7]										77.8	6.7				15.6
	Cattle^{F}	10	[0.5-16.8]										74.2	14.2	1.7			10.0
	Sheep^{F}	< 1	[0.0-4.6]										93.2	5.9				0.8
Streptomycin	Cattle^{M}	17	[9.6-26.0]							2.2	23.3	53.3	4.4	2.2	2.2	6.7	4.4	1.1
	Cattle^{F}	13	[7.2-19.8]							0.8	30.8	51.7	4.2	0.8	2.5	3.3	3.3	2.5
	Sheep^{F}	2	[0.0-6.0]							5.1	66.9	23.7	2.5			1.7		
Gentamicin	Cattle^{M}	0	[0.0-4.0]					14.4	78.9	5.6	1.1							
	Cattle^{F}	0	[0.0-3.0]					23.3	71.7	3.3	1.7							
	Sheep^{F}	0	[0.0-3.1]					55.1	44.9									
Neomycin	Cattle^{M}	0	[0.0-4.0]					.		91.1	8.9							
	Cattle^{F}	< 1	[0.0-4.6]							97.5	1.7	0.8						
	Sheep^{F}	0	[0.0-3.1]							100.0								
Enrofloxacin	Cattle^{M}	0	[0.0-4.0]	42.2	56.7	1.1												
	Cattle^{F}	0	[0.0-3.0]	28.3	64.2	7.5												
	Sheep^{F}	0	[0.0-3.1]	44.1	52.5	3.4												
Nalidixic acid	Cattle^{M}	0	[0.0-4.0]						2.2	52.2	43.3	2.2						
	Cattle^{F}	0	[0.0-3.0]						3.3	41.7	52.5	1.7	0.8					
	Sheep^{F}	0	[0.0-3.1]						3.4	54.2	41.5	0.8						

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

* M=Meat, F=Faeces.

** CI = Confidence interval.

*** Amoxi/clav = Amoxicillin/clavulanic acid. Due to a possible methodological problem, the MIC-values for the combination amoxicillin/clavulanic acid are probably not reliable (the obtained MIC-values are likely too high) and interpretation of these data should therefore be done with caution.

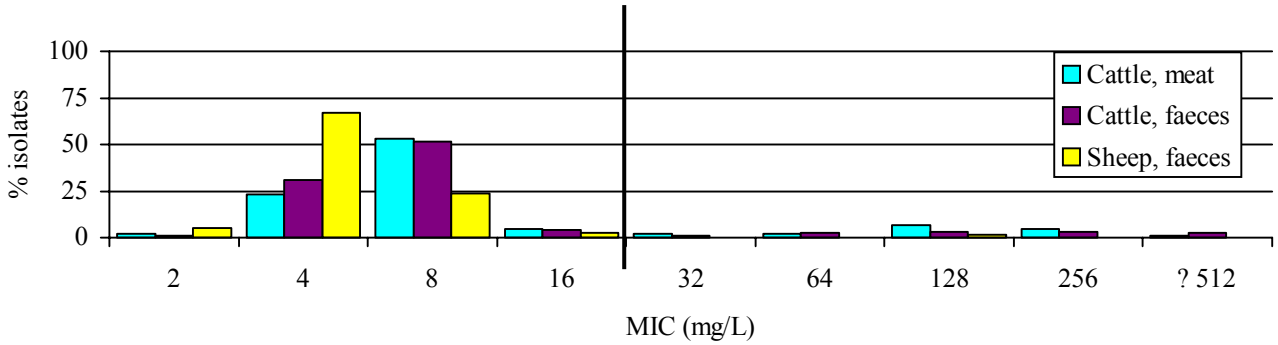

FIGURE 9. Distribution (%) of minimum inhibitory concentrations (MICs) for streptomycin among *E. coli* isolates from cattle and sheep. Microbiological cut-off value for streptomycin is shown as a black vertical line.

CATTLE

The data indicate a moderate occurrence of resistance among *E. coli* from faecal and meat samples from a representative group of Norwegian cattle presented for slaughter. In total, 84.2% and 73.3% of the isolates, respectively, were susceptible to all antimicrobials included. Altogether, 6.7% and 11.1% of the faecal and meat isolates, respectively, were resistant to one antimicrobial (predominantly streptomycin), 4.2% and 6.7%, respectively, to two antimicrobials (mainly streptomycin and sulfamethoxazoles) and 5.0% and 8.9%, respectively, to three or more antimicrobials. Resistance to streptomycin was most frequent in both categories (faeces and meat), followed by resistance to sulfamethoxazole, oxytetracycline and ampicillin. All these antimicrobials are commonly used for clinical purposes in cattle (sulfonamides in combination with trimethoprim). One faecal isolate was resistant to neomycin and one isolate from faecal and meat samples, respectively, was resistant to chloramphenicol. The aminoglycoside neomycin is approved for treatment of diarrhoea in calves. Veterinary drugs containing chloramphenicol, on the other hand, were withdrawn from the Norwegian market in 1992.

No resistance to the fluoroquinolone enrofloxacin or to the quinolone nalidixic acid was observed. The usage of fluoroquinolones in food producing animals in Norway is very limited. No resistance to ceftiofur or gentamicin was observed. No preparations containing cephalosporins or the aminoglycoside gentamicin have been approved for veterinary use in Norway.

SHEEP

The occurrence of resistance among *E. coli* from sheep faecal samples was low. In total, 97.5% of the isolates were susceptible to all antimicrobials included. Two isolates were resistant to one (streptomycin or ampicillin) and one to two antimicrobials (streptomycin and sulfamethoxazole). All these antimicrobials may be used for clinical purposes in sheep production. Lambs slaughtered at an age of about six months account for approximately 70% of the group of slaughtered sheep, and thus dominate the material included in NORM-VET 2003. Lamb production in Norway is very extensive and the lambs spend a large part of their lives roaming freely on rough, upland grazing. Consequently, the antimicrobial use in lambs is very limited, which is also reflected in the resistance prevalences observed.

Escherichia coli from wild reindeer

Faecal samples from 50 wild reindeer were included. *E. coli* was isolated from 42 of the samples, and these isolates were susceptibility tested. The results are presented in Table 14 and figure 10.

TABLE 14. Antimicrobial resistance in *Escherichia coli* isolated from faecal samples from reindeer (n=42).

	Resistance (%)		Distribution (%) of MIC values (mg/L)														
Substance	[95% CI*]		0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	7	[1.5-19.5]						52.4	40.5						7.1		
Chloramphenicol	0	[0.0-8.4]							16.7	52.4	31.0						
Florfenicol	0	[0.0-8.4]								54.8	42.9	2.4					
Ampicillin	0	[0.0-8.4]						14.3	76.2	9.5							
Amoxi/clav**	0	[0.0-8.4]							64.3	35.7							
Ceftiofur	0	[0.0-8.4]			2.4	16.7	81.0										
Trimethoprim	0	[0.0-8.4]				28.6	66.7	4.8									
Sulfamethoxazole	10	[2.7-22.6]											88.1	2.4			9.5
Streptomycin	24	[12.1-39.5]								33.3	40.5	2.4	2.4	9.5	9.5	2.4	
Gentamicin	0	[0.0-8.4]					50.0	45.2	4.8								
Neomycin	0	[0.0-8.4]							97.6	2.4							
Enrofloxacin	0	[0.0-8.4]	35.7	61.9	2.4												
Nalidixic acid	0	[0.0-8.4]							50.0	50.0							

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

RESULTS AND COMMENTS

The data indicate a moderate occurrence of resistance among *E. coli* from faecal samples from wild reindeer. In total, 76.2% of the isolates from reindeer were susceptible to all antimicrobials included. Six isolates (14.3%) were resistant to one antimicrobial (streptomycin), one (2.4%) to two (streptomycin and sulfa-methoxazole) and three (7.1%) to three antimicrobials (streptomycin, oxytetracycline and sulfamethoxazole). Resistance was more common in *E. coli* from wild reindeer (24% of isolates being resistant to at least one of the antimicrobials included) as compared to *E. coli* from other cervids presented in NORM/NORM-VET 2002 (2.2% of isolates being resistant to at least one of the antimicrobials included).

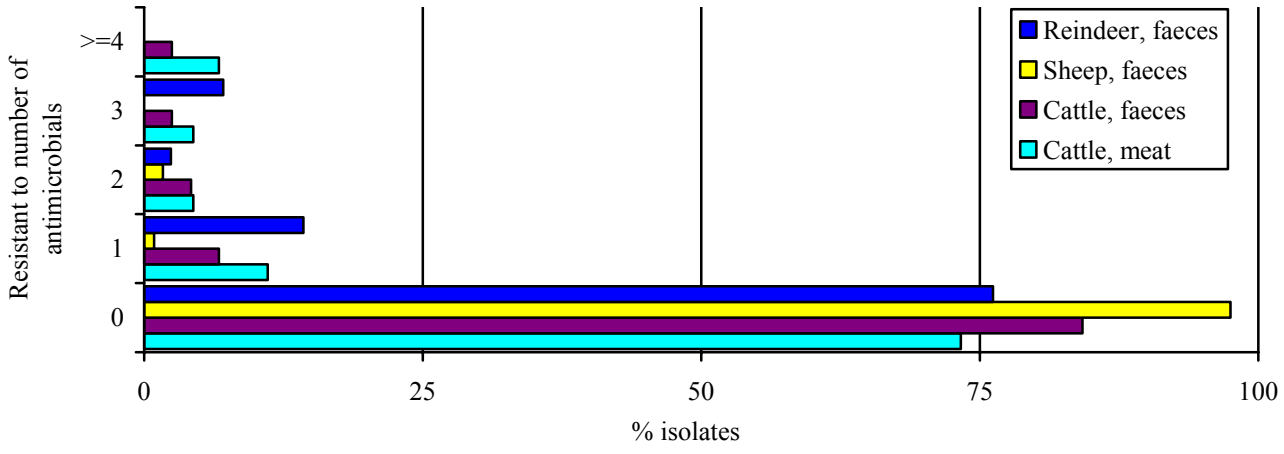

FIGURE 10. Antimicrobial resistance profile for *Escherichia coli* from faecal samples from reindeer (n=42), sheep (n=118) and cattle (n=120) and from meat samples from cattle (n=90). Proportion of isolates susceptible to all antimicrobials included and resistant to one, two, three, and four or more antimicrobials.

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Enterococcus spp. from cattle and sheep

A total of 141 faecal and 157 meat samples from cattle and 137 faecal samples from sheep were collected. *E. faecium* or *E. faecalis* was isolated from 11 (7.8%) of the cattle faecal samples, 108 (68.8%) of the cattle meat samples and three (2.2%) of the sheep faecal samples. The isolates were susceptibility tested. The results are presented in Tables 15 and 16.

TABLE 15. Antimicrobial resistance in *Enterococcus faecium* from faecal (n=5) and meat samples from cattle (n=18)

		Resistance	Distribution (number of isolates) of MIC values (mg/L)														
Substance	Sample	(number)	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	> 2048
Oxytetracycline	Faeces	0				4	1										
	Meat	1			1	14	2						1				
Chloramphenicol	Faeces	0							5								
	Meat	0						2	15	1							
Ampicillin	Faeces	0				3	2										
	Meat	0			3	5	6	2	2								
Erythromycin	Faeces	1					2	2	1								
	Meat	1			1		6	10		1							
Streptomycin	Faeces	0												5			
	Meat	0												18			
Gentamicin	Faeces	0												5			
	Meat	0												18			
Neomycin	Faeces	0								3	2						
	Meat	0								15	1	2					
Vancomycin	Faeces	0				5											
	Meat	0				10	4	4									
Bacitracin*	Faeces	0						1	1		3						
	Meat	1						1	2	6	8	1					
Avilamycin	Faeces	0					2	1	2								
	Meat	0				1	2	12	3								
Virginiamycin	Faeces	0			1	2	2										
	Meat	2			2	10	1	2	1	2							
Flavomycin**	Faeces	NR**												5			
	Meat	NR**						1						17			
Narasin	Faeces	0			2	3											
	Meat	0		2	11	4	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.
** Not relevant, as *E. faecium* is inherently resistant to flavomycin.

TABLE 16. Antimicrobial resistance in *Enterococcus faecalis* from meat samples from cattle (n=90).

	Resistance (%)		Distribution (%) of MIC values (mg/L)														
Substance	[95% CI*]		0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	1024	> 2048
Oxytetracycline	8	[3.2-15.4]			12.2	72.2	5.6	2.2		2.2	1.1	2.2	2.2				
Chloramphenicol	0	[0.0-4.0]					2.2	35.6	62.2								
Ampicillin	0	[0.0-4.0]			22.2	73.3	4.4										
Erythromycin	1	[0.0-6.0]			12.2	10.0	54.4	22.2	1.1								
Streptomycin	6	[1.8-12.5]												94.4			5.6
Gentamicin	0	[0.0-4.0]												100			
Neomycin	1	[0.0-6.0]								17.8	33.3	31.1	15.6	1.1		1.1	
Vancomycin	0	[0.0-4.0]				13.3	61.1	25.6									
Bacitracin**	2	[0.0-7.8]				1.1	1.1	5.6	24.4	60.0	5.6	2.2					
Avilamycin	0	[0.0-4.0]			2.2	17.8	61.1	14.4	4.4								
Virginiamycin[#]	NR[#]								7.8	64.4	27.8						
Flavomycin	1	[0.0-6.0]				15.6	1.1	35.6	35.6	4.4	6.7	1.1					
Narasin	0	[0.0-4.0]	10.0	61.1	24.4	3.3	1.1										

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.
*Confidence interval.
** Measured in U/ml.
[#] Not relevant, as *E. faecalis* is inherently resistant to virginiamycin.

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. The situation is reversed for flavomycin. The use of virginiamycin in animal production in Norway has been negligible, and the substance was banned in 1998. Flavomycin has never been approved in Norway. Resistance to virginiamycin and flavomycin is not included in the following discussion.

SHEEP

All three isolates from faecal samples, two *E. faecalis* and one *E. faecium*, were susceptible to all the antimicrobials included

CATTLE

Of the 90 *E. faecalis* isolates from meat samples, 88.9% were susceptible to all antimicrobials included. In total, five isolates (5.6%) were resistant to one antimicrobial (oxytetracycline, bacitracin or erythromycin) and five (5.6%) were resistant to two or more antimicrobials (four resistant to oxytetracycline and streptomcyin and one isolate also to neomycin). Some tetracycline and streptomycin is used for clinical purposes in Norwegian cattle.

E. faecalis was isolated from only six faecal samples. Of these six isolates, three were susceptible to all antimicrobials included, whereas two were resistant to one antimicrobial (erythromycin or streptomycin).

Of the 18 *E. faecium* isolates from meat samples, 15 were susceptible to all antimicrobials included. Three isolates were resistant to one antimicrobial (tetracycline, erythromycin or bacitracin).

Of the five isolates of *E. faecium* from cattle faeces, only one was resistant to one antimicrobial (erythromycin).

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C. ZOONOTIC AND OTHER ENTEROPATHOGENIC BACTERIA

Zoonotic and other enteropathogenic bacteria represent a considerable public health problem. Furthermore, the increasing occurrence of antimicrobial resistance in such bacteria is a major public health concern. Therefore, it is of uttermost 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 feed, animals and food are monitored for antimicrobial resistance, as well as a representative number of *Campylobacter* isolates from broiler and broiler meat. Regarding human isolates, all *Salmonella*, *Shigella* and *Yersinia enterocolitica* isolates are monitored, as well as a representative number of *Campylobacter* isolates. Sampling, laboratory methods and data processing are described in Appendix 4.

SALMONELLA SPP.

Salmonella from animals

The situation regarding *Salmonella* spp. in food production animals in Norway is very good. Such animals are virtually free from *Salmonella* spp. except for an endemic occurrence of *S. enterica* subsp. *diarizonae* in sheep. To document this favourable situation, Norway runs an extensive surveillance programme that covers both live animals (cattle, pigs and poultry) and meat (cattle, pigs, sheep and poultry). The *Salmonella* isolates examined in NORM-VET include those that are detected in this programme in addition to isolates from other relevant projects as well as clinical submissions to the National Veterinary Institute. The data are presented in Table 17.

TABLE 17. Antimicrobial resistance in *Salmonella* spp. (n=14), *S.* Typhimurium (n=10) and other *Salmonella* spp. (n=4).

	Resistance	Distribution (number of isolates) of MIC values (mg/L)														
Substance	(number)	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	1						6	7					1			
Chloramphenicol	0							1	11	2						
Florfenicol	0								9	5						
Ampicillin	1					2	11						1			
Amoxi/clav*	1							13					1			
Ceftiofur	0				1	2	11									
Trimethoprim	1				2	10		1					1			
Sulfamethoxazole	1										3	8	2			1
Streptomycin	1								2	4	6	1				1
Gentamicin	1					10	3							1		
Neomycin	1							13				1				
Enrofloxacin	0	2	2	10												
Nalidixic acid	0								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.
*Amoxicillin/clavulanic acid

RESULTS AND COMMENTS

In 2003, only five isolates of *Salmonella* were detected in the national surveillance programme; three isolates of *S.* Typhimurium (from poultry, cattle and pig, respectively), one isolate of *S.* Senftenberg (cattle) and one isolate of *S.* Hessarek (pig). All five isolates were susceptible to all antimicrobials included. In addition, one *S. enterica* subsp. *diarizonae* (61:k:1,5,7) from crushed meat was detected in the surveillance programme. This isolate was not susceptibility tested based on the experience from previous years that this type of bacterium generally is pan-susceptible.

Also seven clinical isolates of *S.* Typhimurium (from one horse (DT42), three cats, two dogs and one roe-deer, respectively) and two isolates of *S.* Gallinarum/Pullorum (from poultry) were tested. All isolates were susceptible to all antimicrobials included, except for the isolate from the horse, which was multiresistant.

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

Salmonella from human clinical specimens

In 2003, 1,539 human cases of salmonellosis, excluding typhoid and paratyphoid fever, were reported in Norway (incidence rate 33.8 per 100,000). In 80% of the cases, the infection was reported as having been acquired abroad, whereas for 15.9% the infection was classified as domestically acquired. For the remaining cases, the place of acquisition was unknown.

For *S.* Enteritidis (55% of the cases), the proportion of cases reported as imported was particularly high (90%). *S.* Enteritidis has never been detected in Norwegian poultry. For *S.* Typhimurium (15% of the cases), approximately 41% of the infections were acquired in Norway, which is partly explained by the endemic occurrence of specific clones of this serovar in Norwegian wildlife. Data from surveillance programmes show that domestically produced food is not an important source of human salmonellosis acquired in Norway. The most likely sources in these cases are direct or indirect contamination from wildlife, imported food products, or secondary infections from other patients. Thus, the isolates categorized as "infected in Norway" also partly reflect the resistance situation outside Norway.

For the *S.* Typhimurium infections acquired domestically, only three (3.1%) of the isolates were caused by multiresistant DT104, as opposed to 13 (10.4%) for those *S.* Typhimurium infections acquired abroad. The incidence of multiresistant *S.* Typhimurium DT104 infection, especially domestically acquired cases, was reduced markedly in 2003 as compared to 2002.

In 2003, 239 isolates of *S.* Typhimurium, 869 isolates of *S.* Enteritidis, 14 isolates of S. Typhi, 13 isolates of *S.* Paratyhi A, 25 isolates of *S.* Paratyphi B, and 435 isolates of other *Salmonella* spp. were susceptibility tested. The results are presented in Figure 11, Tables 18-25, and in the text.

TABLE 18. *Salmonella* Typhimurium isolates (n=79), including DT104 (n=3), from patients infected in Norway. Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	86.1	0.0	13.9	6	-	30
Chloramphenicol	≥ 20	≤ 19	94.9	-	5.1	6	-	29
Ampicillin	≥ 32	≤ 12	0.0	87.3	12.7	6	-	31
TMS**	≥ 20	≤ 12	97.5	0.0	2.5	6	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	100.0	0.0	0.0	27	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	97.5	-	2.5	6	-	28

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

TABLE 19. *Salmonella* Typhimurium isolates (n=79) including DT104 (n=3) from patients infected in Norway. Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	6.3	2.5	1.3	1.3	2.5											2.5
Chloramph.	5.1															
Ampicillin	12.7															1.3
TMS**	2.5															
Ciprofloxacin																
Nalidixic acid	2.5															

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	3.8	1.3	17.7	17.7	25.3	7.6	8.9		1.3						
Chloramph.		1.3	13.9	31.6	22.8	11.4	11.4	2.5							
Ampicillin			1.3	3.8	12.7	8.9	27.8	11.4	15.2	5.1					
TMS**				2.5		1.3	1.3	2.5	5.1	7.6	21.5	11.4	19.0	7.6	17.7
Ciprofloxacin						2.5	1.3						12.7	5.1	78.5
Nalidixic acid	3.8	3.8	17.7	26.6	30.4	6.3	8.9								

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

TABLE 20. *Salmonella* Typhimurium isolates (n=119) including DT104 (n=13) from patients infected outside Norway. Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)
	S	R	S	I	R	
Tetracycline	≥ 20	≤ 16	53.8	3.4	42.9	6 - 30
Chloramphenicol	≥ 20	≤ 19	72.3	-	27.7	6 - 29
Ampicillin	≥ 32	≤ 12	0.0	65.5	34.5	6 - 31
TMS**	≥ 20	≤ 12	90.8	0.0	9.2	6 - ≥ 36
Ciprofloxacin	≥ 27	≤ 18	96.6	3.4	0.0	24 - ≥ 36
Nalidixic acid	≥ 17	≤ 16	89.9	-	10.1	6 - ≥ 36

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

TABLE 21. *Salmonella* Typhimurium isolates (n=119) including DT104 (n=13) from patients infected outside Norway. Distribution (%) of zone diameters (mm)*.

