NATIONAL MASTITIS COUNCIL
RESEARCH COMMITTEE REPORT

BOVINE MASTITIS PATHOGENS AND TRENDS IN RESISTANCE TO ANTIBACTERIAL DRUGS

Subcommittee of the NMC Research Committee
Ron Erskine, Jim Cullor, Mel Schaellibaum, Bob Yancey and Alfonso Zecconi

Introduction

Bovine mastitis is the single most common cause for antibacterial use in lactating dairy cattle (18, 24). Treatment of this disease is also the most common cause of illegal antibacterial residues in marketed milk (10). Antibacterial therapy of bacterial-induced diseases in cattle has been incriminated as a catalyst for resistance in bacteria isolated from treated animals, other animals within the herd, and food derived from cattle for human consumption (2, 15, 30, 37, 41). Additionally, antibacterial use has been suggested as a selective force in determining the bacterial ecology of bovine mastitis (25). Numerous studies have determined the antibacterial susceptibility patterns of mastitis pathogens isolated from clinical studies or submissions to diagnostic laboratories (1, 3, 6, 7, 8, 9, 12, 22, 23, 27). Overall, susceptibility patterns for various bacteria are similar between studies, but few studies have compared trends in susceptibility patterns over a period of several years from the same laboratory and geographical region (11, 20, 21, 26).

The purpose of this document is to present a position regarding the resistance of mastitis pathogens isolated from dairy cows to antibacterial drugs. Key questions investigated are: 1) after four decades of antibacterial drug use in dry cow and lactation therapy, does scientific data exist to demonstrate emerging antimicrobial resistance in mastitis pathogens; and, 2) does scientific data exist that supports the need for systematic change in dry cow therapy to prevent the development of resistance of mastitis pathogens against antibacterial drugs within a herd.

Evidence of Emerging Antimicrobial Resistance

Overview of Literature

Resistance of bacteria to antibacterial drugs was first reported soon after antibacterial drugs were accepted for use in both human and veterinary medicine. Articles describing antibacterial resistance patterns among bovine mastitis pathogens have been published for more than thirty years. To determine if the resistance is emerging, or progressing, ideally the resistance observed historically would be compared with that of the present. Unfortunately, the continuity between studies for this comparison is tenuous at best. Most studies have reported susceptibility data determined from the disk diffusion (Kirby-Bauer) method**.** However, differences among specific techniques used to test isolates by the disk diffusion method do not allow valid comparisons among studies. Other studies reported susceptibility data obtained from serial broth dilution or minimal inhibitory concentration (MIC) testing. Additionally, numerous studies did not offer data by bacterial species, often grouping coliforms and streptococci together. As can be seen from Table 4 (29, 31), a 30-fold difference in MIC values among species of streptococci for the same drug can exist. The proportion of resistant isolates identified by disk diffusion methods can also vary widely for the same drug when compared among streptococcal and coliform species (Tables 5 and 6). An attempt to compare results of antibacterial susceptibility among studies lacking species differentiation can lead to erroneous assumptions. Therefore, studies lacking delineation of species were not used in the assessment of emerging resistance.

Staphylococcus aureus

The most extensive antimicrobial resistance studies involving mastitis isolates have investigated *Staphylococcus aureus*. Jones, et al. (17; Table 1) noted over thirty years ago *S. aureus* isolates had relatively high MIC values for penicillin and ampicillin, and implied this was because of Beta-lactamase inactivation of the drugs. Beta-lactamase production is induced in some bacteria when exposed to Beta-lactam drugs. The importance of continued Beta-lactamase related resistance in *S. aureus* was underscored by the Watts and Salmon (40; Table 1) report of higher MIC values for isolates that produced this enzyme as compared with isolates that did not. No evidence exists to suggest that this adaptation of *S. aureus*, or resistance to other classes of antibacterial drugs, is any different from those noted thirty-five years ago. The MIC values (Tables 1 and 2) and disk diffusion results (Table 3) demonstrate ampicillin and penicillin are consistently the antimicrobial drugs to which *S. aureus* are most commonly resistant. However, comparing values within the tables from one time period to another should be avoided. Any comparison of this kind should be done with skepticism because of the differences in geography, numbers of isolates used within a study, and inconsistencies in laboratory methods. As an example, two studies performed in the same year by Costa, et al. (5) and Gentilini, et al. (14) reported the proportion of oxacillin resistant strains of *S. aureus* as 42.0 and 0%, respectively (Table 3). Interestingly, MIC values measured for isolates from the mammary gland of heifers (Table 2) do not vary from those for isolates obtained from cows, despite the fact that isolates from heifers would likely have had little previous exposure to antibacterial drugs.

Staphylococcus aureus are common causes of bacteraemia in humans (19). Among the different strains involved, the sources of major concern are the methicillin-resistant *S. aureus* (MRSA) and vancomycin resistant *S. aureus* (VRSA). MRSA strains emerged as early as 1961, and became a major concern for hospital epidemics in many countries. VRSA strain emerged in 1996 from MRSA strains a few years after introducing vancomycin in human therapy (16, 19). MRSA strains often express another resistance gene called *emr* which induces resistance to erythromycin. This gene has been identified in many isolates of poultry origin, but never from bovine or milk sources (38). Transmission of MRSA seems to be mediated by person-to-person transmission, and to the best of our knowledge, transmission of MRSA from bovine to humans has not been reported. A Belgian study identified MRSA resistance types in 1 to 3% of *S. aureus* isolates in the 1970's. However, these resistance types were not found in later years (1980's through 1990's) and they were also determined to be of human origin (9). Based on the scientific evidence available, the presence of MRSA strains originating from the use of lactating or dry cow therapy seems unlikely.