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	18.5	4.2	5.9	5.9	7.6	0.8						1.7	1.7		0.8	1.7
Chloramph.	26.9													0.8		
Ampicillin	34.5															0.8
TMS**	9.2															
Ciprofloxacin																
Nalidixic acid	10.1											0.8				1.7

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	1.7	5.9	6.7	10.9	15.1	7.6	2.5		0.8						
Chloramph.	3.4	7.6	15.1	21.8	16.8	4.2	1.7	1.7							
Ampicillin	0.8		1.7	10.1	6.7	4.2	19.3	13.4	7.6	0.8					
TMS**		1.7	1.7	0.8	3.4	2.5	9.2	2.5	10.1	7.6	16.0	11.8	16.0	3.4	4.2
Ciprofloxacin			0.8	1.7	0.8	0.8	0.8	5.0	3.4	0.8	0.8	5.0	10.9	9.2	59.7
Nalidixic acid	5.0	6.7	16.8	22.7	22.7	10.9	1.7								0.8

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

TABLE 22. *Salmonella* Enteritidis isolates from patients (n=869[#]). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)
	S	R	S	I	R	
Tetracycline	≥ 20	≤ 16	97.0	0.1	2.9	6 - ≥ 36
Chloramphenicol	≥ 20	≤ 19	98.8	-	1.2	7 - ≥ 36
Ampicillin	≥ 32	≤ 12	1.4	95.1	3.6	6 - ≥ 36
TMS**	≥ 20	≤ 12	97.9	0.5	1.6	6 - ≥ 36
Ciprofloxacin	≥ 27	≤ 18	93.1	6.9	0.0	20 - ≥ 36
Nalidixic acid	≥ 17	≤ 16	73.4	-	26.6	6 - ≥ 36

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.
Place of infection; Norway (n=54), Abroad (n=748), Unknown (n=67).

TABLE 23. *Salmonella* Enteritidis isolates from patients (n=869[#]). Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	0.9	1.8								0.1			0.1		0.8	0.6
Chloramph.		0.1								0.1		0.5	0.5		0.3	
Ampicillin	3.6									0.1	0.2	0.5	0.8		0.1	0.5
TMS**	1.6											0.3		0.1		
Ciprofloxacin															0.1	
Nalidixic acid	26.4			0.1						0.1			0.1		0.6	0.8

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	1.6	2.4	6.8	13.6	19.3	18.8	21.4	5.3	5.2	0.1	0.8				0.3
Chloramph.	0.8	2.0	9.7	25.8	29.9	17.3	9.2	1.0	1.7	0.1	0.3			0.3	0.3
Ampicillin	0.8	1.6	3.7	6.2	10.7	13.2	35.8	10.6	9.7	0.6	1.0			0.2	0.1
TMS**		0.1	0.1	0.7	1.0	0.8	3.2	2.5	11.4	10.7	22.2	11.5	19.3	5.9	8.4
Ciprofloxacin		0.5	0.7	2.2	3.5	3.3	5.9	3.6	6.1	0.9	2.3	2.4	7.9	6.7	54.0
Nalidixic acid	2.0	6.4	18.3	23.6	15.7	3.9	1.3	0.3	0.1				0.1		0.2

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
**TMS=Trimethoprim/sulfamethoxazole.
Place of infection; Norway (n=54), Abroad (n=748), Unknown (n=67).

TABLE 24. *Salmonella* spp. (excluding *S.* Typhimurium, *S.* Enteritidis, *S.* Typhi and *S.* Paratyphi) (n=435[#]). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	74.0	0.2	25.7	6	-	34
Chloramphenicol	≥ 20	≤ 19	90.8	-	9.2	6	-	≥ 36
Ampicillin	≥ 32	≤ 12	1.6	88.0	10.3	6	-	≥ 36
TMS**	≥ 20	≤ 12	89.0	0.5	10.6	6	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	93.3	6.7	0.0	21	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	79.1	-	20.9	6	-	32

*S=Susceptible, I=Intermediately susceptible, R=Resistant. **TMS=Trimethoprim/sulfamethoxazole.
Place of infection; Norway (n=56), Abroad (n=319), Unknown (n=60).

TABLE 25. *Salmonella* spp. (excluding *S.* Typhimurium, *S.* Enteritidis, *S.* Typhi and *S.* Paratyphi) (n=435). Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	10.6	6.7	6.9	0.9	0.2	0.5							0.2		1.4	1.4
Chloramph.	5.5	0.9	0.2	0.2	0.2		0.2				0.5	0.5	0.5	0.5	0.5	0.7
Ampicillin	10.3								0.5	0.2			0.2		0.2	
TMS**	10.3				0.2								0.5			
Ciprofloxacin																0.2
Nalidixic acid	19.5		0.2						0.5	0.2	0.5	0.7	0.5	0.2	1.4	2.1

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	1.4	3.9	9.7	14.5	15.2	13.1	9.2	1.6	2.1		0.2		0.5		
Chloramph.	1.1	4.6	12.6	28.5	17.2	9.7	7.8	2.5	4.1	0.2	0.2	0.2	0.2		0.5
Ampicillin	1.4	2.1	2.3	3.7	7.8	11.7	29.2	12.9	14.5	1.4	0.9	0.5			0.2
TMS***		0.2	0.7	1.1	1.6	2.1	3.7	4.1	11.0	6.9	20.5	9.9	15.6	3.9	7.6
Ciprofloxacin	0.2	0.2	1.1	1.4	3.4	4.4	6.0	2.3	3.2	0.7	1.6	3.9	7.8	9.2	54.3
Nalidixic acid	2.1	4.1	14.7	16.8	21.1	8.5	5.3	0.7	0.5	0.2	0.2				

*Shaded areas in each row indicate susceptibility (light), intermediate susceptibility (medium) and resistance (dark).
**TMS=Trimethoprim/sulfamethoxazole. #Place of infection; Norway (n=56), Abroad (n=319), Unknown (n=60)

RESULTS AND COMMENTS

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

The proportion of *S.* Typhimurium isolates (excluding multiresistant DT104 isolates) susceptible to all antimicrobials was higher for the category "infected in Norway" (88.2%) than for the "infected abroad" category (59.4%) (Figure 11). Moreover, multiresistance (resistance to more than two antimicrobials) was more common in the category "infected abroad" (19.8%) as compared to the category "infected in Norway" (5.3%). A significant discrepancy for the two categories (including DT104 isolates) was observed for quinolones; in the category "infected abroad", 10.1% of the isolates were resistant to nalidixic acid as opposed to 2.5% among those from patients "infected in Norway". However, the very few isolates of multiresistant DT104 are accounting for the majority of this resistance, and by excluding DT104 the differences between the categories are not quite as striking with 4.7% in the category "infected abroad" and 1.3% in the category "infected in Norway" being resistant to nalidixic acid. None of the non-DT104 isolates in the category "infected in Norway" showed reduced susceptibility to ciprofloxacin, whereas 0.9% of the isolates in the category "infected abroad" showed reduced susceptibility. It is emphasized that the use of fluoroquinolones in Norway is limited in both human and veterinary medicine.

The vast majority of *S.* Enteritidis isolates had been acquired abroad. The proportion of *S.* Enteritidis isolates resistant to tetracycline, chloramphenicol and ampicillin, respectively, was considerably lower than for *S.* Typhimurium, including those *S.* Typhimurium infections acquired in Norway. Resistance to nalidixic acid on the other hand, was more widespread among *S.* Enteritidis as compared to *S.* Typhimurium. Of the nalidixic acid resistant *S.* Enteritidis isolates, 6.9% showed reduced susceptibility to ciprofloxacin indicating that fluoroquinolone resistance could be developing. In total, all of the isolates intermediately susceptible to ciprofloxacin were also resistant to nalidixic acid. The resistance frequencies observed for *S.* Enteritidis in NORM/NORM-VET 2003 are in accordance with those observed in NORM/NORM-VET 2002 also when taking into account the change of breakpoints applied.

Altogether, 14 isolates of *S.* Typhi, 13 isolates of *S.* Paratyphi A and 25 of *S.* Paratyhi B were susceptibility tested. The majority of these infections had been acquired outside Norway. Only five cases were aquired domestically. Among the isolates of *S.* Paratyphi B, all but one isolate (multiresistant) were susceptible to all antimicrobials included, whereas a higher proportion of the isolates of *S.* Paratyphi A and *S.* Typhi were resistant to one or more of the antimicrobials included. Only three of 13 isolates of *S.* Paratyphi A were susceptible to all antimicrobials included, as opposed to 10 of 14 *S.* Typhi isolates. Of the 27 isolates of *S.* Paratyphi A and *S.* Typhi,

four isolates were resistant to one antimicrobial (nalidixic acid), and 10 isolates to two or more antimicrobials, of which six were resistant to four antimicrobials (except to nalidixic acid and ciprofloxacin).

With regard to isolates of *Salmonella* spp. other than Typhimurium, Enteritidis, Typhi and Paratyphi, the vast majority of infections had been acquired abroad. Resistance was quite widespread. Resistance to tetracycline was most common, followed by resistance to nalidixic acid, trimethoprim-sulfamethoxazole, ampicillin and chloramphenicol. The prevalence of resistance to nalidixic acid was relatively high (20.9%). Similar to what was observed for *S.* Enteritidis isolates, although ciprofloxacin resistance was not detected, 6.7% showed reduced susceptibility to ciprofloxacin indicating that fluoroquinolone resistance could be developing.

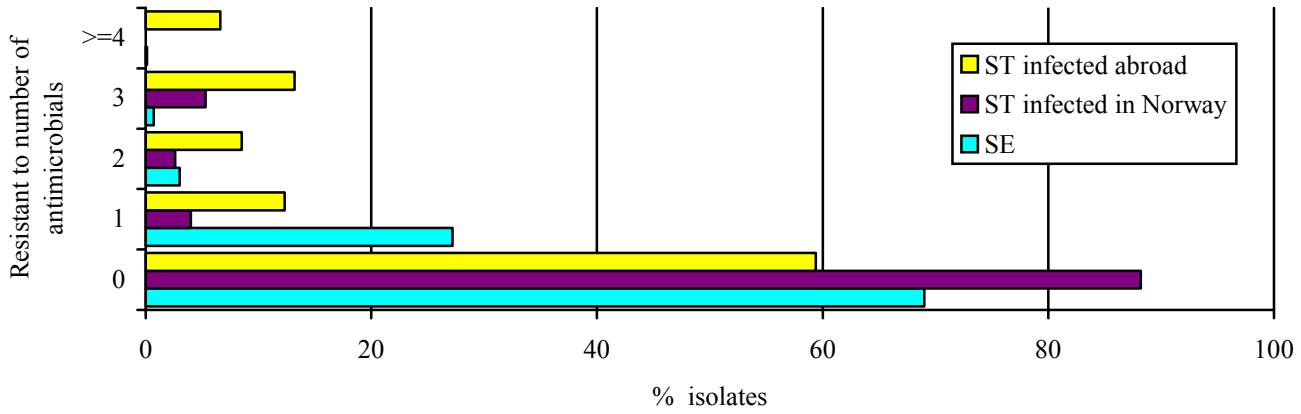

FIGURE 11. Antimicrobial resistance profiles for all *Salmonella* Enteritidis from humans (n=869) and for *Salmonella* Typhimurium (excluding DT104) from humans infected in Norway (n=76) and infected abroad (n=106). Proportion of isolates susceptible to all antimicrobials included and resistant to one, two, three, or four or more antimicrobials.

Studies on mutations that cause quinolone resistance and surveillance of integrons in *Salmonella enterica*.

Quinolone resistance

The quinolones are a class of antibiotic agents that act by inhibiting the action of type II topoisomerases, DNA gyrase and topoisomerase IV[1-3]. DNA gyrase is a tetrameric enzyme composed of two A subunits and two B subunits, encoded by *gyrA* and *gyrB* respectively. Topoisomerase IV is an A_2B_2 enzyme as well, encoded by *parC* and *parE*. In bacteria these enzymes change the topology of DNA by cleaving both strands and allow one doubled-stranded DNA molecule to pass through another and then reseal the break[1-3]. Quinolones inhibit these enzymes by stabilising the complex between DNA and DNA gyrase or topoisomerase IV and thus block progression of polymerase and DNA replication.

An increase in quinolone resistance among *Salmonellae* has been reported. Most notably is the rise in quinolone resistance in the *Salmonella enterica* serovars Enteritidis (*S.* Enteritidis), Hadar (*S.* Hadar), Typhimurium (*S.* Typhimurium) and Virchow (*S.* Virchow)[4-6]. All of these serotypes have their primary reservoirs in food animals and their primary method of spread is likely to be through the food chain.

A Finnish study showed reduced quinolone susceptibility in *Salmonella enterica* from travellers returning from Southeast Asia[4]. The most common serotypes with reduced ciprofloxacin susceptibility were *S.* Hadar, *S.* Typhimurium, *S.* Virchow and *S.* Enteritidis. The data for Thailand were especially prominent showing an increase in quinolone-resistant isolates from 5.6% in 1995 to 50% in 1999[4].

A recent study from the Norwegian Institute of Public Health showed that mutations in *gyrA* codons 83 and 87 are the primary means of nalidixic acid resistance in *S.* Enteritidis and *S.* Hadar[7]. All the examined isolates displayed at least one mutation at codon 83 or 87, which are well known mutational sites associated with quinolone resistance.

Results from broth-dilution MIC assays showed that isolates with mutations in *gyrA* had elevated MIC values for ciprofloxacin with MIC values equal to or higher than 4 mg/L for all *gyrA* mutated isolates[7]. The NCCLS resistance breakpoint for ciprofloxacin is set at ≥4 mg/L for *Enterobacteriaceae* making all the *gyrA*-mutated isolates in this study resistant to ciprofloxacin. The tablet diffusion method, however, resulted in inhibition zones that were not considered as fully resistant for any of the mutated isolates towards ciprofloxacin. The majority of these isolates did however fall into inhibition zone ranges that were classified as reduced susceptible. To obtain fluoroquinolone resistance it is generally believed that a *gyrA* double mutation in codon83/codon87 coupled with a *parC* mutation is probably required. In contrast to this, our nalidixic acid resistant isolates, including single *gyrA* mutations, all showed resistance to ciprofloxacin by the broth dilution assay, indicating a higher degree of cross-resistance to fluoroquinolones by single codon mutations in nalidixic acid resistant strains than previously presumed[7].

Very unexpected was the discovery that mutations in *S.* Hadar showed a very strong correlation between mutations at codon 83

and isolates from Southeast Asia, and codon 87 mutations with isolates from mainly Southern Europe and North Africa ($P<0.0001$ in Fisher's exact test)[7]. Mutations at *gyrA* codon 83 was found in 95.5% (21/22) of the Southeast Asian isolates, while only 4.5% (1/22) of the Southeast Asian isolates had codon 87 mutations. The isolates from mainly Southern Europe and North Africa displayed mutations at codon 87 in 96.7% (29/31) of the isolates, whereas these isolates only displayed codon 83 mutations in 3.2% (1/31) of the isolates[7]. It is quite unlikely that the geographic location *per se* is the cause of the different mutational targets we observed in *S*. Hadar. A more likely explanation would be that the *S*. Hadar isolates with codon 83 and codon 87 mutations have been exposed to different quinolone antibiotics at sublethal concentrations in East Asia and Europe/North Africa.

Surveillance of integrons

Integrons are genetic units that include components of a site-specific recombination system enabling them to capture and mobilize genes[8,9]. Integrons contain a gene of the λ family of integrases that carries out recombination between two distinct target sites; the *attI* site and the 59-base element where *attI* is the target site for cassette integration[8,9]. The genes captured by this system are packaged as small discrete units with essentially all flanking sequences removed but with the 59-base element included, and are named 'gene cassettes' [8,9]. The majority of known gene cassettes are antibiotic resistance genes, but it is likely that any gene could be included in a cassette and several uncharacterized open reading frames have been found in cassettes. Integrons are classified by comparison of the amino acid sequence of their integrases, coded for by the *intI* genes.

In a recent study of class-I integrons and the genetic units that control their expression (promoters), 156 multi-resistant strains of the important zoonotic pathogens *S.* Typhimurium and *S.* Enteritidis from human clinical isolates submitted to the Reference Laboratory for Enteropathogenic Microbes at the Norwegian Institute of Public Health were characterized by DNA sequencing[10]. Integrons were found in 64 of 66 *S.* Typhimurium isolates (97%) and in 20 of 90 *S.* Enteritidis isolates (22.2%) with the following sizes; 650bp, 1,000bp, 1,200bp, 1,500bp, 1,600bp, 1,700bp, 2,000bp and 2,100bp[10]. Our integron sequences were compared with GenBank sequences and the *aadA1*, *aadA2*, *aadA5*, *aadB*, *pse-1*, c*atB3*, *oxa1*, *dfrA1*, *dfrA12, dfrA17* antibiotic resistance genes as well as a fragment of the *sat1* gene were identified based on DNA homology[10]. An internal fragment of the *purG* gene was additionally found as an artifact PCR amplicon. Integrons have been located within transposons, and all isolates were additionally tested for the presence of the mercury resistance gene *merA* that is associated with the transposon *Tn*21 as well as others. The *merA* gene was found in 27 of the integron carrying strains indicating the presence of transposons, which will give the integrons additional mobility[10]. Searches for integrons in all multi-resistant *Salmonella* isolates that are submitted to the Reference Laboratory for Enteropathogenic Microbes are performed, and the integrons are characterized by DNA sequencing. During the last year and the first months of 2004, integrons were found in the *Salmonella* serovars; Enteritidis, Typhimurium, Concord, Saintpaul, Paratyphi A, Paratyphi B, Scwarzengrund, Albany, Panama, Newport, Virchow, Stanley and Derby. The resistance genes and promoters found were the same as in our previous study indicating that the *Salmonella* integron pool is fairly stable.

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CAMPYLOBACTER SPP.

Campylobacter jejuni from broilers

The isolates of *Campylobacter jejuni* in broilers originate from the Norwegian action plan against *Campylobacter* spp. in broilers (www.zoonose.no). All broiler flocks slaughtered before 50 days of age are tested for the presence of *Campylobacter* spp. In addition, 100 samples of broiler meat products from retail level are tested monthly. In 2003, one isolate per positive farm as well as one isolate from each batch of positive broiler meat products were submitted for susceptibility testing.
A total of 139 isolates, 108 from cloacal samples and 31 from broiler meat, were susceptibility tested. The results are presented in Table 26 and Figures 12 and 13.

TABLE 26. Antimicrobial resistance in *Campylobacter jejuni* (n=139) from broiler cloacal samples (n=108) and from broiler meat products (n=31).

	Resistance (%)		Distribution (%) of MIC values (mg/L)														
Substance	[95% CI*]		0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	≥ 512
Oxytetracycline	2	[0.5-6.2]				97.1	0.7							2.2			
Ampicillin	3	[0.8-7.2]					5.0	7.2	33.8	37.4	10.1	3.6	2.2	0.7			
Erythromycin	0	[0.0-2.6]			1.4	6.5	44.6	43.2	4.3								
Gentamicin	0	[0.0-2.6]				5.0	21.6	51.8	21.6								
Enrofloxacin	1	[0.2-5.1]	4.3	14.4	72.7	7.2				0.7	0.7						
Nalidixic acid	1	[0.2-5.1]						0.7	7.9	71.2	18.7				0.7	0.7	

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

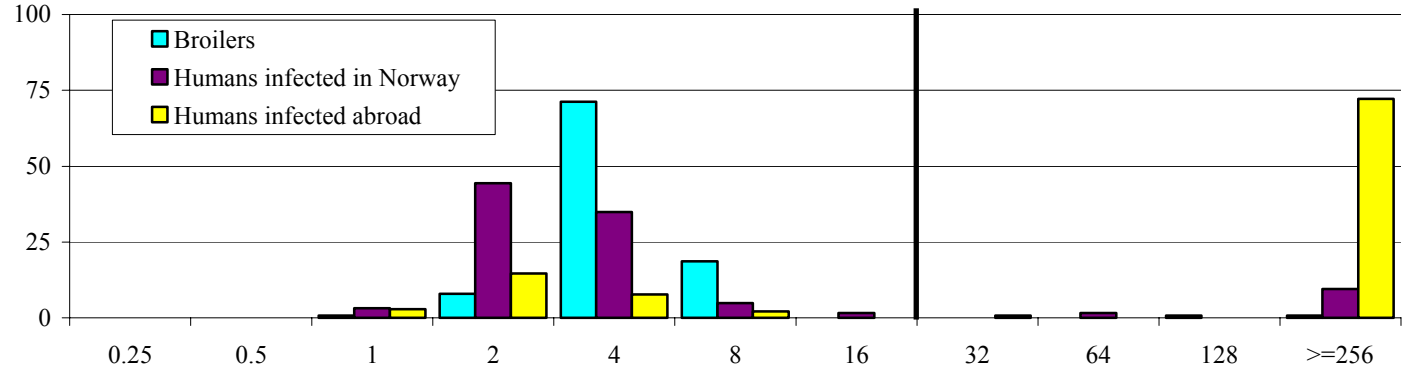

FIGURE 12. Distribution (%) of minimum inhibitory concentrations (MICs) for nalidixic acid among *Campylobacter jejuni* isolates from broilers and humans. The microbiological cut-off value/breakpoint for nalidixic acid is shown as a black vertical line.

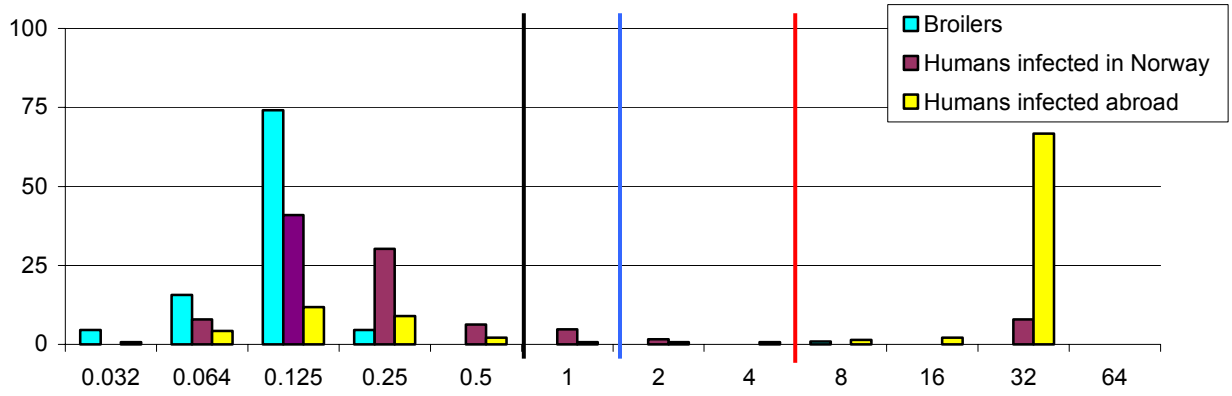

FIGURE 13. Distribution (%) of minimum inhibitory concentrations (MICs) for ciprofloxacin/enrofloxacin among *Campylobacter jejuni* isolates from broilers and humans. The microbiological cut-off value for enrofloxacin is shown as a black vertical line and the AFA breakpoints for ciprofloxacin are shown in red (breakpoint for resistance) and blue (breakpoint for susceptibility).

RESULTS AND COMMENTS

The results show that the prevalence of resistance among *C. jejuni* isolates from Norwegian broilers is low. A total of 95.0% of the isolates tested were susceptible to all antimicrobials included. Altogether 3.6% were resistant to one antimicrobial (ampicillin or oxytetracycline) and 1.4% to three antimicrobials (oxytetracycline, nalidixic acid and ciprofloxacin). The results reflect the usage of antimicrobials in poultry production. Antimicrobials (except coccidiostats) are rarely used, and only for therapeutical purposes. If used, amoxicillin (cross-resistance with ampicillin) and tetracycline are the drugs of choice. However, several quinolone preparations are licensed for use in poultry in the EU, and may therefore perhaps also be used in poultry production in Norway if specifically applied for.

The results are similar to those presented in previous NORM/NORM-VET reports (2001 and 2002).

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

Campylobacter spp. from human clinical specimens

Of the 2,270 cases of human campylobacteriosis recorded in Norway in 2003 (incidence rate 49.9 per 100,000), 53% were reported as acquired abroad. The vast majority of cases were sporadic. Norwegian case-control studies have revealed that consumption of broiler meat purchased fresh and drinking untreated water are important risk factors for domestically acquired campylobacteriosis.

A total of 207 isolates of *C. jejuni*, 63 from patients infected in Norway and 144 from patients infected abroad, as well as 16 isolates of *C. coli* were susceptibility tested. Due to the selection procedure where equal numbers of cases are selected each month, the numbers of Norwegian cases (highest incidence during the summer months) are under-represented in the material. The results are presented in Tables 27-30, Figures 12-14 and in the text.

TABLE 27. *Campylobacter jejuni* isolates from patients infected in Norway (n=63). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)			MIC_{50}	MIC_{90}
	S	R	S	I	R					
Doxycycline	≤ 2	≥ 4	90.5	-	9.5	0.064	-	48	0.19	0.75
Erythromycin	≤ 0.5	≥ 8	33.3	65.1	1.6	0.125	-	64	0.75	1.5
Gentamicin	≤ 4	≥ 8	100.0	-	0.0	0.125	-	4	0.38	1
Ciprofloxacin	≤ 1	≥ 4	90.5	1.6	7.9	0.047	-	32	0.19	1
Nalidixic acid	≤ 16	≥ 32	88.9	-	11.1	1	-	256	3	64

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

TABLE 28. *Campylobacter jejuni* isolates from patients infected in Norway (n=63). Distribution (%) of MICs (mg/L).*

Substance	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Doxycycline		6.3	41.0	28.5	12.7	1.6		3.2		1.6	3.2	1.6		
Erythromycin			1.6	1.6	30.1	50.8	11.1	3.2				1.6		
Gentamicin			6.3	26.9	46.1	12.7	3.2	4.8						
Ciprofloxacin		7.9	41.0	30.2	6.3	4.8	1.6				7.9			
Nalidixic acid						3.2	44.4	34.9	4.8	1.6		1.6		9.5

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

TABLE 29. *Campylobacter jejuni* isolates from patients infected outside Norway (n=144). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Doxycycline	≤ 2	≥ 4	37.5	-	62.5	0.032 - 128	8	48
Erythromycin	≤ 0.5	≥ 8	40.3	56.9	2.8	0.125 - 8	0.75	2
Gentamicin	≤ 4	≥ 8	96.5	-	3.5	0.047 - 16	0.25	1
Ciprofloxacin	≤ 1	≥ 4	28.5	0.7	70.8	0.032 - 32	32	32
Nalidixic acid	≤ 16	≥ 32	27.1	-	72.9	0.75 - 32	256	256

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

TABLE 30. *Campylobacter jejuni* isolates from patients infected outside Norway (n=144). Distribution (%) of MICs (mg/L).*

Substance	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Doxycycline	0.7		9.7	16.0	6.3	1.4	3.5	7.0	6.3	18.7	18.0	9.8	0.7	2.1
Erythromycin			0.7	8.3	31.2	40.3	13.8	2.8	0.7					2.1
Gentamicin		4.9	21.5	23.6	29.9	13.2	2.8	0.7	0.7	2.1				0.7
Ciprofloxacin	0.7	4.2	11.8	9.0	2.1	0.7	0.7	0.7	1.4	2.1	66.7			
Nalidixic acid						2.8	14.6	7.7	2.1		0.7			72.2

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

RESULTS AND COMMENTS

The data show that resistance was significantly more widespread among *C. jejuni* isolates derived from patients infected abroad. Only 16% of these isolates were susceptible to all antimicrobials included as opposed to 84.1% of the isolates from patients infected in Norway (Figure 14). These discrepancies are explained by the widespread occurrence among isolates acquired abroad of resistance to ciprofloxacin/nalidixic acid (70.8%/72.9% versus 7.9%/11.1%/) and to tetracycline (62.5% versus 9.5%) (Tables 27 and 29 and Figures 12-13).

The resistance frequencies for domestically acquired human isolates are in accordance with data for Norwegian broilers, although resistance to quinolones was more prevalent among the human isolates (Figures 12-14).

Only two of the 16 isolates of *C. coli* were acquired in Norway. In total, 11 of the *C. coli* isolates were resistant to at least one of the antimicrobials included (quinolones and/or doxycycline). Seven isolates were resistant to both nalidixic acid and ciprofloxacin, and four of these were also resistant to doxycycline. *C. coli* is typically associated with pigs and pork.

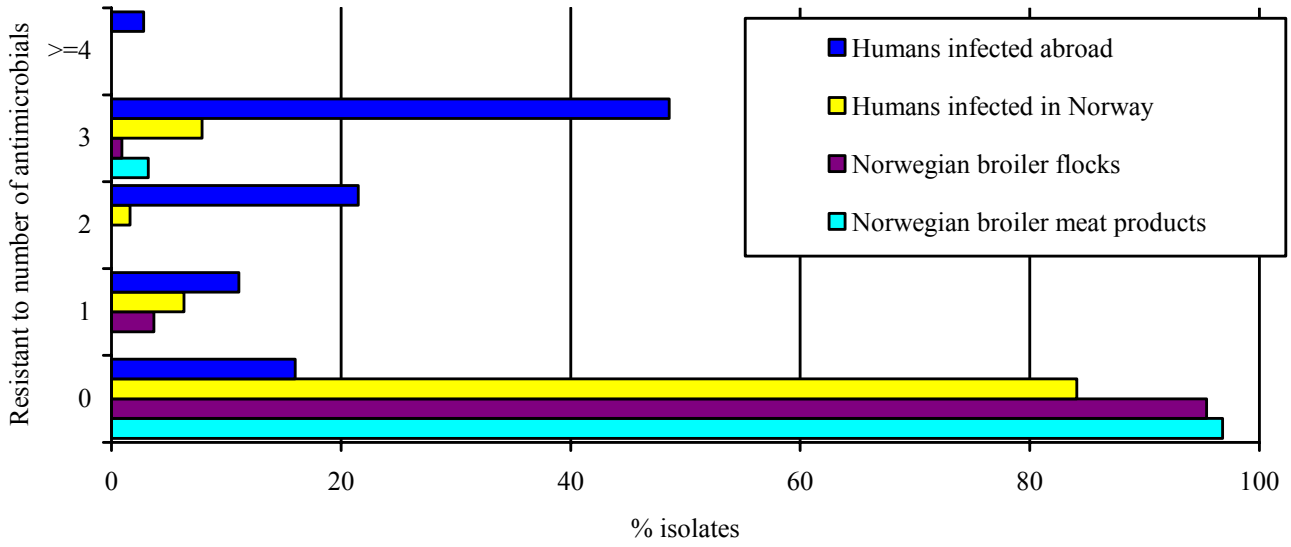

FIGURE 14. Antimicrobial resistance profiles for *Campylobacter jejuni* from Norwegian broiler flocks (n=108), Norwegian broiler meat products (n=31), humans infected in Norway (n=63) and humans infected abroad (n=144). Proportion of isolates susceptible to all antimicrobials included or resistant to one, two, three, or four or more antimicrobials. The isolates from humans were tested for susceptibility to doxycycline, erythromycin, gentamicin, ciprofloxacin and nalidixic acid, whereas the broiler and broiler meat isolates in addition were tested for susceptibility to ampicillin (and to oxytetracycline rather than doxycycline).

Yersinia enterocolitica from human clinical specimens

Most cases of *Yersinia enterocolitica* infection in Norway are domestically acquired. In 2003, only 19% of the 86 reported cases were classified as imported.

A total of 58 isolates of *Y. enterocolitica* O:3 were susceptibility tested. The results are presented in Tables 31 and 32.

TABLE 31. *Yersinia enterocolitica* serogroup 0:3 isolates from human clinical cases (n=58). Distribution (%) of antimicrobial susceptibility groups.

	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)		
Substance	S	R	S	I	R			
Tetracycline	≥ 20	≤ 16	100.0	0.0	0.0	24	-	≥ 36
Chloramphenicol	≥ 20	≤ 19	94.8	-	5.2	6	-	≥ 36
Ampicillin	≥ 32	≤ 12	0.0	0.0	100.0	6	-	11
TMS**	≥ 20	≤ 12	96.6	1.7	1.7	9	-	≥ 36
Ciprofloxacin	≥ 27	≤ 18	96.6	3.4	0.0	22	-	≥ 36
Nalidixic acid	≥ 17	≤ 16	96.6	-	3.4	6	-	32

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

TABLE 32. *Yersinia enterocolitica* serogroup 0:3 isolates (n=58) from human clinical cases. Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline																
Chloramph.	5.2															
Ampicillin	56.9	24.1	13.8		3.4	1.7										
TMS**				1.7							1.7					1.7
Ciprofloxacin																
Nalidixic acid	3.4														1.7	

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline			3.4	3.4	1.7	8.6	22.4	19.0	27.6	6.9	3.4	1.7			1.7
Chloramph.			1.7	6.9	8.6	13.8	12.0	8.6	17.2	3.4	8.6	5.2	5.2		3.4
Ampicillin															
TMS**		1.7	1.7			6.9	8.6	10.3	20.7	5.2	10.3	8.6	5.2	6.9	8.6
Ciprofloxacin	1.7			1.7		1.7	5.2	6.9	8.6		5.2	8.6	8.6	6.9	44.8
Nalidixic acid	3.4		6.9	3.4	17.2	13.8	21.0	15.5	1.7	3.4	8.6				

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

RESULTS AND COMMENTS

Compared to data from 2001 and 2002 and taking into account change of the breakpoints applied, the resistance prevalences are quite similar. None of the isolates tested were resistant to ciprofloxacin. However, two isolates were classified as resistant to nalidixic acid and one of these was also intermediately susceptible to ciprofloxacin. All isolates expressed reduced susceptibility to ampicillin, an intrinsic resistance trait in strains of serogroup O:3.

Shigella spp. from human clinical specimens

It is emphasized that almost all the reported *Shigella* infections in Norway were acquired abroad, mostly in Egypt, Tunisia, Pakistan and Turkey. In 2003, only 6% of the reported cases were classified as domestically acquired. Thus, the resistance prevalences reported here predominantly relate to isolates originating in other countries. The distribution of the *Shigella* species was as follows: *S. sonnei* 74 (57%), *S. flexneri* 43 (33%), *S. boydii* 7 (5%), and *S. dysenteriae* 5 (4%). The results for *S. sonnei* and *S. flexneri* are presented in Tables 33-36 and in the text.

TABLE 33. *Shigella sonnei* isolates from human clinical cases (n=74). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)
	S	R	S	I	R	
Tetracycline	≥ 20	≤ 16	41.9	0.0	58.1	6 - 28
Chloramphenicol	≥ 20	≤ 19	91.9	-	8.1	6 - 33
Ampicillin	≥ 32	≤ 12	0.0	78.4	21.6	6 - 29
TMS**	≥ 20	≤ 12	25.7	5.4	68.9	6 - ≥ 36
Ciprofloxacin	≥ 27	≤ 18	95.9	4.1	0.0	22 - ≥ 36
Nalidixic acid	≥ 17	≤ 16	81.1	-	18.9	6 - 34

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

TABLE 34. *Shigella sonnei* isolates from human clinical cases (n=74). Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	54.1	2.7	1.4												1.4	
Chloramph.	8.1															1.4
Ampicillin	21.6								1.4	1.4	1.4	1.4	1.4	4.1	28.4	14.9
TMS**	66.2			1.4		1.4		1.4				1.4	2.7			
Ciprofloxacin																
Nalidixic acid	14.9	2.7							1.4							

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline	5.4	2.7	1.4	5.4	16.2	6.8	2.7								
Chloramph.	2.7	2.7	17.6	9.5	23.0	12.2	8.1	4.1	6.8		2.7	1.4			
Ampicillin	9.5	2.7	5.4	2.7	2.7			1.4							
TMS**						1.4	1.4	1.4			4.1	4.1	6.8	2.7	4.1
Ciprofloxacin	1.4				2.7	2.7	8.1	1.4	4.1			1.4	5.4	1.4	71.6
Nalidixic acid			1.4	2.7	4.1	2.7	25.7	20.3	16.2	4.1	2.7		1.4		

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

TABLE 35. *Shigella flexneri* isolates from human clinical cases (n=43). Distribution (%) of antimicrobial susceptibility groups.

Substance	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)
	S	R	S	I	R	
Tetracycline	≥ 20	≤ 16	27.9	0.0	72.1	6 - 32
Chloramphenicol	≥ 20	≤ 19	34.9	-	65.1	6 - ≥ 36
Ampicillin	≥ 32	≤ 12	0.0	27.9	72.1	6 - 30
TMS**	≥ 20	≤ 12	30.2	0.0	69.8	6 - ≥ 36
Ciprofloxacin	≥ 27	≤ 18	93.0	4.7	2.3	6 - ≥ 36
Nalidixic acid	≥ 17	≤ 16	86.0	-	14.0	6 - ≥ 36

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

TABLE 36. *Shigella flexneri* isolates from human clinical cases (n=43). Distribution (%) of zone diameters (mm).*

Substance	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Tetracycline	60.5	7.0	2.3		2.3											
Chloramph.	41.9		9.3	9.3	2.3		2.3									
Ampicillin	72.1															
TMS**	69.8															
Ciprofloxacin	2.3															
Nalidixic acid	9.3	2.3				2.3										

Substance	22	23	24	25	26	27	28	29	30	31	32	33	34	35	≥ 36
Tetracycline					2.3	4.7	7.0	2.3	4.7	2.3	4.7				
Chloramph.											4.7		9.3	2.3	18.6
Ampicillin		2.3		2.3	4.7	4.7	4.7	4.7	4.7						
TMS**				2.3			2.3			4.7	2.3		2.3		16.3
Ciprofloxacin		2.3	2.3							2.3		4.7		4.7	81.4
Nalidixic acid						4.7	18.6	18.6	18.6	9.3	7.0	4.7	2.3		2.3

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

RESULTS AND COMMENTS

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

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

D. BACTERIA FROM HUMAN CLINICAL SPECIMENS

Escherichia coli in blood cultures

TABLE 37. *Escherichia coli* blood culture isolates (n=966). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Ampicillin	≤ 1	≥ 32	3.5	66.4	30.1	0.25 - ≥ 256	4	≥ 256
Amoxi/Clav**	≤ 0.5	≥ 32	0.3	97.6	2.1	0.5 - ≥ 256	4	8
Cefpirome	≤ 1	≥ 32	99.8	0.2	0.0	0.016 - 2	0.064	0.125
Ceftazidime	≤ 1	≥ 32	99.2	0.7	0.1	0.016 - ≥ 256	0.125	0.25
Cefuroxime	≤ 1	≥ 32	4.2	94.4	1.3	0.25 - ≥ 256	4	4
Ciprofloxacin	≤ 0.125	≥ 4	94.9	2.8	2.3	0.002 - ≥ 32	0.002	0.032
Gentamicin	≤ 2	≥ 8	99.3	0.1	0.6	0.032 - ≥ 256	0.25	0.5
Meropenem	≤ 0.5	≥ 4	100.0	0.0	0.0	0.002 - 0.125	0.016	0.016
Pip/Tazo***	≤ 8	≥ 32	98.9	0.3	0.8	0.032 - ≥ 256	2	4
TMS****	≤ 2	≥ 16	82.2	0.2	17.5	0.008 - ≥ 32	0.064	≥ 32
ESBL			99.7	-	0.3			

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

TABLE 38. *Escherichia coli* blood culture isolates (n=966). Distribution (%) of MICs (mg/L).*

	≤0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Ampicillin						0.2	0.2	3.1	18.4	38.6	8.2	1.0	0.4	0.5	0.7	28.4
Amoxi/Clav**							0.2	3.5	13.1	54.3	20.6	5.0	1.0	0.1		0.9
Cefpirome		5.8	43.5	40.7	7.0	1.9	0.7	0.2	0.2							
Ceftazidime		0.2	2.8	12.6	50.1	29.1	3.4	0.9	0.4	0.2	0.1					0.1
Cefuroxime						0.2	0.4	3.6	32.8	53.0	6.6	2.0	0.9		0.1	0.3
Ciprofloxacin	40.2	46.7	5.2	1.1	1.7	1.9	0.7	0.2		0.1	0.2	0.3	1.7			
Gentamicin			0.6	1.2	16.2	54.9	19.1	5.8	1.3	0.1		0.1	0.2	0.1		0.2
Meropenem	11.8	78.5	8.8	0.5	0.3											
Pip/Tazo***			0.1	0.3	0.3	0.5	3.8	22.3	60.2	9.7	1.6	0.3	0.1		0.1	0.6
TMS****	0.1	1.3	14.4	41.0	15.5	6.0	2.5	0.9	0.4	0.2			17.5			

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

RESULTS AND COMMENTS

The prevalence of resistance to older antimicrobials such as trimethoprim/sulfamethoxazole and gentamicin was stable. There was a minor increase in ampicillin resistance from 26.5% in 2002 to 30.1% in 2003 due to an increase in the proportion of high-level resistance to this agent. As seen in Figure 15, there was also a slight shift to the right of the wild-type MIC distribution leading to a higher prevalence of intermediate susceptibility.

The isolates were generally susceptible to broad-spectrum antimicrobials including ceftazidime, cefpirome, meropenem, ciprofloxacin and piperacillin/tazobactam. All isolates were specifically examined for production of extended spectrum β-lactamases by a disk approximation test, and isolates with a positive ESBL test and/or reduced susceptibility to ceftazidime (MIC ≥ 1 mg/L) and/or cefpirome (MIC ≥ 1 mg/L) were further characterized by combination Etests and/or molecular examinations. A total of three ESBL producing isolates were detected giving a 0.3% prevalence of ESBL producers among all isolates. Two isolates originated from the same local hospital two weeks apart, while the third isolate was detected at a university clinic in a different part of the country. The findings are in accordance with the results from 2001 (0%) and 2002 (0.3%). The three isolates were detected by elevated MICs of ceftazidime (n=2), elevated MIC of cefpirome (n=1) or a positive disk approximation test (n=3). Conversely, there were no false positive ESBL disk approximation tests, whereas a total of 17 isolates with elevated MICs of ceftazidime and/or cefpirome were ESBL negative. The optimal screening procedure for ESBL cannot be deduced from these data, especially since cefotaxime and cefpodoxime were not included in the

initial screening step. This may be of great significance as preliminary studies suggest that cefotaximases of the CTX-M group are the most prevalent ESBL phenotypes in Norway. The NORM 2004 protocol will evaluate a more complete range of cephalosporin Etests and disks in order to resolve this issue. The occurrence of ESBL producing strains should be closely monitored in light of the increasing usage of cephalosporins in Norwegian hospitals.

The prevalence of intermediate susceptibility and resistance to quinolones is still low but may be increasing. As seen in Figure 16, the overall prevalence of ciprofloxacin non-susceptibility is now 5.1%, which is approximately 40% above the prevalence seen in 2000. This parallels the increasing use of ciprofloxacin from 0.29 to 0.42 DDD/1,000 inhabitants/day in the same period (Figure 16). The present strict indications for ciprofloxacin use should be maintained if this agent is to be sustained as a broad-spectrum alternative for critically ill patients.

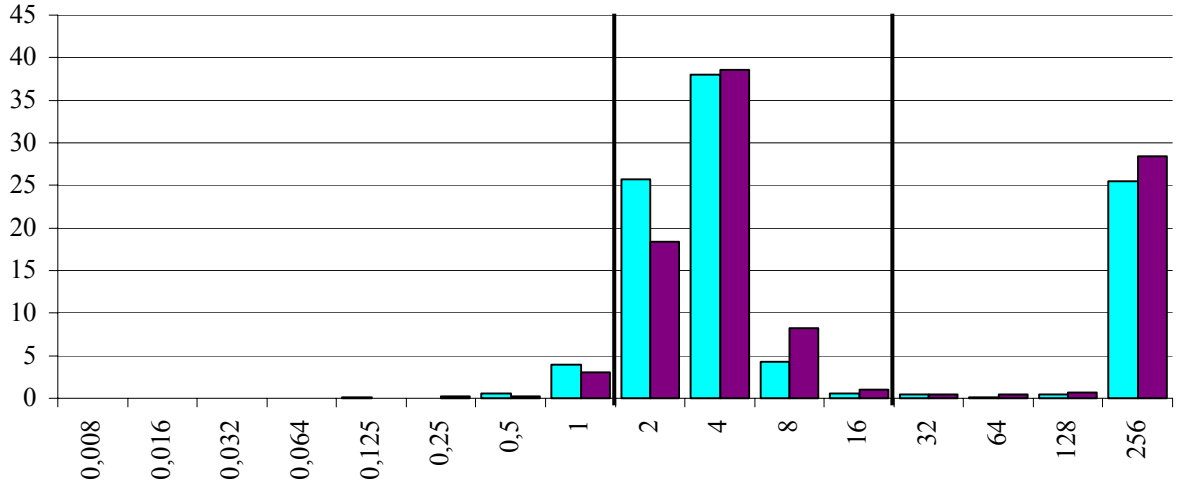

FIGURE 15. Distribution (%) of minimum inhibitory concentrations (MICs) of ampicillin for *E. coli* in 2002 (turquoise) and 2003 (magenta). The breakpoints of the Norwegian Reference Group for Antibiotic Susceptibility Testing (AFA) are indicated by vertical black bars.

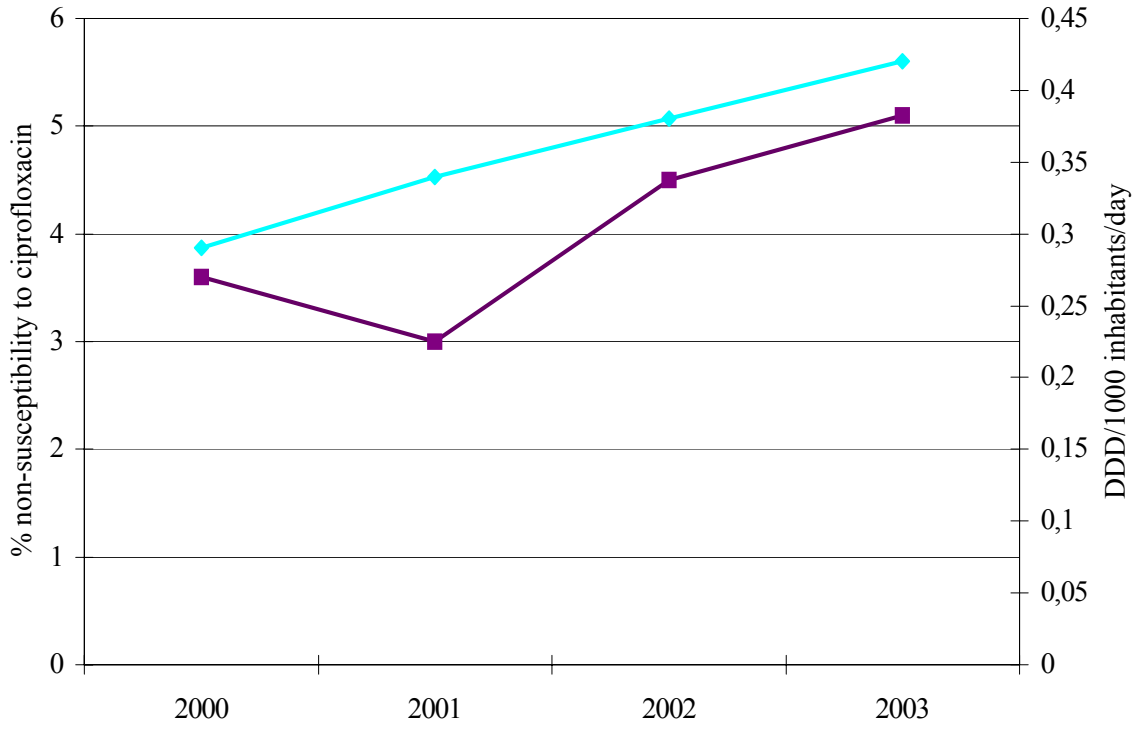

FIGURE 16. Prevalence of ciprofloxacin non-susceptibility in *E. coli* (magenta) and usage of ciprofloxacin (turquoise) 2000 – 2003.

Klebsiella spp. in blood cultures

TABLE 39. *Klebsiella* spp. blood culture isolates (n=299). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Ampicillin	≤ 1	≥ 32	2.0	38.9	59.1	0.5 - ≥ 256	32	≥ 256
Amoxi/Clav**	≤ 0.5	≥ 32	2.0	97.0	1.0	0.125 - ≥ 256	2	4
Cefpirome	≤ 1	≥ 32	100.0	0.0	0.0	0.016 - 1	0.064	0.125
Ceftazidime	≤ 1	≥ 32	98.3	1.3	0.3	0.016 - 128	0.125	0.5
Cefuroxime	≤ 1	≥ 32	19.4	77.9	2.7	0.25 - 128	2	8
Ciprofloxacin	≤ 0.125	≥ 4	91.6	8.0	0.3	0.004 - 8	0.032	0.125
Gentamicin	≤ 2	≥ 8	100.0	0.0	0.0	0.016 - 2	0.25	0.5
Meropenem	≤ 0.5	≥ 4	100.0	0.0	0.0	0.004 - 0.064	0.032	0.032
Pip/Tazo***	≤ 8	≥ 32	95.7	3.0	1.3	0.125 - ≥ 256	2	4
TMS****	≤ 2	≥ 16	96.0	1.0	3.0	0.016 - ≥ 32	0.125	0.5
ESBL			99.7	-	0.3			

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

TABLE 40. *Klebsiella* spp. blood culture isolates (n=299). Distribution (%) of MICs (mg/L).*

	≤ 0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Ampicillin								0.3	1.7	3.0	5.4	13.1	17.4	24.8	10.4	4.7	19.1
Amoxi/Clav**						0.3		1.7	13.7	59.5	17.4	4.3	2.0	0.3		0.3	0.3
Cefpirome			5.0	35.2	42.6	10.4	2.7	2.7	1.3								
Ceftazidime			1.0	6.4	25.2	34.9	20.5	6.7	3.7	1.3						0.3	
Cefuroxime							1.7	3.7	14.0	52.2	17.1	4.3	4.3	1.7	0.7	0.3	
Ciprofloxacin	1.0	8.4	22.4	40.1	16.1	3.7	5.4	2.0	0.7			0.3					
Gentamicin			0.3	0.3	1.0	14.4	56.5	18.7	7.7	1.0							
Meropenem	0.3	1.7	35.5	59.2	3.0												
Pip/Tazo***						0.3	1.3	5.0	11.0	48.2	24.7	5.0	3.0	0.3	0.3		0.6
TMS****			0.3	3.4	23.6	42.1	17.8	6.1	1.3	1.3	0.3	0.6	0.3	2.7			

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

RESULTS AND COMMENTS

Klebsiella spp. blood culture isolates included 215 *K. pneumoniae*, 44 *K. oxytoca* and 40 isolates not identified to the species level (Figure 17). There were no significant differences in the pattern of antimicrobial resistance among the different species and they were therefore analysed together.

The prevalence of resistance to β–lactams did not change from earlier years. The majority of isolates were phenotypically resistant to ampicillin as would be expected due to the chromosomally encoded SHV and K1 β–lactamases of *K. pneumoniae* and *K. oxytoca*, respectively. In the diagnostic laboratory, *Klebsiella* should be reported as ampicillin resistant without testing. As seen in Figure 18, wild-type *Klebsiella* isolates were intermediately susceptible to cefuroxim and susceptible to 3rd (ceftazidime) and 4th (cefpriome) generation cephalosporins and carbapenems. As for *E. coli*, blood culture isolates of *Klebsiella* spp. isolates were examined specifically for production of extended spectrum β-lactamases by a disk approximation test, and isolates with a positive ESBL test and/or reduced susceptibility to ceftazidime (MIC ≥ 1 mg/L) and/or cefpirome (MIC ≥ 1 mg/L) were further characterized by combination Etests and/or molecular examinations. One ESBL producing *K. pneumoniae* isolate was detected giving a prevalence of 0.3%. The issue of ESBL detection will be further evaluated in NORM 2004.

Ciprofloxacin resistance was detected in less than 1% of isolates, but the increasing prevalence of intermediate susceptibility is a cause for great concern. The uniform susceptibility to gentamicin indicates that aminoglycosides may be a therapeutic alternative in *Klebsiella* infections in certain clinical situations.

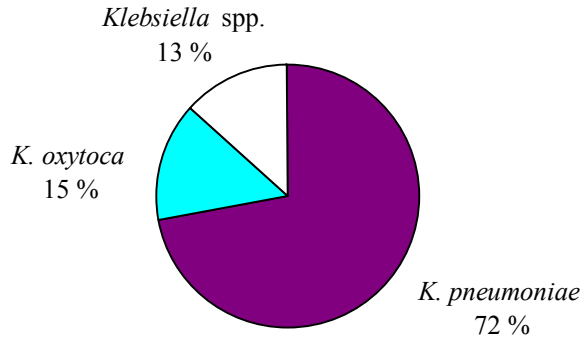

FIGURE 17. Distribution (%)of *Klebsiella* species from blood cultures in NORM 2003.

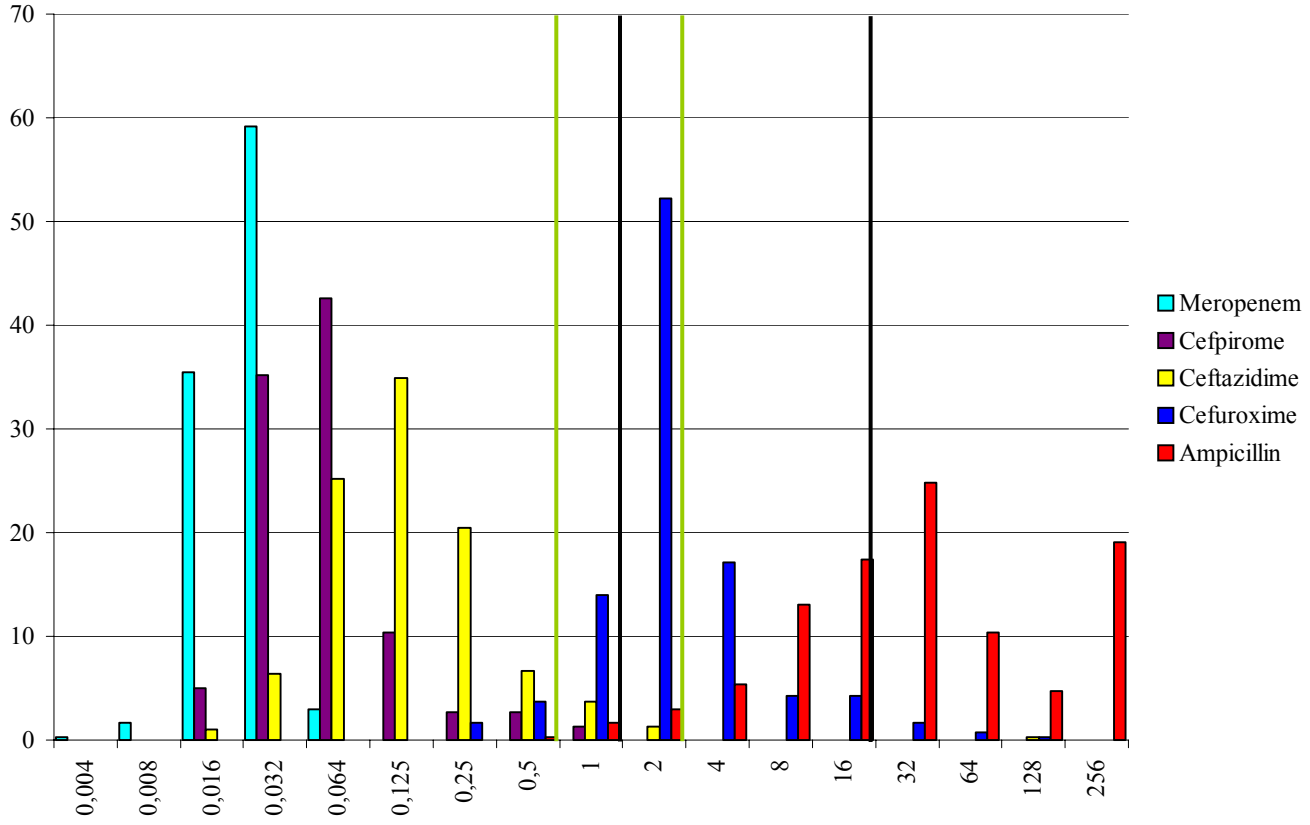

FIGURE 18. Distribution (%) of minimum inhibitory concentrations (mg/L) of different β-lactam antibiotics in *Klebsiella* spp. isolates from blood cultures. Vertical bars indicate breakpoints for susceptibility (left) and resistance (right) for meropenem (green bars) and cefpirome, ceftazidime, cefuroxime and ampicillin (black bars).

Enterococcus spp. in blood cultures

TABLE 41. *Enterococcus* spp. blood culture isolates (n=252). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints mg/L		Proportion of isolates (%)*			MIC range	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Ampicillin	≤ 2	≥ 16	87.7	2.4	9.5	0.016 - 256	0.5	8
Gentamicin	≤ 512	≥ 1024	88.9	-	11.1	0.016 - ≥ 1024	4	1024
Penicillin G	≤ 4	≥ 16	85.6	3.6	10.8	0.008 - ≥ 256	2	32
Streptomycin	≤ 512	≥ 1024	84.5	-	15.5	0.064 - ≥ 1024	32	≥ 1024
Vancomycin Screen			99.6	-	0.4			
β-lactamase			100.0	-	0.0			

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

TABLE 42. *Enterococcus* spp. blood culture isolates (n=252). Distribution (%) of MICs (mg/L).*

	≤ 0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	≥ 1024
Ampicillin	2.0	2.0	3.1	4.3	25.7	38.7	12.3		0.8	1.6	1.6	3.2	2.4	0.8	1.6		
Gentamicin	0.4		0.4	0.8	2.4	7.5	10.7	19.4	19.0	15.5	6.3	3.6	0.8	1.2	0.4	0.4	11.1
Penicillin G	1.2	2.0	0.8	0.8	2.4	10.4	26.7	31.9	9.6	3.6		4.0	1.2		5.6		
Streptomycin			1.6	0.4		0.8	1.6	2.4	6.3	9.5	20.6	21.8	11.1	5.2	2.0	1.2	15.5

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

TABLE 43. *Enterococcus faecalis* blood culture isolates (n=183). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints mg/L		Proportion of isolates (%)*			MIC range	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Ampicillin	≤ 2	≥ 16	100.0	0.0	0.0	0.016 - 1	0.5	1
Gentamicin	≤ 512	≥ 1024	85.7	-	14.2	0.064 - ≥ 1024	4	1024
Penicillin G	≤ 4	≥ 16	95.1	4.4	0.5	0.008 - 32	2	4
Streptomycin	≤ 512	≥ 1024	85.8	-	14.2	0.064 - ≥ 1024	32	≥ 1024
Vancomycin Screen			100.0	-	0.0			
β-lactamase			100.0	-	0.0			

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

TABLE 44. *Enterococcus faecalis* blood culture isolates (n=183). Distribution (%) of MICs (mg/L).*

	≤ 0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	≥ 1024
Ampicillin	1.1	0.5	2.2	4.4	31.1	45.9	14.8										
Gentamicin			0.5	0.5	1.1	6.6	8.7	16.9	17.5	18.0	7.1	4.9	1.1	1.6	0.5	0.5	14.2
Penicillin G	0.5	0.5		0.5	1.1	11.0	31.8	36.6	12.6	4.4		0.5					
Streptomycin			1.1			1.1		2.7	5.5	7.7	20.8	25.1	13.7	5.5	1.6	1.1	14.2

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

TABLE 45. *Enterococcus faecium* blood culture isolates (n=40). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints mg/L		Proportion of isolates (%)*			MIC range	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Ampicillin	≤ 2	≥ 16	37.5	12.5	50.0	0.016 - ≥ 256	8	64
Gentamicin	≤ 512	≥ 1024	97.5	-	2.5	0.016 - ≥ 1024	2	8
Penicillin G	≤ 1	≥ 16	42.5	2.5	55.0	0.016 - ≥ 256	32	≥ 256
Streptomycin	≤ 512	≥ 1024	77.5	-	22.5	0.064 - ≥ 1024	16	≥ 1024
Vancomycin Screen			97.5	-	2.5			
β-lactamase			100.0	-	0.0			

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

TABLE 46. *Enterococcus faecium* blood culture isolates (n=40). Distribution (%) of MICs (mg/L).*

	≤ 0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	512	≥ 1024
Ampicillin	7.5	2.5	7.5	2.5	7.5	7.5	2.5		5.0	7.5	7.5	20.0	12.5	5.0	5.0		
Gentamicin	2.5			2.5	7.5	10.0	20.0	20.0	17.5	12.5	5.0						2.5
Penicillin G	5.0	5.0	5.0		7.5	2.5	5.0	10.0	2.5	2.5		20.0	7.5		27.5		
Streptomycin			5.0	2.5			7.5		15.0	12.5	15.0	7.5	5.0	2.5	2.5	2.5	22.5

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

RESULTS AND COMMENTS

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

The susceptibility to β-lactams was again shown to be much lower in *E. faecium* than in *E. faecalis*. The latter species was uniformly susceptible to ampicillin, and 95% of the isolates were also susceptible to penicillin G. In contrast, only 37.5% of *E. faecium* isolates were ampicillin susceptible. The prevalence of intermediate susceptibility and resistance was comparable to 2002 when new breakpoints were introduced.

High-level resistance to aminoglycosides is a threat to traditional combination regimens used for treatment of serious enterococcal infections. Figure 19 shows the alarming trend in enterococcal gentamicin resistance caused by a 14.2% resistance rate to this agent in *E. faecalis*.

Vancomycin resistance was reported in a single VanB *E. faecium* isolate. This isolate was also resistant to ampicillin (MIC=64 mg/L), but high-level resistance to aminoglycosides was not detected. Surveillance protocols for enterococci will from 2004 include more modern agents such as quinupristin/dalfopristin and linezolid.

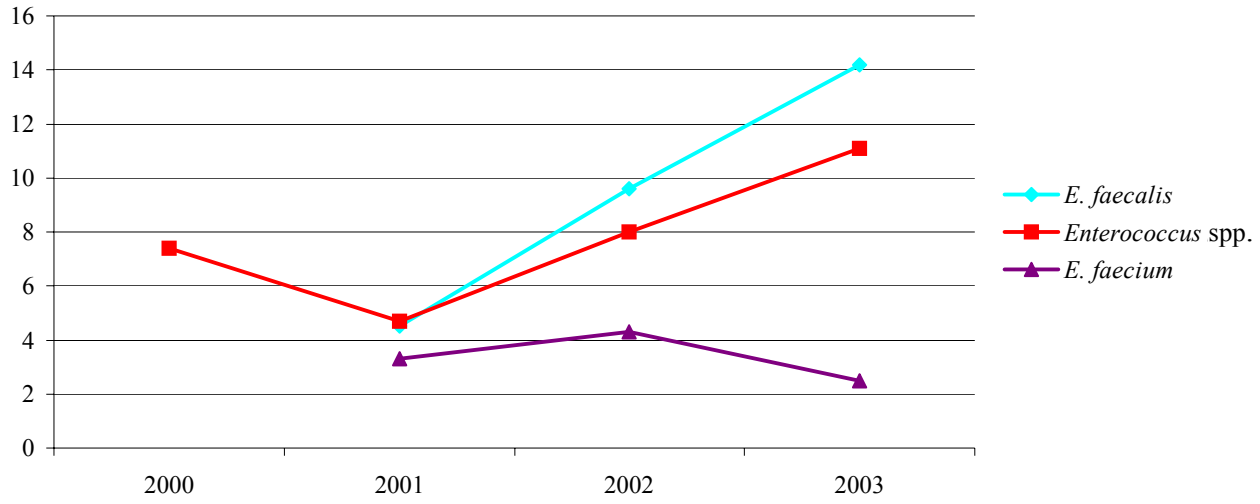

FIGURE 19. Prevalence of high-level resistance to gentamicin in blood culture isolates of *E. faecalis*, *E. faecium* and all enterococci combined 2000-2003.

Aspects of bacterial resistance to disinfectants

Disinfectants are widely used in human and veterinary medicine and in the food processing industry. In addition, a recent trend is the inclusion of antimicrobial agents (including disinfectants) into a multitude of traditional consumer products[1]. Several researchers have expressed their concern that the widespread use of disinfectants may contribute to the development and selection of bacteria resistant to various antimicrobial agents, including antibiotics. This has led to increased focus on potential ecological consequences and elucidation of resistance mechanisms.

Important aspects of the development of resistance and cross-resistance include types, frequency and concentrations of antimicrobials used. Although concentration dependent, disinfectants (multiple targets) and antibiotics (specific targets) often differ in their modes of action. Thus, resistance mechanisms towards antibiotics and disinfectants also differ. Bacterial disinfectant resistance mechanisms include intrinsic mechanisms where resistance and cross-resistance occur as a result of natural properties of the organism, e.g. impermeability barriers like cell wall properties and slime production. Disinfectant resistance can also occur as a result of efflux pumps, mutations in target sites, enzymatic inactivation or a combined effect of these mechanisms[2, 3].

Resistance to disinfectants based on quaternary ammonium compounds (QAC), one of the most widespread used antimicrobial compounds, have been extensively studied. In staphylococci, efflux mechanisms contributing to low-level QAC resistance is widespread in environments where QAC-based disinfectants are used on a routine basis. To date six different staphyloccal QAC resistance efflux genes, *qacA*, *qacB*, *smr* (*qacC*), *qacG*, *qacH*, *qacJ* have been characterized[4-8]. The general plasmid borne location of these genes contributes to the dissemination of these resistance genes among various staphylococcal species. Genetic linkage between QAC resistance genes and antibiotic resistance genes (most frequently *blaZ*; beta-lactamase resistance) is common[9]. A study of clinical staphylococci showed that QAC resistant strains are more often resistant to antibiotics than QAC sensitive isolates[10]. This indicates that the presence of *qac* genes in staphylococci results in the selection of antibiotic resistant strains, probably by co-selection. Several efflux-based resistance mechanisms (e.g. *acrAB*, *mex*) have also been characterized in gram-negative bacteria. In combination with other resistance mechanisms, efflux is regarded an important contributor to high-level resistance, not only to disinfectants, but also to a range of antimicrobial agents including antibiotics. Adaptation of bacteria to low-level concentrations of antimicrobials and other stress factors may induce resistance mechanisms to various antimicrobial compounds[11].

The widespread use of triclosan has led to concerns that it could exert a selective pressure for the development of antibiotic resistant bacteria. In laboratory experiments, low-level concentrations of triclosan have the same mechanism of action as isoniazid, a drug used in the treatment of tuberculosis. Laboratory mutants of *Mycobacterium smegmatis* and *Mycobacterium tuberculosis* obtained by triclosan selection conferred resistance to both triclosan and isoniazid. However, there is (still) no evidence that the use of triclosan has resulted in clinical development of isoniazid-resistant mycobacteria or other antibiotic resistant gram-positive or gram-negative bacteria[1, 3, 12].

The significance of disinfectant use and measure of resistance in laboratory model systems and its relevance to clinical practice and the food processing industry is debated. In nature, bacteria are present in both planctonic and biofilm communities, which affects both the biocidal effects and the bacterial reponse. Consequently, translations of laboratory findings to clinical and environmental situations are not straightforward. A further understanding of factors affecting biocidal effects and resistance/cross-resistance development is needed for future safe and effective use of antimicrobial agents including disinfectants.

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Even Heir

Streptococcus pneumoniae in blood cultures

TABLE 47. *Streptococcus pneumoniae* blood culture isolates (n=514). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Cefotaxime	≤ 0.5	≥ 4	100.0	0.0	0.0	0.002 - 0.5	0.016	0.016
Cefuroxime	≤ 0.5	≥ 4	99.7	0.2	0.0	0.016 - 1	0.016	0.016
Chloramph.	≤ 4	≥ 8	99.4	0.0	0.6	0.016 - 16	2	2
Ciprofloxacin	≤ 0.125	≥ 4	0.2	97.9	1.9	0.064 - 16	1	2
Clindamycin	≤ 0.25	≥ 4	99.0	0.2	0.8	0.016 - ≥ 256	0.125	0.125
Doxycycline	≤ 1	≥ 4	98.6	0.2	1.2	0.064 - 16	0.125	0.25
Erythromycin	≤ 0.5	≥ 1	94.0	0.0	6.0	0.016 - ≥ 256	0.064	0.125
Oxacillin screen	≥ 20 mm	≤ 19 mm	96.6	-	2.9			
Pen G**	≤ 0.064	≥ 2	99.5	0.6	0.0	0.002 - 0.5	0.016	0.016
TMS***	≤ 0.5	≥ 4	96.7	1.9	1.4	0.002 - ≥ 32	0.25	0.25
Vancomycin	≤ 2	≥ 8	100.0	0.0	0.0	0.125 - 2	0.5	1

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

TABLE 48. *Streptococcus pneumoniae* blood culture isolates (n=514). Distribution (%) of MICs (mg/L).*

	≤0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	≥ 128
Cefotaxime	3.1	30.0	60.5	3.9	1.4	1.0		0.2								
Cefuroxime			93.3	5.2	1.2				0.2							
Chloramph.			0.2	0.2	0.8	1.8	3.3	4.5	26.1	61.4	1.2	0.4	0.2			
Ciprofloxacin					0.2		2.7	23.4	53.0	18.7	1.6		0.4			
Clindamycin			6.6	4.5	29.8	50.8	7.4		0.2							0.8
Doxycycline					10.7	43.0	37.2	6.2	1.6	0.2	0.2	0.6	0.4			
Erythromycin			6.2	7.4	37.2	42.8	0.4		0.4	0.4	1.4	1.6	1.2	0.4		0.8
Pen G**	1.2	25.9	64.4	7.0	1.0	0.4		0.2								
TMS***	0.4	0.2	0.8	1.0	5.4	28.8	57.4	2.7	1.9			0.6	0.6	0.2		
Vancomycin						1.6	10.7	50.2	31.5	6.0						

	≤ 19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	≥ 34
Oxacillin disk	2.9	0.8		1.0	4.5	7.0	10.9	12.7	11.7	14.8	7.6	11.5	3.7	6.0	2.5	1.9

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

RESULTS AND COMMENTS

As in previous years, *Streptococcus pneumoniae* from blood cultures were generally susceptible to all relevant antimicrobials. Two isolates had MICs for penicillin G of 0.125 mg/L. A single isolate had an MIC of 0.5 mg/L, and this isolate also had reduced susceptibility to cefuroxime (MIC=1 mg/L) and an MIC to cefotaxime above the wild-type distribution for this agents (MIC=0.5 mg/L). The oxacillin screening test for non-susceptibility to penicillin identified all the non-susceptible isolates, and the isolate with MIC of 0.5 mg/L had no zone of inhibition around the oxacillin disk. The screening procedure consequently had 100% sensitivity in our low-prevalence situation. Conversely, 14 isolates with penicillin G MICs in the susceptible range were positive in the oxacillin screening test giving a specificity of 97.3%.

The prevalence of non-susceptibility to erythromycin increased to 6.0% due to increased low-level resistance. A total of 27 isolates with MIC values of 1–32 mg/L were detected indicating further expansion of pneumococcal clones harbouring the *mef*-encoded efflux mechanism for macrolides. As seen in Figure 20, the prevalence of combined high-level resistance to macrolides (erythromycin) and lincosamides (clindamycin) remained stable below 1%. The four isolates with high-level resistance presumably expressed *erm*-encoded methylation of ribosomal RNA. One of these isolates was the strain with reduced susceptibility to both penicillin G and cefuroxime. The change of breakpoints for erythromycin and clindamycin from 2001 to 2002 should not bias these results significantly as very few isolates have MICs close to the breakpoints.

It should be noted that the results demonstrated chlorampenicol, doxycycline and trimethoprim/sulfamethoxazole as viable second-line alternatives when first-line antimicrobials cannot be used.

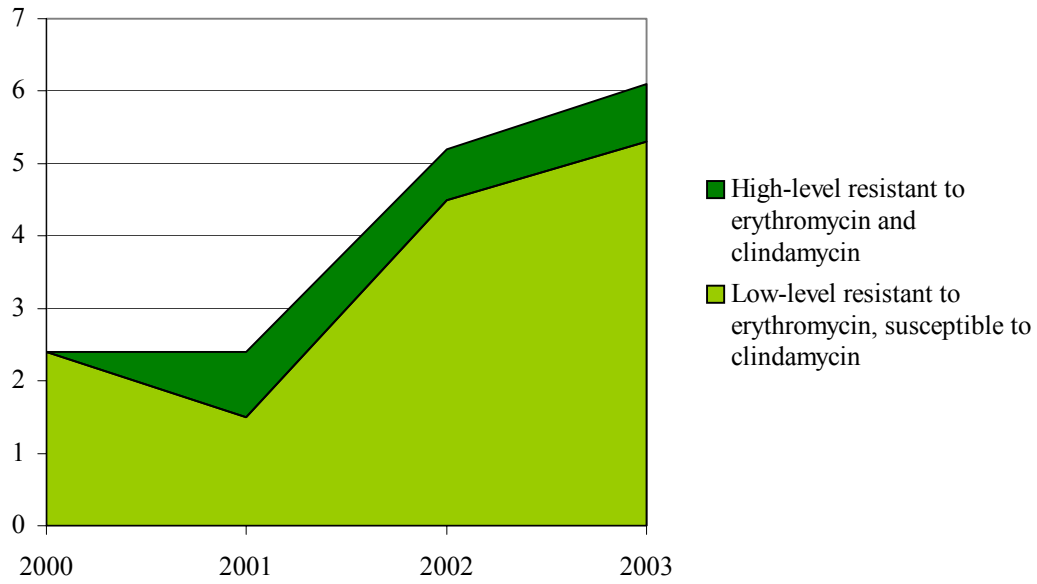

FIGURE 20. Prevalence (%) *of Streptococcus pneumoniae* with high-level resistance to erythromycin and clindamycin or low-level resistance to erythromycin and susceptibility to clindamycin 2000-2003.

Staphylococcus aureus in blood cultures

TABLE 49. *Staphylococcus aureus* blood culture isolates (n=637). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Cefuroxime	≤ 2	≥ 8	99.4	0.6	0.0	0.25 - 4	1	1
Clindamycin	≤ 1	≥ 4	98.7	0.0	1.3	0.016 - ≥ 256	0.064	0.125
Doxycycline	≤ 1	≥ 4	95.6	1.7	2.7	0.032 - 32	0.125	0.25
Erythromycin	≤ 1	≥ 4	96.5	0.2	3.3	0.016 - ≥ 256	0.25	0.25
Fusidic acid	≤ 0.5	≥ 1	93.9	-	6.1	0.016 - ≥ 256	0.064	0.25
Gentamicin	≤ 2	≥ 8	98.9	0.5	0.6	0.016 - 32	0.25	0.5
Oxacillin	≤ 2	≥ 4	99.4	0.0	0.6	0.032 - ≥ 256	0.5	1
Oxacillin screen			99.4	-	0.6			
Penicillin G**	≤ 0.064	≥ 0.25	26.1	3.8	70.2	0.016 - ≥ 256	0.5	4
β-lactamase			25.3	-	74.7			
Vancomycin screen			100.0	-	0.0			

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

TABLE 50. *Staphylococcus aureus* blood culture isolates (n=637). Distribution (%) of MICs (mg/L).*

	≤0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Cefuroxime						1.2	39.8	58.4		0.6						
Clindamycin		0.2	3.3	57.3	35.2	1.7	0.3	0.8								1.3
Doxycycline			0.9	13.0	54.9	24.2	1.4	1.1	1.7	1.7	0.5	0.3	0.2			
Erythromycin		0.3	2.2	4.7	40.3	45.4	3.3	0.3	0.2	0.2	0.3	0.2	0.3	0.2		2.2
Fusidic acid		1.6	12.9	52.0	23.4	2.5	1.6	0.6	0.5	2.2	1.9	0.6	0.2			0.2
Gentamicin		0.2	0.6	0.8	22.0	47.3	22.3	5.2	0.6	0.5	0.2	0.3	0.2			
Oxacillin			0.3	1.1	13.2	32.3	41.3	10.2	0.8			0.2	0.3			0.2
Penicillin G**		6.3	16.3	3.5	3.6	4.9	15.8	24.3	13.0	6.4	2.8	0.9	1.7	0.2		0.2

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

RESULTS AND COMMENTS

A total of four *Staphylococcus aureus* isolates (0.63%) were verified as methicillin resistant *S. aureus* (MRSA) by *mecA* and *nuc* PCRs. This is in accordance with the reults from NORM 2000-2002 and the Norwegian Surveillance System for Communicable Diseases (MSIS) where 2-6 cases of septicaemiae have been reported each year since 1997. The four MRSA strains were isolated at four different hospitals, and there were no apparent epidemiological links between them. All isolates were detected both by use of the oxacillin screening agar and the oxacillin Etest. Conversely, no isolates were identified as MRSA by these tests without being *mec*/*nuc* PCR positive. Thus, both tests were able to reliably identify MRSA in a low prevalence population.

The discrepancy between the increasing total number of MRSA reported to MSIS (see below) and the stability of MRSA prevalence in NORM blood culture isolates and yearly numbers of septicaemiae cases in MSIS is difficult to explain. The proportion of invasive isolates is obviously small, and it may therefore take some time before an upward trend is noted. Alternatively, the MRSA strains circulating in Norway may have virulence factors needed to establish soft tissue infections without causing systemic disease. Both the epidemiological and the microbiological surveillance system for MRSA will be further improved in 2004 and hopefully give answers to some of these questions.

Figure 21 shows the prevalence of non-susceptibility to various antimicrobials other than β-lactams. The most striking finding was the decline in the prevalence of resistance to fusidic acid. Norway has in recent years experienced an epidemic of impetigo bullosa caused by fusidic resistant *S. aureus*. Among *S. aureus* from wound specimens, the epidemic seems to have reached its peak with 20.8% resistance in 2001 and 23.0% in 2003. As seen in Figure 21, the prevalence of resistance to fusidic acid has been much lower in systemic isolates and is now declining. For the other antimicrobials in Figure 21 there were slowly increasing trends except for vancomycin.

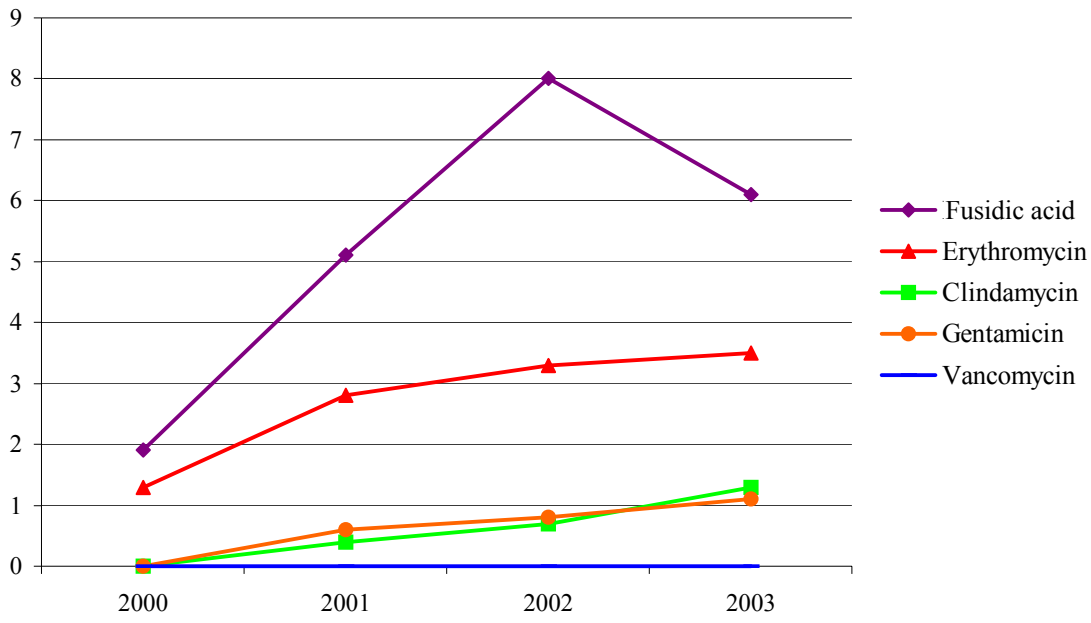

FIGURE 21. Prevalence (%) of non-suscptibility to antimicrobials other than β-lactams in *S. aureus* from blood cultures.

MRSA infections in Norway

Infections with methicillin resistant *Staphylococcus aureus* (MRSA) have been notifiable in Norway since 1995, but colonisation without infection is not notifiable. Consistent discrimination between the two can be difficult. In 2003 there was once again a substantial increase in the number of reported cases with 216 cases compared to 143 and 121 the two previous years (MSIS report 2004; 32:25, Figure 22). Of the cases, 116 were men (54%). Median age in 2003 was 42 years (range 0–97 years) and 40 years for all nine years reported. Less than one third (30%) of the patients falling ill in 2003 was hospitalised at the time of diagnosis.

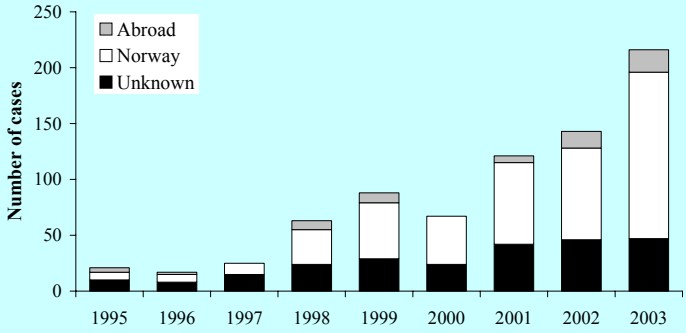

FIGURE 22. Reported cases of MRSA infection 1995–2003 and whether the infection was contracted abroad or not.

MRSA was found in blood cultures in seven patients in 2003 and only 29 for all nine years reported. The clinical picture shows a majority of wound infections or abscesses (Table 51).

TABLE 51. Clinical picture of reported cases of MRSA infection in Norway 1995–2003.

Clinical picture	1995	1996	1997	1998	1999	2000	2001	2002	2003	Total
Septicaemia	1		3	3	4	2	6	4	5	28
Septicaemia and meningitis				1						1
Meningitis				1						1
Osteomyelitis	2	1		2			2	2	2	11
RTI*, incl. otitis media	1	1	1	14	8	5	8	13	7	58
Urinary tract infection		1		4	3	3	2	9	12	34
Wound infection, abscess	17	14	19	36	71	54	97	115	189	612
Other, unknown			2	2	2	3	6		1	16
Total	21	17	25	63	88	67	121	143	216	761

* RTI = Respiratory tract infection

The number of cases of MRSA infection has increased by 51% from the previous year. The overwhelming majority consists of wound infections. The number of serious infections is still very low with five clinical septicaemias and seven positive blood cultures. The numbers are too small to ascertain whether there has been a true increase in serious infections.
How large the true increase in the total number of infections is, has to be interpreted with caution. The increase is mainly seen in non-hospitalised patients with minor infections and who have contracted the disease in Norway. This may indicate increased testing of patients outside hospitals.
The national surveillance of MRSA in Norway is not satisfactory. Only MRSA infections are notifiable and not colonisations. Consequently only a part of the picture is seen and the possibility to discover outbreaks is made difficult. Furthermore we are in a process to establish a national reference laboratory where all isolates are to be sent, analysed and genetically compared.

Bjørn G. Iversen

Streptococcus pneumoniae in respiratory tract specimens

TABLE 52. *Streptococcus pneumoniae* in respiratory tract specimens (n=752). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Clindamycin	≤ 0.25	≥ 4	98.4	0.7	0.9	0.016 - ≥ 256	0.125	0.125
Doxycycline	≤ 1	≥ 4	92.3	1.5	6.3	0.016 - 64	0.125	0.5
Erythromycin	≤ 0.5	≥ 1	94.3	0.0	5.7	0.008 - ≥ 256	0.064	0.125
Oxacillin screen	≥ 20 mm	≤ 19 mm	94.9	-	5.1			
Pen G**	≤ 0.064	≥ 2	97.2	2.8	0.0	0.002 - 1	0.016	0.032
TMS***	≤ 0.5	≥ 4	95.6	1.9	2.5	0.032 - ≥ 32	0.25	0.25

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

TABLE 53. *Streptococcus pneumoniae* in respiratory tract specimens (n=752). 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
Clindamycin			0.5	5.1	36.8	50.9	5.1	0.3	0.4		0.1					0.8
Doxycycline			0.1	0.4	4.9	45.9	31.7	8.3	0.9	1.5	1.1	2.0	2.1	0.7	0.4	
Erythromycin		0.1	0.5	6.0	47.3	39.8	0.3	0.3		1.1	1.7	1.6	0.4	0.3		0.7
Pen G**	2.4	22.3	59.4	11.7	1.3	0.8	1.2	0.4	0.4							
TMS***				0.4	2.5	33.0	56.8	2.8	1.3	0.5	1.1	0.5	0.3	0.7		

	≤ 19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	≥ 34
Oxacillin disk	4.8	1.3	1.3	1.9	2.3	8.7	11.3	14.0	13.4	13.2	9.3	6.6	3.4	3.2	1.1	4.0

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

RESULTS AND COMMENTS

NORM 2003 was the third year of NORM surveillance of *S. pneumoniae* in respiratory tract specimens, the two first being conducted in 2000 and 2001. As in the previous surveys, the vast majority of isolates was fully susceptible to penicillin G with only 21 isolates (2.8%) intermediately susceptible to this agent. Three of these isolates had MICs of 1 mg/L, which would have defined them as penicillin resistant in cerebrospinal fluid specimens. The prevalence of non-susceptibility to penicillin (PNSP) was comparable to the findings in 2000 (2.6%) and 2001 (2.6%).

The oxacillin screening disk for non-susceptibility to β-lactams identified 36 isolates as possible PNSP. Etest confirmed eleven of these isolates as PNSP, whereas 25 were in fact penicillin susceptible. Conversely, 17 of the 21 isolates defined as PNSP by Etest were also examined by the oxacillin disk test, and six of these isolates were not detected by this method. It is thus a question whether the oxacillin screening test is sufficiently sensitive and specific for the present situation of low PNSP prevalence in Norway.

The results for erythromycin and clindamycin cannot be directly compared to the figures from 2000 and 2001 due to changes of breakpoints in 2002. However, the bias is very small due to the low number of isolates around the breakpoints. Reduction of the lower breakpoint for clindamycin has led to a low number of intermediately susceptible strains (0.7% in 2003), which would earlier have been categorized as susceptible. The upper breakpoint for clindamycin has not been changed. The lower breakpoint for erythromycin (S ≤ 0.5 mg/L instead of S ≤ 1 mg/L) did not change the distribution between susceptible and non-susceptible isolates, but reduction of the upper breakpoint (R ≥ 1mg/L instead of R ≥ 4 mg/L) transferred 1.1% of the isolates from the intermediately susceptible to the resistant category. As seen in Figure 23, the prevalence of low-level resistance to erythromycin combined with clindamycin susceptibility is increasing. This is in accordance with the results from blood culture isolates and indicates further dissemination of clones harbouring the *mef*-encoded macrolide efflux mechanism. The prevalence of combined high-level resistance to erythromycin and clindamycin, compatible with *erm*-encoded methylation, was apparently unchanged.

The prevalence of non-susceptibility to doxycycline has increased from 5.5 in 2000 and 4.6 in 2001 to 7.8 in 2003. This can of course be a random fluctuation, but it may also reflect an upward trend in response to the considerable usage of doxycycline for treatment of respiratory tract infections. Further surveillance studies are needed to clarify this issue.

The changes in the prevalences of intermediate susceptibility and resistance to trimethoprim/sulfamethoxazole are entire due to changes of breakpoints. This was underlined by the stable MIC_{50} and MIC_{90} values.

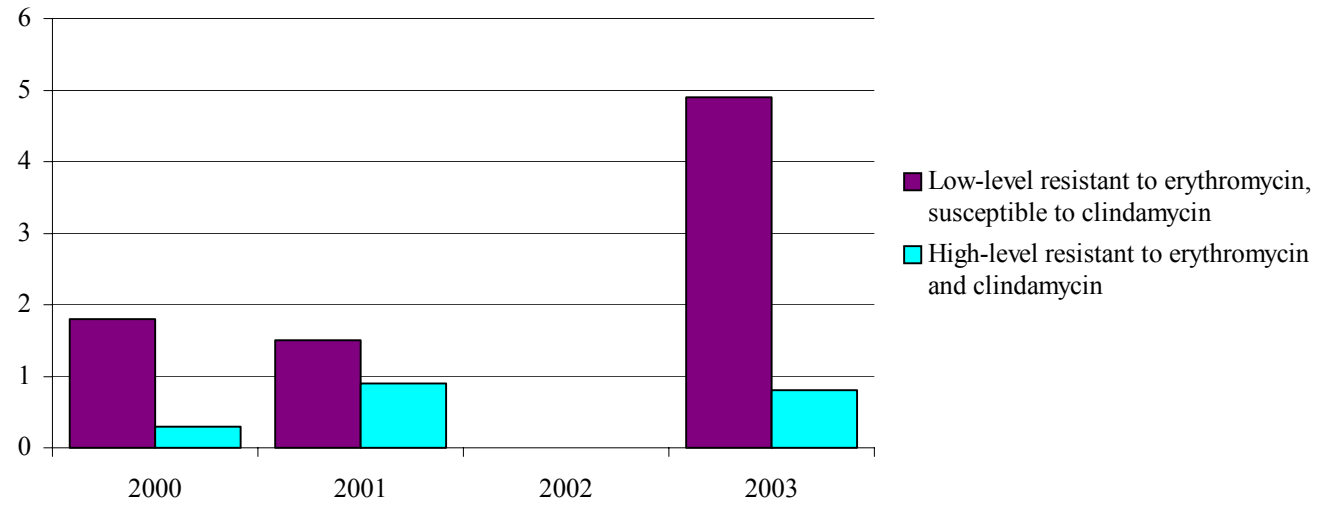

FIGURE 23. Prevalence (%) *of Streptococcus pneumoniae* with high-level resistance to erythromycin and clindamycin or low-level resistance to erytrhomycin and susceptibility to clindamycin in 2000, 2001 and 2003.

Moraxella catarrhalis in respiratory tract specimens

TABLE 54. *Moraxella catarrhalis* in respiratory tract specimens (n=287) analysed by Etest. Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Amoxi/Clav**	≤ 0.5	≥ 32	99.0	1.0	0.0	0.016 - 1	0.125	0.25
Aztreonam	≤ 1	≥ 32	61.0	39.0	0.0	0.125 - 8	1	2
Cefotaxime	≤ 1	≥ 32	97.9	2.0	0.0	0.032 - 4	0.25	1
Cefuroxime	≤ 1	≥ 32	83.6	16.4	0.0	0.032 - 8	1	2
Ciprofloxacin	≤ 0.125	≥ 4	99.0	1.0	0.0	0.008 - 0.5	0.032	0.064
Doxycycline	≤ 1	≥ 16	99.0	1.0	0.0	0.125 - 4	0.5	0.5
Erythromycin	≤ 1	≥ 4	99.3	0.0	0.7	0.032 - 8	0.25	0.25
Pen G***	≤ 1	≥ 32	23.7	40.1	36.2	0.032 - 64	6	32
Tetracycline	≤ 1	≥ 4	99.3	0.0	0.7	0.125 - 32	0.5	0.5
TMS****	≤ 2	≥ 16	99.3	0.0	0.7	0.032 - ≥ 32	0.25	0.5
β-lactamase			9.7	-	90.3			

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

TABLE 55. *Moraxella catarrhalis* in respiratory tract specimens (n=287) analysed by Etest. 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
Amoxi/Clav**			1.0	27.5	16.7	11.8	35.5	6.3	1.0							
Aztreonam						0.3	3.8	32.4	24.4	35.5	3.1	0.3				
Cefotaxime				1.4	9.8	29.6	10.1	27.9	19.2	1.7	0.3					
Cefuroxime				0.7	0.3	0.7	5.6	29.3	47.0	15.7		0.3	0.3			
Ciprofloxacin		0.7	1.7	50.9	43.6	2.1	0.7	0.3								
Doxycycline						5.6	31.0	55.7	6.6	0.3	0.7					
Erythromycin				0.3	1.4	32.8	57.1	6.3	1.4		0.3	0.3				
Pen G***				3.1	5.2	1.0	1.7	2.1	10.5	14.6	9.4	9.4	6.6	36.2		
Tetracycline						3.5	24.0	63.1	8.7				0.3	0.3		
TMS****				0.3	3.8	32.1	48.1	12.9	1.7	0.3			0.3	0.3		

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

TABLE 56. *Moraxella catarrhalis* in respiratory tract specimens (n=287) analysed by the disk diffusion method. Distribution (%) of zone diameters (mm). The Norwegian Reference Group on Antibiotic Susceptibility Testing (AFA) has not determined breakpoints for susceptibility or resistance for this microorganism using disk diffusion methodology.

	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Amoxi/Clav*															
Aztreonam												0.4			
Cefotaxime															
Cefuroxime												0.4			
Ciprofloxacin															
Doxycycline									0.4					0.4	
Erythromycin					0.4	0.4									
Pen G**			2.4	5.2	2.8	5.6	11.5	4.2	7.3	3.1	7.3	3.1	2.4	3.1	7.7
Tetracycline									0.4	0.4	0.4				
TMS***				0.4	0.4		1.0	0.4	0.4	0.7	1.0	0.7	1.8	3.5	4.6

	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35
Amoxi/Clav*						0.7			1.4	1.0	3.5	5.6	3.5	8.7	6.3
Aztreonam			0.4	3.4	7.0	4.5	5.2	9.4	9.4	12.6	5.2	10.1	4.9	7.3	4.5
Cefotaxime				0.4	0.7	1.4	1.0	3.1	4.9	9.4	5.2	8.4	7.3	8.4	4.2
Cefuroxime		1.4	1.0	1.7	5.9	8.4	4.5	8.4	10.4	15.0	8.0	8.7	4.5	4.5	3.5
Ciprofloxacin				0.4	0.7	1.7	1.0	3.8	2.8	5.9	7.0	8.7	4.9	9.4	4.2
Doxycycline	0.7	1.4	2.4	4.2	9.8	13.3	13.3	15.0	4.5	4.9	3.1	5.2	4.5	3.5	1.0
Erythromycin					1.3	1.3	2.1	7.3	5.6	13.3	9.4	9.1	5.6	9.1	10.8
Pen G**	3.1	2.4	4.5	3.8	2.4	1.7	0.4	1.3	0.7	1.0	0.4	1.3		1.7	0.7
Tetracycline						1.3	2.4	9.4	14.0	18.9	7.0	10.8	9.1	5.9	5.6
TMS***	4.2	7.4	6.4	6.4	10.2	8.1	8.5	9.9	5.3	6.4	1.8	3.9	1.4	2.8	0.4

	36	37	38	39	40	41	42	43	44	45	> 45
Amoxi/Clav*	6.3	7.3	11.9	7.0	8.0	2.1	5.2	3.8	5.2	3.5	8.7
Aztreonam	4.2	2.8	2.8	1.0	2.8	0.4	0.4	0.4	0.4		0.4
Cefotaxime	7.7	6.6	7.3	4.9	5.2	2.8	3.1	0.7	2.4	1.7	2.8
Cefuroxime	4.9	1.7	2.4		0.4	0.7	1.0	0.4	0.7		1.0
Ciprofloxacin	8.4	5.6	9.1	3.8	8.4	4.2	3.8	1.4	1.4	0.7	2.4
Doxycycline	3.5	3.5	1.0	1.3	2.8						
Erythromycin	10.5	2.8	4.9	1.3	3.1	0.4	0.7		0.4		
Pen G**	0.7	0.4	1.3	0.7	1.0		0.7	0.3			1.7
Tetracycline	2.8	2.1	1.7	2.1	2.8	1.0	1.7				
TMS***	0.7				1.1						0.4

*Amoxi/Clav=Amoxicillin/clavulanic acid.
**Pen G=Benzylpenicillin.
***TMS= Trimethoprim/sulfamethoxazole. Breakpoints for the TMS combination are given for the trimethoprim component only.

RESULTS AND COMMENTS

Moraxella catarrhalis was included in NORM for the first time in 2003, and to our knowledge this is the first systematic survey of antimicrobial resistance in this species in Norway. The aims of the protocol were to determine the prevalence of resistance to antimicrobials used for treatment of respiratory tract infections, and to evaluate the utility of disk diffusion and Etest for susceptibility testing in *M. catarrhalis*.

The Etest classified 76.3% of isolates as intermediately susceptible or resistant to penicllin G, whereas β-lactamase production was detected in 92.3% of the isolates. As seen in Figure 24, this misclassification would have been reduced by lowering the breakpoint for susceptibility to 0.5 mg/L. The disk diffusion assay was obviously of little value in β-lactams as the population was distributed over a wide range of zone diameters without any clear separation of subpopulations. The results support the recommendation of using a specific β-lactamase test to determine susceptibility to penicillins. The prevalence of β-lactamase production was similar to what has been reported from other Western countries.

The histograms in Figure 24 further demonstrate the distribution of MICs and zone diameters for a range of β-lactams. The majority of *M. catarrhalis* isolates have been found to contain the BroI β-lactamase which efficiently hydrolyses the β-lactam ring. A minority population harbours BroII which is less efficient than BroI. However, there is no direct relationship between hydrolytic activity and the level of resistance. It has been suggested that other factors such as permeability may be involved.

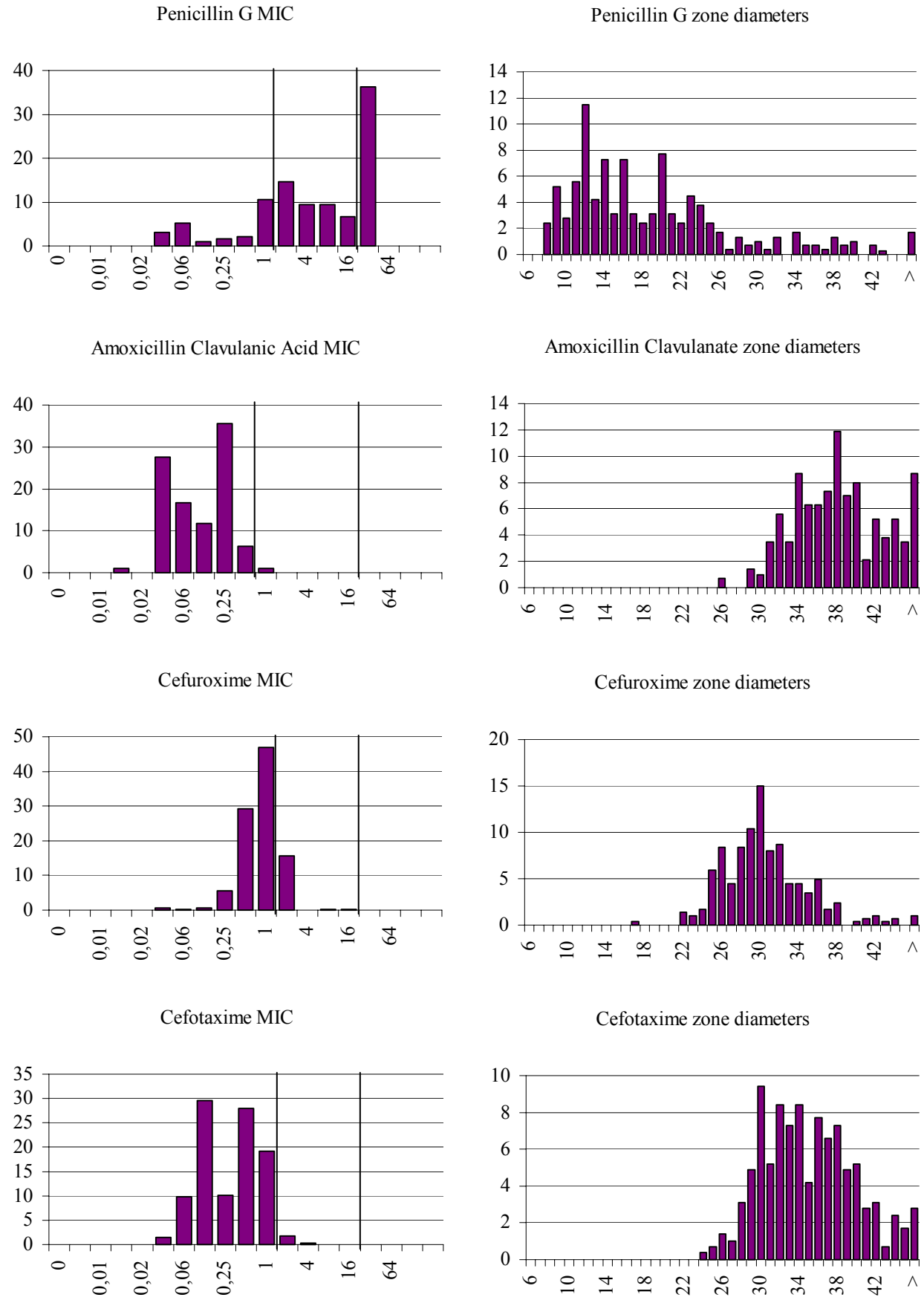

FIGURE 24. Distributions (%) of minimum inhibitory concentrations (mg/L) and disk diffusion zone diameters (mm) of β-lactams in *Moraxella catarrhalis* from respiratory tract specimens. Breakpoints are indicated by vertical black bars.

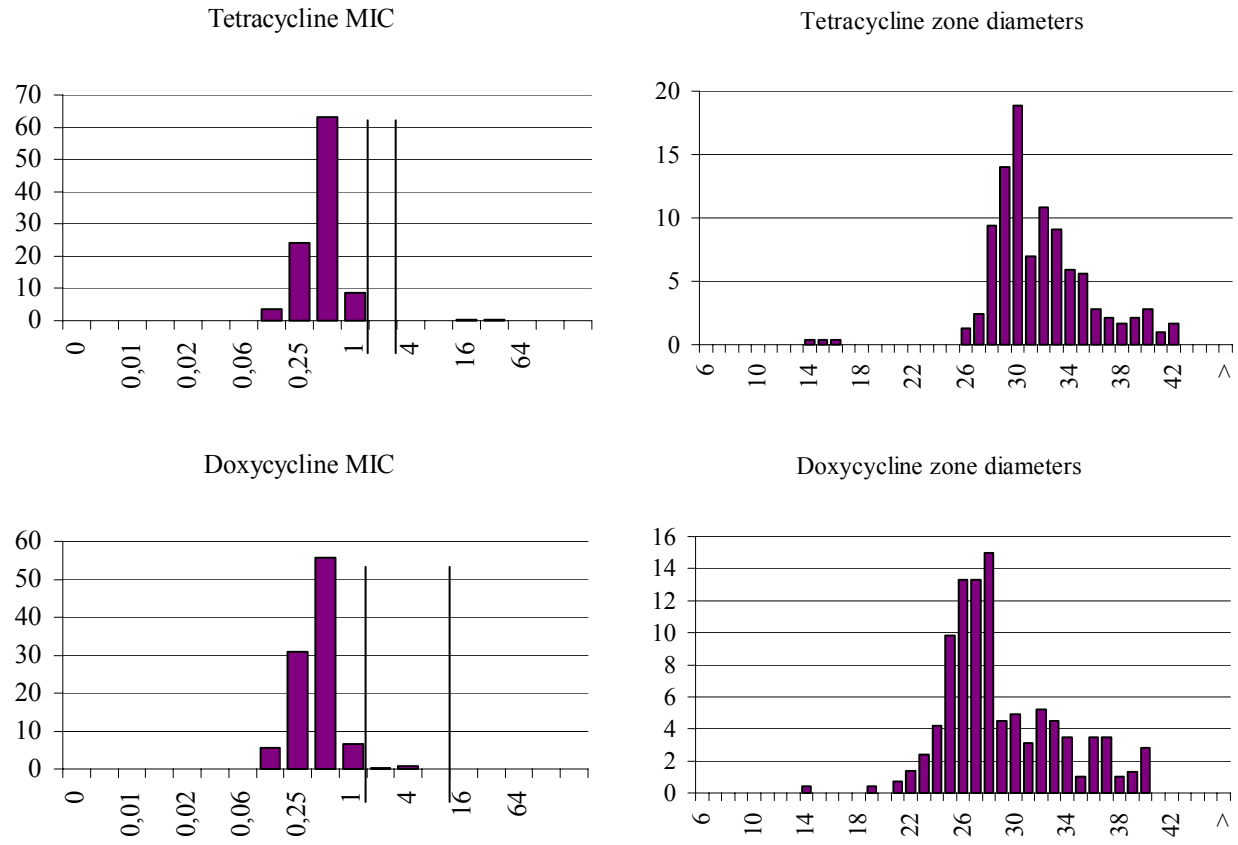

FIGURE 25. Distributions (%) of minimum inhibitory concentrations (mg/L) and disk diffusion zone diameters (mm) of tetracyclines in *Moraxella catarrhalis* from respiratory tract specimens. Breakpoints are indicated by vertical black bars.

As seen in Figure 24, several β-lactams seem to display a biphasic MIC distribution. This is even evident for amoxicillin/clavulanate which one would think should eliminate differences in MIC caused by different β-lactamases. The underlying mechanism for the observed distributions can only be elucidated by molecular characterization of the strain collection.

The isolates were generally susceptible to tetracyclines, and the Etest successfully defined normal distributions around 0.5 mg/L for both tetracycline and doxycycline. However, the zone diameters for tetracycline conformed much better to a normal distribution than was the case for doxycycline. If routine susceptibility testing of *M. catarrhalis* is performed by disk diffusion, tetracycline is obviously a better choice than doxycycline. More than 99% of the isolates were susceptible to ciprofloxacin, trimethoprim/sulfamethoxazole and erythromycin. The three agents also proved reliably distributed both by Etest and disk diffusion (Figure 26).

In conclusion, the majority of *M. catarrhalis* isolates were, as expected, β-lactamase producers, and this phenotype should be tested by a specific enzyme assay. Disk diffusion assays were unable to define normal distributions or well-defined subpopulations for β-lactams, but the Etest data indicate the presence of groups of isolates possibly correlated to different β-lactamases. Almost all *M. catarrhalis* isolates were susceptible to tetracyclines, erythromycin, ciprofloxacin and trimethoprim/sulfamethoxazole. These agents can be tested by disk diffusion with tetracycline being preferred over doxycycline.

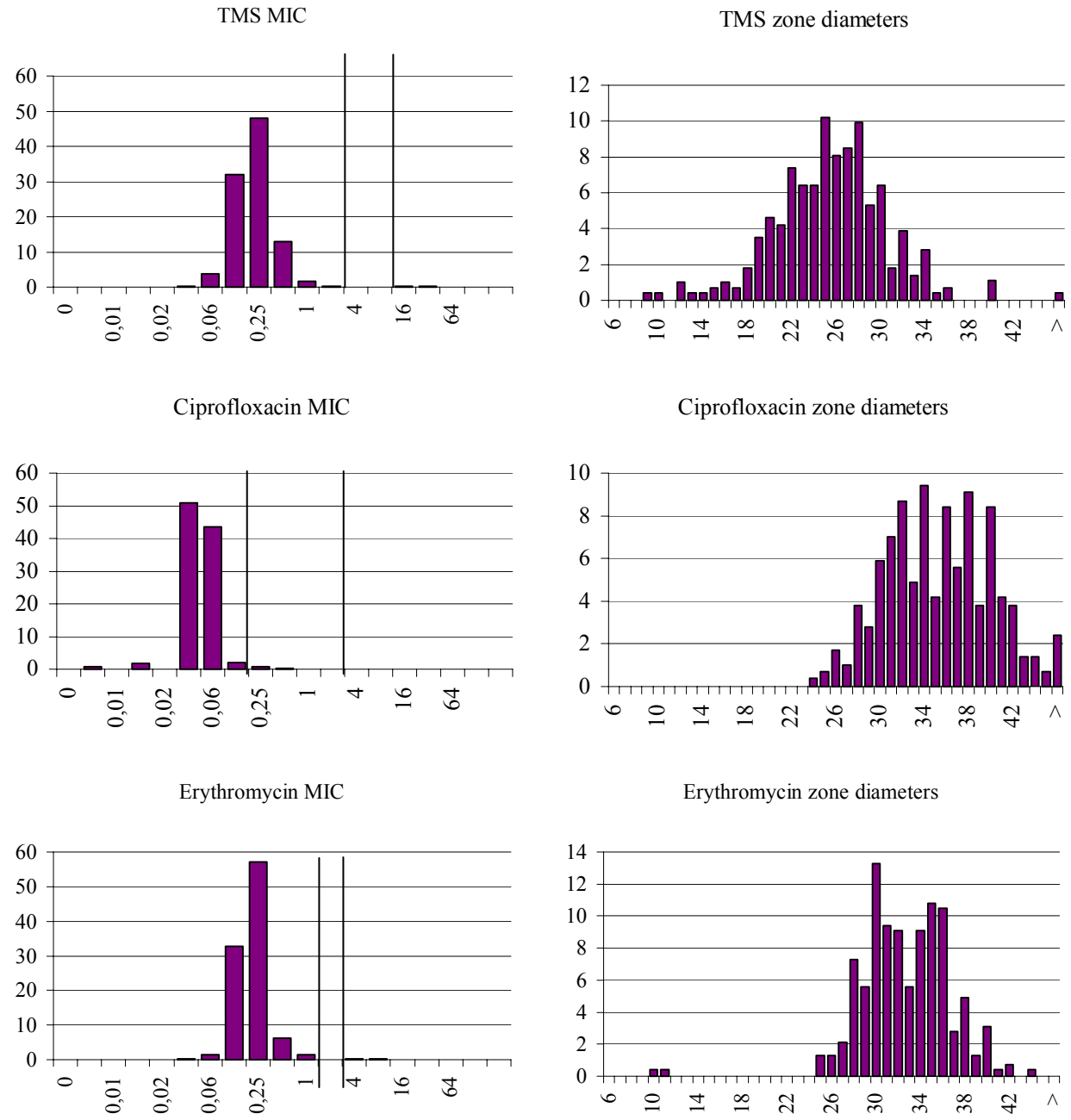

FIGURE 26. Distributions (%) of minimum inhibitory concentrations (mg/L) and disk diffusion zone diameters (mm) of trimethoprim/sulfamethoxazole (TMS), ciprofloxacin and erythromycin in *Moraxella catarrhalis* from respiratory tract specimens. Breakpoints are indicated by vertical black bars.

Staphylococcus aureus in wound specimens

TABLE 57. *Staphylococcus aureus* isolates from wound specimens (n=1,144). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mg/L)		Proportion of isolates (%)*			MIC range (mg/L)	MIC_{50}	MIC_{90}
	S	R	S	I	R			
Clindamycin	≤ 1	≥ 4	99.7	0.1	0.3	0.016 - ≥ 256	0.064	0.125
Doxycycline	≤ 1	≥ 4	95.0	1.0	4.0	0.016 - 128	0.125	0.25
Erythromycin	≤ 1	≥ 4	96.9	0.5	2.6	0.016 - ≥ 256	0.125	0.25
Fusidic acid	≤ 0.5	≥ 1	77.0	-	23.0	0.016 - ≥ 256	0.125	4
Oxacillin	≤ 2	≥ 4	99.7	0.0	0.3	0.016 - 32	0.5	0.5
Oxacillin screen			99.7	-	0.3			
Penicillin V**	≤ 0.064	≥ 0.25	23.8	11.7	64.5	0.016 - ≥ 256	0.25	1
β-lactamase			19.5	-	80.5			
Vancomycin screen			100.0	-	0.0			

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

TABLE 58. *Staphylococcus aureus* isolates from wound specimens (n=1,144). Distribution (%) of MICs (mg/L).*

	≤0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	≥ 256
Clindamycin		1.6	12.8	50.2	33.1	1.2	0.3	0.3	0.1							0.3
Doxycycline		0.6	6.4	23.1	38.7	21.3	3.7	1.2	1.0	1.9	1.0	0.8	0.3		0.1	
Erythromycin		0.8	3.1	10.8	43.7	37.2	1.3		0.5	0.1	0.2	0.1	0.1			2.2
Fusidic acid		5.2	14.4	28.6	23.0	4.7	1.0	2.4	6.6	10.1	1.6	1.1	0.3	0.2	0.1	0.7
Oxacillin		0.1	0.3	1.2	10.6	37.0	42.7	7.0	0.9			0.2	0.1			
Penicillin V**		14.0	5.9	3.9	11.7	22.3	20.6	13.2	3.8	2.1	1.0	0.7	0.4	0.1	0.2	0.1

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

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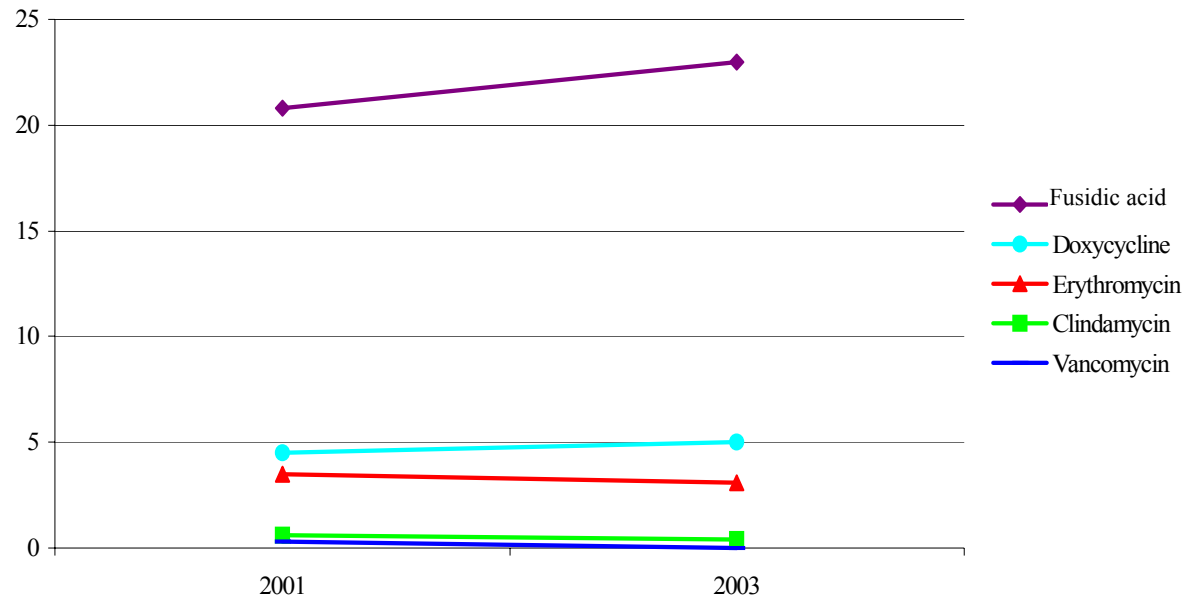

FIGURE 27. Prevalences (%) of non-susceptibility to various antibiotics other than β-lactam in *S. aureus* isolates from wound specimens in 2001 and 2003.

RESULTS AND COMMENTS

Methicillin resistant *S. aureus* (MRSA) isolates were detected in three wound specimens in 2003 (0.3%). This low figure was a slight increase from the findings in NORM 2001 where none of 829 isolates were verified as MRSA by *mec*/*nuc* PCRs. It was not surprising that MRSA should emerge in wound specimens in NORM 2003. The number of MRSA wound infections reported to the Norwegian Surveillance System for Communicable Diseases (MSIS) almost doubled from 97 in 2001 to 189 in 2003. The results nevertheless confirm the very low prevalence of MRSA infections in Norway. The three MRSA isolates were all detected by the oxacillin screening agar containing oxacillin 4 mg/L, and by Etest they had MIC values of 16, 16 and 32 mg/L. Both the screening agar and the oxacillin Etest proved highly specific as no *mecA* negative isolates tested positive in these assays.

Norway has in recent years experienced an epidemic of pediatric skin infections caused by *S. aureus* resistant to fusidic acid. The modest increase from 20.8% fusidic acid resistance in 2001 to 23.0% in 2003 may indicate that the epidemic has now reached its peak. The prevalence of fusidic acid resistance has been much lower in blood culture isolates, but a downward trend can now be seen in the systemic isolates. Future surveillance protocols will determine whether the epidemic is passing or resistance to fusidic acid will remain stable at the present level.

Figure 27 displays the prevalences of non-susceptibility to various antibiotics other than β-lactams. Except for fusidic acid the prevalences have remained stable over the last two years.

Escherichia coli in urine

TABLE 59. *Escherichia coli* urinary tract isolates (n=1,180). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)
	S	R	S	I	R	
Ampicillin	≥ 24	≤ 12	10.1	61.1	28.9	6 - 38
Ciprofloxacin	≥ 25	≤ 20	97.4	0.6	2.0	6 - ≥ 45
Mecillinam	≥ 20	≤ 16	93.5	2.5	4.0	6 - 42
Nalidixic acid	≥ 17	≤ 16	95.1	-	4.9	6 - 36
Nitrofurantoin	≥ 19	≤ 18	96.3	-	3.7	6 - 38
Sulfonamide	≥ 19	≤ 14	73.6	1.8	24.6	6 - 41
Trimethoprim	≥ 21	≤ 19	81.6	0.2	18.3	6 - 40

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

TABLE 60. *Escherichia coli* urinary tract isolates (n=1,180). Distribution (%) of zone diameters (mm).*

	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Ampicillin	25.4	0.7		0.2	0.8	0.6	1.2	1.3	2.3	3.7	4.7	5.4	9.0	8.0	8.5
Ciprofloxacin	1.2	0.1		0.1	0.1	0.2		0.1					0.1	0.1	0.1
Mecillinam	1.2	0.1		0.1	0.1	0.2	0.3	0.2	0.5	0.7	0.8	0.5	1.3	0.8	1.6
Nalidixic acid	4.1	0.2	0.1			0.2			0.1	0.2	0.1	0.1	0.3	0.2	2.4
Nitrofurantoin	0.7		0.2	0.3	0.2		0.2	0.2	0.3	0.4	0.4	0.2	0.8	1.5	3.1
Sulfonamide	23.5	0.6			0.1			0.2	0.3	0.2	0.3	0.3	0.9	0.2	1.6
Trimethoprim	16.5	0.8	0.1	0.1		0.1	0.1	0.1			0.2		0.1	0.3	0.2

	21	22	23	24	25	26	27	28	29	30	31	32	33	34	≥ 35
Ampicillin	6.4	7.1	4.7	4.5	1.4	1.8	0.8	0.6	0.2	0.3	0.2	0.1			0.2
Ciprofloxacin	0.1		0.3	0.3	0.3	0.5	0.8	1.0	0.8	1.7	3.0	7.4	8.7	14.2	59.1
Mecillinam	1.4	1.7	2.4	2.4	3.4	3.0	3.0	4.5	3.5	9.2	10.4	12.0	9.8	11.7	13.3
Nalidixic acid	1.4	4.7	4.9	6.9	9.7	13.3	10.9	15.2	8.6	9.0	2.0	3.2	0.6	1.1	0.5
Nitrofurantoin	3.3	6.4	8.2	10.9	14.8	16.8	10.3	9.5	3.9	4.2	1.2	0.7	0.2	0.6	0.7
Sulfonamide	2.0	3.0	3.4	5.0	7.3	8.3	6.5	7.7	4.7	9.2	2.8	4.5	1.6	2.2	3.7
Trimethoprim	0.2	0.3	0.4	0.3	0.8	2.1	3.0	6.8	5.4	14.0	8.9	12.3	7.8	9.0	10.5

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

RESULTS AND COMMENTS

Urinary tract isolates of *E. coli* are analysed by disk diffusion in NORM. This protocol is used to obtain the dual aims of conducting surveillance and of improving the quality of routine susceptibility testing in Norwegian diagnostic laboratories. The results from the surveillance program have enabled the Norwegian Reference Group on Antibiotic Susceptibility Testing (AFA) to optimize both the MIC breakpoints and the zone diameter interpretations of the breakpoints. However, the changing zone criteria make it difficult to directly compare results between years.

The prevalence of non-susceptibility to ampicillin remained around 90%, as *E. coli* by definition is considered only intermediately susceptible to this agent. The results do not imply that ampicillin will not be effective against *E. coli* in anatomical locations where drug concentrations are high i.e. the urinary tract.

Non-susceptibility to other antimicrobials traditionally used for treatment of urinary tract infections are seen in Figure 28. The prevalences of resistance to trimethoprim and sulfamethoxazole remained stable around 18% and 24%, respectively. Non-susceptibility to mecillinam was also virtually unchanged which is almost surprising due to the difficulties with defining a cut-off value for this microbe/antimicrobial combination (Figure 29). Resistance to nitrofurantoin fluctuated between 2.9% and 4.4% in the period 2000 to 2002, and it did not seem to change significantly in 2003 with 3.7%.

Total non-susceptibility to ciprofloxacin was unchanged from 2002 to 2003 (2.6%), but a slight shift from intermediate susceptibility to resistance was noted. The increasing prevalence of resistance to nalidixic acid may indicate more widespread resistance to quinolones in the future. Only two isolates were reported as susceptible to nalidixic acid and resistant to ciprofloxacin in the raw data. This suggests an improved quality of microbiological analysis and/or interpretation in the laboratories.

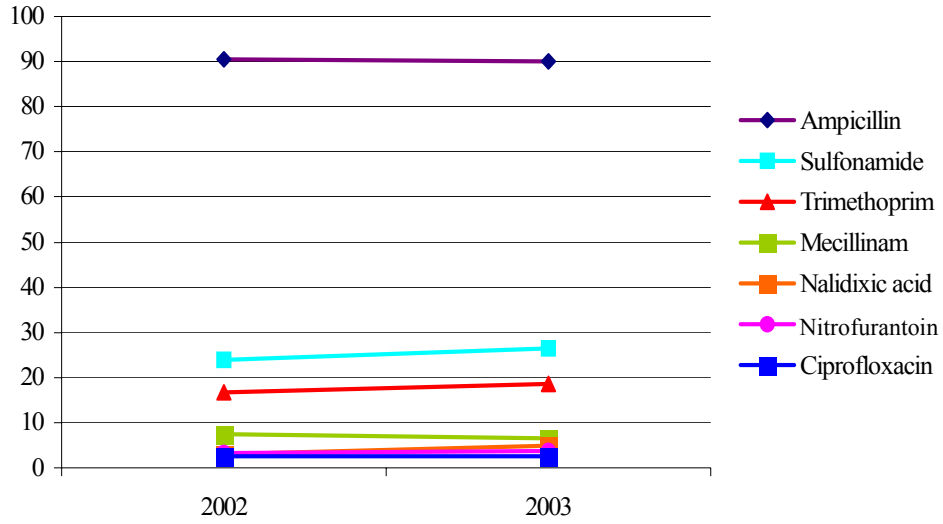

FIGURE 28. Prevalences (%) of non-susceptibility to various antimicrobials in urinary tract *E. coli* in 2002 and 2003.

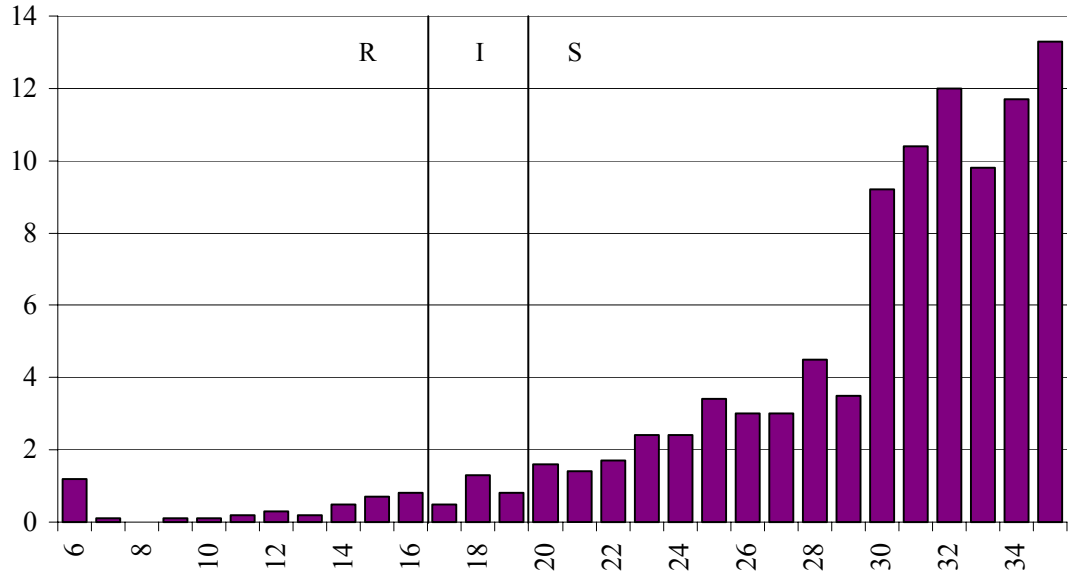

FIGURE 29. Distribution (%) of disk diffusion diameters (mm) of mecillinam for *E. coli* urinary tract isolates from 2003 using 10 µg disks. A small zone diameter indicates resistance, whereas a large zone diameter indicates susceptibility. AFA breakpoints are indicated by vertical black bars.

Klebsiella spp. *coli* in urine

TABLE 61. *Klebsiella* spp. urinary tract isolates (n=1,028). Sampling, laboratory methods, and data handling are described in Appendix 5.

	Breakpoints (mm)		Proportion of isolates (%)*			Range (mm)
	S	R	S	I	R	
Ampicillin	≥ 24	≤ 12	2.4	20.4	77.1	6 - 38
Ciprofloxacin	≥ 25	≤ 20	94.0	3.8	2.2	8 - ≥ 45
Mecillinam	≥ 20	≤ 16	89.4	1.9	8.7	6 - 42
Nalidixic acid	≥ 17	≤ 16	89.7	-	10.3	6 - 36
Nitrofurantoin	≥ 19	≤ 18	71.9	-	28.1	6 - 38
Sulfonamide	≥ 19	≤ 14	86.6	3.3	10.0	6 - 41
Trimethoprim	≥ 21	≤ 19	83.5	1.1	15.4	6 - 40
ESBL			99.3	-	0.7	

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

TABLE 62. *Klebsiella* spp. urinary tract isolates (n=1,028). Distribution (%) of zone diameters (mm).*

	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Ampicillin	39.3	5.4	7.8	4.2	9.9	4.6	5.9	6.2	3.5	2.5	2.5	1.3	1.5	0.1	1.2
Ciprofloxacin			0.2		0.3		0.3		0.2	0.2	0.2		0.3	0.1	0.5
Mecillinam	3.6	0.1	0.4	0.3	0.5	0.8	0.4	0.7	0.7	0.6	0.7	0.4	1.0	0.6	1.1
Nalidixic acid	4.8	0.5	0.4	0.1	0.5	0.7	0.7	0.3	1.0	0.7	0.8	1.0	1.2	1.7	4.6
Nitrofurantoin	2.4	0.1	0.5	1.1	2.2	1.5	1.8	1.8	1.8	2.1	3.5	3.5	5.9	6.1	9.7
Sulfonamide	8.0	0.7	0.2	0.1		0.1	0.2	0.4	0.4	0.6	0.5	0.5	1.8	1.3	4.2
Trimethoprim	8.8	0.6	0.3	0.3	0.8	0.3	0.6	0.8	0.7	0.2	0.4	0.6	0.7	0.4	1.1

	21	22	23	24	25	26	27	28	29	30	31	32	33	34	≥ 35
Ampicillin	0.5	0.5	0.5	0.8		0.7	0.1	0.1	0.1	0.1	0.1	0.2	0.1		0.2
Ciprofloxacin	0.6	0.9	1.1	1.3	1.2	1.1	1.3	3.0	5.1	12.6	10.6	16.5	9.6	11.7	21.4
Mecillinam	1.3	2.1	2.5	2.7	4.8	5.6	6.2	9.6	8.2	15.5	9.4	8.1	5.5	3.0	3.8
Nalidixic acid	7.0	10.2	12.0	11.7	10.7	12.5	5.8	5.2	2.7	2.2	0.7	0.2		0.1	0.2
Nitrofurantoin	7.6	9.5	8.2	7.5	5.9	5.8	3.5	3.6	0.9	1.3	0.4	0.1	0.3	0.7	0.8
Sulfonamide	2.5	5.5	5.2	8.1	7.7	10.9	6.5	10.7	3.8	8.5	2.5	4.2	1.3	1.7	2.2
Trimethoprim	0.4	1.1	2.2	3.1	6.2	8.3	9.3	10.7	7.4	15.5	5.1	6.3	2.6	3.1	2.4

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

RESULTS AND COMMENTS

Klebsiella spp. isolates from urinary tract samples were last analysed in NORM in 2001. Except for nitrofurantoin, all zone diameter breakpoints have been adjusted in order to accommodate new MIC breakpoints or to better delineate the normal distribution. It is therefore impossible to compare resistance rates directly between 2001 and 2003.

Phenotypical ampicillin resistance was found in 77.1% of isolates, whereas 20.4% were intermediately susceptible. However, these figures do not imply that a subpopulation of *Klebsiella* spp. is *in vitro* susceptible to ampicillin. All *Klebsiella* spp. should be categorized as resistant to ampicillin due to chromosomally encoded β-lactamases such as SHV in *Klebsiella pneumoniae* and K1 in *Klebsiella oxytoca*.

More than 80% of isolates were fully susceptible to traditional urinary tract antimicrobials such as mecillinam, trimethoprim and sulfonamide. The breakpoint for nitrofurantoin has remained unchanged, and the prevalence of resistance to this agent is still considerably higher in *Klebsiella* spp. (30.7% in 2001 and 28.1% in 2003) than in *E. coli* (2.9% in 2001 and 3.7% in 2003).

The prevalence of non-susceptibility to ciprofloxacin decreased from 18.7% in 2001 to 6.0% in 2003, but this was entirely due to the changing of breakpoints. The figures for urinary tract isolates (3.8% intermediate susceptibility and 2.2% resistance) were comparable to the corresponding numbers from blood culture isolates (8% intermediate susceptibility and 0.3% resistant) but may indicate a shift towards higher levels of resistance in urinary tract isolates. The 10% prevalence of resistance to nalidixic acid confirms the validity of the results for ciprofloxacin.

Mycobacterium tuberculosis

A total of 339 cases of tuberculosis were reported to the Norwegian Surveillance System for Communicable Diseases (MSIS) in 2003. Of these patients, 320 had not previously been treated with antituberculosis drugs. *Mycobacterium tuberculosis* was isolated in 272 cases and *Mycobacterium africanum* in one case. Susceptibility tests were performed on 272 isolates as one isolate was contaminated before susceptibility testing was performed. Antimicrobial susceptibility testing of *Mycobacterium tuberculosis* complex isolates was performed for 261 patients not previously treated for tuberculosis.

TABLE 63. Antimicrobial susceptibility of *Mycobacterium tuberculosis* complex isolates from 261 patients not previously treated for tuberculosis - 2003.

Geographical origin of patient	No. of isolates	Resistance to antimicrobial agents (isolates)				
		Isoniazid	Rifampicin	Ethambutol*	Streptomycin**	MDRTB
Norway	45	2			1	
Europe outside Norway	18	2	1		5	1***
Asia	84	6			7	
Africa	111	9	1	1	19	
America	3					
Total	261	19	2	1	32	1
Proportion of isolates resistant %		7	0.8	0.4	12.8	
Isolates tested		261	261	261	250	

* Some isolates were tested with a higher concentration of ethambutol. Resistance may therefore be under-reported and will be revised.
** Isolates from 11 patients were not tested for susceptibility to streptomycin
*** One isolate from a patient not previously treated showed combined resistance to rifampicin (R) and isoniazid (H) and was thus multidrug resistant by usual definitions. However, the isolate was susceptible to other first-line drugs.

Susceptibility tests were also performed on *Mycobacterium tuberculosis* isolates from 11 patients who had previously received antituberculosis drug treatment. Two isolates from East Europe were multidrug resistant (MDR) strains. By the usual definition of MDRTB as isolates resistant to rifampicin and isoniazid, three patients (2 previously treated and 1 previously not treated) were therefore diagnosed with MDR tuberculosis in Norway in 2003. In addition, one previously treated patient had an isolate resistant to isoniazid, ethambutol and streptomycin. One isolate was monoresistant to pyrazinamid.

Appendix 1:
Collection of data on usage of antimicrobials in animals

Data sources

Feed additives

The Norwegian Food Safety Authority is responsible for approving and monitoring sales of feed additives including antimicrobial growth promoters and coccidiostats. Reliable data on the use of different substances and categories of feed additives can be obtained from this agency.

Antimicrobial drugs for therapeutic use

In Norway, veterinary antimicrobials for therapeutic use in domestic animals or farmed fish are prescription drugs only. Moreover, veterinary antimicrobial drugs 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 drugs is possible, again subject to veterinary prescription, through the drinking water or as a top dressing on the feed.

The sales figures for veterinary antimicrobial drugs from wholesalers and feed mills are thought to roughly equal the use of these drugs. Sales figures for veterinary antimicrobial drugs are therefore used as synonyms 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 1, 2002 to ensure that all the data are included. Crude sales data of veterinary antimicrobial drugs were obtained from the Norwegian Institute of Public Health and calculated to express kg active substance.

Drug classification system

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

Unit of measurement

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

All veterinary antimicrobial specialities included in this report belong to the following ATCvet groups: gastrointestinal infections (QA07AA), uterine infections (QG01AA+AE), and antimicrobial drugs for systemic use (QJ), including intramammary dose applicators (QJ51). The QJ-group also includes medicated feeds and premixes for farmed fish that are approved by the drug authorities and classified as pharmaceutical specialities (QJ01).

Dermatological preparations (QD) and preparations intended for sensory organs (QS) are not included in the material. Human antimicrobial preparations can be used in small animal practice. However, data on the use of these drugs in animals are not included in this report, as such usage cannot be separated from use in humans.

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

Data sources
In Norway, antimicrobials are prescription only drugs (POM), and only allowed sold by pharmacies. Antimicrobials are normally not reimbursed, exceptions are venereal diseases and chronic infections. Drug statistics on usage of antimicrobials for humans are based on sales of medicaments from drug wholesalers to pharmacies and hospitals in Norway. This data cover total sales of antimicrobials for humans in Norway. Sales to hospitals represent around 8% of the total use of antimicrobials for humans.
The figures presented should be regarded as maximum figures with the assumption that all medicaments sold from the wholesalers 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 has been collected in Norway since the beginning of the seventies.

Drug classification
The data are categorized according to the ATC classification system and Defined Daily Doses (DDDs) are employed as units of measurement. The ATC/ DDD index version 2004 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 use over time, nationally and internationally. The DDD is a theoretical unit of measurement, and does not necessarily reflect the recommended or Prescribed Daily Dose.

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

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

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

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

Sampling strategy
Clinical isolates from animals in 2003 were collected from diagnostic submissions: *Staphylococcus aureus* and coagulase negative *Staphylococcus* spp. (CoNS) from cattle with mastitis and *Staphylococcus aureus* from sheep and goats with mastitis. Milk samples were collected by veterinary practitioners. Only one isolate per herd were tested for antimicrobial susceptibility.
Isolates of *Moritella viscosa* from Atlantic salmon (*Salmo salar*) were collected from diagnostic submissions to the National Veterinary Institute during the periods 1987-1992 and 2000-2002.
Indicator bacteria *Escherichia coli* and *Enterococcus* spp. (*E. faecalis* and *E. faecium*) included in the NORM-VET monitoring programme 2003 were collected from cattle and sheep. Faecal samples (from caecum or colon) were collected at slaughterhouses and samples of minced cattle meat were collected at cutting plants. The sampling period was from January to December. The Municipal Food Control Authorities collected the samples at slaughterhouses. To obtain a representative random sample from cattle and sheep, the number of faecal samples collected at each slaughterhouse was determined by the proportion of animals slaughtered there relative to the total number of animals slaughtered in Norway in 2002. Abattoirs that slaughtered > 1% of the total delivered slaughter in 2002 were included. The meat samples from cattle were collected from 16 cutting plants (10 samples at each facility distributed over the year).
In addition to the monitoring programme, indicator bacteria from faecal samples collected in the National Health Surveillance Programme for Cervids were included.

Isolation and identification of bacteria
Escherichia coli
The *E. coli* isolates included in NORM-VET 2003 were isolated and identified at the National Veterinary Institute. Meat: Five grams of material from each specimen were incubated in 45 ml of MacConkey broth (Oxoid). After incubation at 44°C for 24 h, a small amount (approx. 10 µl) of broth was plated onto the surface of lactose-saccarose-bromthymol blue agar. Faeces: Intestinal content was gathered on swabs and plated directly onto the surface of lactose-saccarose-bromthymol blue agar without broth enrichment.
After incubation of the agar plates at 37°C for 24 h, a typical colony was plated onto blood agar (Heart infusion agar (Difco) containing 5% bovine blood). Colonies were identified as *E. coli* by typical appearance, lactose and/or saccarose fermentation and a positive indole reaction.

***Enterococcus* spp.**
The enterococcal isolates included in NORM-VET 2003 were isolated and identified at the National Veterinary Institute. Meat: Five grams of material from each specimen were incubated in 45 ml of Azide dextrose broth (Oxoid). After incubation at 44°C for 24 h, a small amount (approx. 10µl) of broth was plated onto the surface of Slanetz & Bartley agar (Oxoid). Faeces: Intestinal content was gathered on swabs and plated directly onto the surface of Slanetz & Bartley agar (Oxoid) without broth enrichment.
After incubation of the agar plates at 44°C for 48h, a typical colony was plated onto blood agar (Heart infusion agar (Difco) with 5% bovine blood). Typical colonies were tested by negative catalase reaction and *E. faecium* and *E. faecalis* were identified by *ddl*-PCR (Dutka-Malen et al., 1995).

Moritella viscosa
M. viscosa isolates were identified using the species description according to Lunder, T. et al., Int J Syst Evol Microbiol. 2000.

***Staphylococcus* spp.**
The staphylococcal isolates included in NORM-VET 2003 were isolated at the National Veterinary Institute or at the Mastitis Laboratory in Molde. Secretions (0.01 ml) were plated on blood agar (Heart infusion agar (Difco) containing 5% washed bovine erythrocytes). The plates were incubated at 37°C for 24 and 48 h. If no growth was detected after incubation for 24 h, the original secretion sample was preincubated for 4 h at 37°C, and a larger inoculum (0.05 ml) was cultivated on another blood agar as described above. Species identification of the isolates was performed at the National Veterinary Institute and was based on the occurrence of haemolytic zones, Gram stain, production of catalase and coagulase, growth on peptone agar with acriflavine and anaerobic fermentation of manitol. The Staph-Zym® biochemical test kit (Rosco) was used to identify the coagulase negative staphylococci.

Susceptibility testing
Only one isolate per herd or product were tested for antimicrobial susceptibility. Bacterial isolates were tested for antimicrobial susceptibility at the National Veterinary Institute. The VetMIC™ microdilution method (Dept. of Antibiotics, National Veterinary Institute, Sweden) was used for the susceptibility testing of all isolates but *Moritella viscosa.* For this pathogen, an in-house broth dilution method was used. All *Staphylococcus* spp. isolates were tested for production of β-lactamase using the cloverleaf method.
Microbiological cut-off values were used to classify the isolates as resistant or susceptible (Appendix 6). The microbiological cut-off value is defined as the highest MIC-value of isolates that belong to the original genetically unchanged population (wild-type). It classifies the isolates with a MIC-value greater than the microbiological cut-off value as resistant. However, NCCLS breakpoints were applied when available and appropriate.

Quality assurance systems

The following bacteria were included as quality controls on a weekly basis, *E. coli* ATCC 25922, *E. faecalis* ATCC 29212 and *S. aureus* ATCC 29213. The following resistant bacteria were tested on a regular basis: *S. aureus* CCUG 35602 and CCUG 35603, *E. faecium* CCUG 33829 and CCUG 36804 and *E. faecalis* CCUG 37389. The results were approved according to reference values given by NCCLS when available.

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

Data processing

Susceptibility test results were recorded and processed in WHONET 5.3, a program developed by the World Health Organization (WHO) for analysis of antimicrobial resistance data (**ftp.who.int/data/cds/csreph**). The susceptibility data were stored as continuous values (MIC). In addition data was imported into SAS, Enterprise guide V.2. to obtain exact 95% confidence intervals for the prevalence's of resistance. For this purpose the susceptibility data were categorised as susceptible or resistant, respectively, as defined by the relevant microbiological cut-off value. The function Proc freq, using exact binomial proportion test for one-way tables, was used for the calculation of prevalences of resistance including 95% confidence intervals.

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

Sampling strategy - animals

Salmonella

Samples from animals were collected according to The Norwegian Salmonella Control Programme for Live Animals. Additional samples were obtained from animals during clinical examinations or necropsies at the National Veterinary Institute. One isolate of each serovar per incident was included for susceptibility testing.

Campylobacter jejuni

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

Sampling strategy - humans

Salmonella, Yersinia enterocolitica* and *Shigella

All the human isolates were obtained from clinical specimens. One isolate per patient was included for susceptibility testing.

Campylobacter

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

Isolation and identification of bacteria

Isolation and identification of *Salmonella* from animals was carried out by the National Veterinary Institute according to the Nordic Committee on Food Analyses (NMKL) method number 71. Isolation of *Campylobacter jejuni* from broilers was carried out by the National Veterinary Institute or local food control laboratories according to the Nordic Committee on Food Analyses (NMKL) method number 119, with minor modifications. Identification of *C. jejuni* was carried out by the National Veterinary Institute or the Norwegian Institute of Public Health.

Isolation and identification of bacteria from humans were performed according to conventional methods described in standard reference literature (e.g. the ASM Manual of Clinical Microbiology, Edwards and Ewings Identification of Enterobacteriaceae). The identification of all isolates from animals and humans were verified at the National Reference Laboratory for Enteropathogenic Bacteria at the Norwegian Institute of Public Health.

Susceptibility testing

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

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

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

Quality assurance systems

The National Veterinary Institute and the Reference Laboratory for Enteropathogenic Bacteria at the Norwegian Institute of Public Health have a quality assurance system according to the requirements of NS-EN ISO/IEC 17025.

Campylobacter jejuni subsp. *jejuni* CCUG 33057 and CCUG 33560 were used as quality control at the National Veterinary Institute on a weekly basis. The National Veterinary Institute participates in an external quality assurance programme for veterinary pathogens organized by the VLQAS (Veterinary Laboratories Agency Quality Assurance Unit, Loughborough, England) and participates also in the external quality assurance programmes organized by ARBAO-II http://fvm-dvi.dynamicweb.dk/Default.asp?ID=9753 and Global Salm-Surv (GSS) http://www.who.int/salmsurv/en/

The Norwegian Institute of Public Health participates in the external quality assessment programme for *Salmonella* organized by Enter-Net.

Data processing

Susceptibility data were recorded and processed in WHONET5.3, a program developed by the World Health Organization (WHO) for analysis of antimicrobial resistance data (ftp.who.int/data/cds/csreph). The susceptibility data were stored as continuous values (MIC). In addition the animal data concerning *C. jejuni* was imported into SAS, Enterprise guide V.2. to obtain exact 95% confidence intervals for the prevalence's of resistance. For this purpose the susceptibility data were categorised as susceptible or resistant, respectively, as defined by the relevant cut-off value. The function Proc freq, using exact binomial proportion test for one-way tables, was used for the calculation of prevalences of resistance including 95% confidence intervals.

Appendix 5
Sampling, microbiological methods and data processing in NORM

General considerations
NORM is based upon periodic sampling of bacteria from patients with respiratory tract infections, wound infections, urinary tract infections, or septicaemiae. For enteric infections see Appendix 4. 2003 was the fourth year of surveillance, and all twenty-four laboratories in Norway participated in the surveillance system in addition to the Norwegian Institute of Public Health. The surveillance strategy is based on sampling and local testing of bacterial isolates from defined clinical conditions. All laboratories follow the same sampling strategy and use identical criteria for the inclusion of bacterial strains. Only one isolate per patient and infectious episode is included. All bacteria were identified using conventional methods as described in the ASM Manual of Clinical Microbiology (8th ed). The surveillance period started in the beginning of January, and consecutive bacterial isolates were included up to a defined maximum of isolates for each surveillance category. The surveillance categories in 2003 were: *E. coli*, *Klebsiella* spp., *Staphylococcus aureus*, *Streptococcus pneumoniae* and *Enterococcus* spp. from blood cultures; *Streptococcus pneumoniae* and *Moraxella catarrhalis* from respiratory tract infections, *S. aureus* from wound infections and *E. coli* and *Klebsiella* spp. from urinary tract infections. Blood culture isolates, respiratory tract isolates and isolates from wound specimens were tested using Etest, while isolates from urinary tract infections were examined by a disk diffusion method in accordance with the Norwegian Reference Group on Antibiotic Susceptibility Testing (AFA). *M. catarrhalis* isolates were tested by both methods. All resistance values were recorded either as MICs or mm inhibition zone sizes in order to monitor trends in the occurrence of resistance. Suspected MRSA (*S. aureus* with oxacillin MIC $\geq$ 4 mg/L) were examined by *mecA* PCR, and suspected VRE (enterococci growing on BHI with 6 mg/L vancomycin) were examined by PCRs for the *van* gene complex. The NORM computer program was used for the registration of patient data, sample data and resistance data. Data were analyzed by WHONET5 with the aid of a special program (NORMlink developed by John Stelling) converting the data base structure of NORM to a single file format.

Blood culture isolates
Consecutive isolates of up to 50 each of *E. coli*, *S. aureus*, and pneumococci, up to 25 isolates of *Klebsiella* spp., and up to 20 isolates of enterococci from January until testing time in September to October were included in the surveillance. All isolates were identified to the species level using conventional bacteriological methods. All isolates were tested using Etest (AB Biodisk, Solna, Sweden). A total of 966 isolates of *E. coli*, 299 isolates of *Klebsiella* spp, 637 isolates of *S. aureus* and 252 isolates of enterococci were tested on PDM agar at 35ºC in ambient air, while the 514 isolates of pneumococci were tested on PDM (AB Biodisk, Solna, Sweden) agar supplemented with 5% lysed horse blood at 35ºC in 5% CO_2. All *E. coli* and *Klebsiella* spp. isolates were tested for ESBL production using a disk approximation test including amoxicillin/clavulanic acid, aztreonam, ceftazidime, cefotaxime and cefpirome. All *S. aureus* isolates were tested for β-lactamase production using the nitrocefin disk, the acidometric agar plate (3.6 mg/L penicillin G and phenol red) or the clover leaf method. All *S. aureus* isolates were screened for methicillin resistance using MH agar (Difco) with 4% NaCl and oxacillin 4 mg/L and a spot inoculum of 10^6 cfu/spot. All enterococci were screened for vancomycin resistance using BHI agar (Difco) and vancomycin 6 mg/L. The following strains were used for quality control: *E. coli* ATCC 25922, *K. pneumoniae* ATCC 700603 (ESBL positive), *E. faecalis* ATCC 29212, *S. pneumoniae* ATCC 49619, *S. aureus* ATCC 29213, *S. aureus* ATCC 43300 (heterogeneous methicillin resistance), and *S. aureus* CCUG 35600 (homogeneous methicillin resistance).

Respiratory tract isolates
Up to 50 consecutive isolates each of *S.pneumoniae* and *M. catarrhalis* from patients with respiratory tract infections were collected in each laboratory from January to March. The *M. catarrhalis* survey was optional for the laboratories. All isolates were kept in a freezer and tested in batch using Etest and paper disks (*M. catarrhalis*) from AB Biodisk, Solna Sweden. A total number of 752 *S. pneumoniae* and 287 *M. catarrhalis* were included in the study. Both species were tested on PDM agar supplemented with 5% lysed horse blood at 35ºC in 5% CO_2. *M. catarrhalis* was in addition tested for β-lactamase production as described above. *S. pneumoniae* ATCC 49619 was used for quality control.

Wound specimens
Up to 50 consecutive isolates of *S. aureus* from patients with wound infections were collected in each laboratory from January to March. All isolates were kept in a freezer and tested in batch using Etest (AB Biodisk, Solna Sweden). A total of 1,144 *S. aureus* were included in the study. The isolates were analysed as described above for blood culture isolates.

Urinary tract isolates
Up to 50 consecutive isolates each of *E. coli* and *Klebsiella* spp. from patients with urinary tract infections were collected in each lab during January and February. All isolates were either kept on bench or in a freezer until tested in batch using a disk diffusion method with PDM agar and paper disks (AB Biodisk, Solna Sweden) at 35ºC in ambient air. The study included 1,180 *E. coli* isolates and 1,028 *Klebsiella* spp. isolates. *E. coli* ATCC 25922 was used for quality control.

Mycobacterium tuberculosis
In the year 2003, antimicrobial susceptibility testing of *M. tuberculosis* was performed at the following institutions: Norwegian Institute of Public Health, Oslo, Ullevål University Hospital, Oslo, National Hospital, Oslo, and Haukeland Hospital, Bergen. The majority of isolates were tested using the BACTEC (Norwegian Institute of Public Health and Ullevål University Hospital) or MGIT systems (National Hospital). All four laboratories participate in an external quality control program organized by the WHO.

Appendix 6
Breakpoints NORM-VET

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

Antimicrobials	Resistant (MIC values, mg/L)	*Campylobacter*	*E. coli* / *Salmonella*	*Staphylococcus*	*Enterococcus*
Oxytetracycline	> 2	■		■	
	> 4				■
	> 8		■		
Chloramphenicol	> 16		■	■	■
Florfenicol	> 16		■		
Ampicillin	> 8		■		■
	> 16	■			
Amoxi./clav	> 16		■		
Penicillins	Based upon β-lactamase production			■	
Oxacillin	> 2			■*	
Cephalothin	> 1			■	
Ceftiofur	> 2		■		
Trimethoprim	> 4		■		
	> 8			■	
Sulfonamides	> 256		■		
Erythromycin	> 2			■	
	> 4				■
	> 8	■			
Clindamycin	> 2			■	
Streptomycin	> 16		■**		
	> 32		■**	■	
	> 256				■
Gentamicin	> 2			■	
	> 4	■	■		
	> 256				■
Neomycin	> 2			■	
	> 4		■		
	> 256				■
Enrofloxacin	> 0.125		■		
	> 0.5	■		■	
Nalidixic acid	> 16	■	■		
Vancomycin	> 4			■	■
Fusidic acid	> 0.5			■	
Avilamycin	> 16			■	■
Bacitracin	> 32				■
Flavomycin	> 32				■
Virginiamycin	> 4			■	
	> 8				■
Narasin	> 2				■

* *Staphylococcus aureus*
** > 16 for *Escherichia coli*, > 32 for *Salmonella* spp.

Appendix 7
Breakpoints NORM

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

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

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