Other Mastitis Pathogens

The data for streptococci resistance to antimicrobial drugs are not as extensive as for *S. aureus*. Variability in drug resistance is more pronounced among individual streptococcal species than among studies. Again, the temptation to seek trends by comparing results from different studies should be avoided. For example, a wide disparity was observed between two studies performed in 2002 in the proportion of resistant isolates of *Str. uberis* for several drugs (11, 13). McDonald, et al. (22) reported virtually no resistance among strains of *Str. agalactiae* for Beta-lactam antibacterial drugs like penicillin and ampicillin. A small proportion of resistant strains were reported for these drugs 26 years later (11). However, little evidence has been reported to suggest this increase is significant. When coliform organisms were divided into *Klebsiella* and *Escherichia coli,* a marked variability in resistance to antibacterial drugs was observed between these genera, but there was a consistent pattern of resistance within species (Tables 6 and 7).

Studies Comparing Isolates Within a Geographical Region

A few studies have determined the proportion of resistant isolates over time performed at the same laboratory and geographic region. Antimicrobial susceptibility of *S. aureus* over a four-year period was determined for both clinical and subclinical isolates from British dairy cows (20). No trends over time in the proportion of resistant strains for clinical or subclinical isolates were reported. A survey of the prevalence of mastitis and antimicrobial resistance of intramammary pathogens was carried out in 1988 (17,111 quarter milk samples) and again in 1995 (10,410 quarter milk samples) in Finland (25). The proportion of isolates resistant to at least one antibacterial increased from 32 to 52% during 1988 to 1995. Of the antibacterial drugs tested on the panel, appreciable increased resistance was only observed for penicillin. Studies from Michigan (11) and Wisconsin (21) surveyed the antibacterial susceptibility results from all milk sample submissions to the respective diagnostic laboratories for the years 1994 to 2001. A trend for an increase in the proportion of *S. aureus* isolates that were resistant to any antibacterial drugs was not observed in either study. In contrast, the proportion of resistance isolates actually decreased for some of the Beta-lactam drugs. Thus, three of the four studies did not determine any appreciable difference in mastitis pathogen resistance for *S. aureus*, and of the study that did, it was primarily for Beta-lactamase susceptible drugs. In these four studies, the proportion of resistant streptococci or coliforms did not change over the duration of the studies.

The largest study of this type included a 25-year perspective of *S. aureus* mastitis isolates in Belgium (9). Percentages of strains producing penicillinase were noted to rapidly increase from 38% in 1971 to between 70 to 80 % throughout the next two decades. However, after the rapid increase in penicillinase resistance observed initially, the percentage of resistant strains has attained a plateau. In 1996 the percentage of resistant strains was reported to be 51% (9). Developing resistance was not observed for other antibacterial drug classes, including Beta-lactamase stable drugs such as the cephalosporins (9).

Assessment of Literature

Definitive conclusions on resistance patterns of mastitis pathogens are difficult given the limitations of the data presented in the literature. Logically, antibacterial resistance can be induced in most mastitis pathogens. However, as stated earlier, there is no evidence that antibacterial resistance is an emerging process in mastitis pathogens. Comparisons should not be made between studies, except to gain an overall appreciation of which classes of drugs are more active than others. Apparently the breakpoints used to determine drug susceptibility were not identical for all studies for the past thirty-five years. Most of the studies, including those completed over a time period, report data from essentially unknown conditions with respect to farms, i.e., the number of samples from each farm, and most importantly of all, prior drug administration history of the cow that was sampled for bacterial isolation. If we are to infer developing resistance in relation to antibacterial drug therapy, the history of the drug administration, including dose, frequency, and duration must be known. Scientific evidence would also require an antibiogram before and after drug administration. Because of the quantitative nature of the data, MIC values would be preferred, with a histogram of all isolates displayed rather than just MIC values at the 50^{th} and 90^{th} percentile.

Need for Systematic Change in Dry Cow Therapy

Properly controlled studies have not examined the need for systematic change in dry cow therapy to prevent the development of resistance of mastitis pathogens against antibacterial drugs within a herd. Therefore, no data are evident to support such a change based on increased antibacterial drug resistance in mastitis pathogens. Berghash, et al. (2) reported that MIC levels for *Str. agalactiae* isolates were higher for herds that reported dry cow treating all cows, as opposed to herds that did not dry cow treat or only selectively treated cows. This study reiterated that strains of *Str. agalactiae* exist that may be potentially resistant to Beta-lactam antibacterial drugs. However, the drug therapy records and exposure of cows in these herds was unknown, other than the survey of dry cow therapy use. Interestingly, the MIC_{90} for penicillin of *Str. agalactiae* strains from the herds that did not use total dry cow treatment, although lower than the other herds, was still above the breakpoint that is considered to determine susceptibility.

Conclusion

Scientific evidence does not support a widespread, emerging resistance among mastitis pathogens to antibacterial drugs. Although resistance to antibacterial drugs among mastitis pathogens has been well documented for nearly four decades, evidence has not been presented to suggest that this is either an emerging or progressing phenomenon. Controlled studies have not determined, on a pharmacodynamic basis, which drug therapeutic regimens may increase this risk, or for that matter, help to decrease it. Monitoring should be continued, preferably by studies that follow data over a course of time and not one point in time. Data derived for MIC testing would be preferred for this purpose because of a more consistent and quantitative nature compared with the disk diffusion method.

References

1. Anderson, K.L. 1989. Therapy for acute coliform mastitis. 1989. Compend. Contin. Educ Pract. Vet. 11:1125-1132.

2. Berghash, S.R., J.N. Davidson, J.C. Armstrong, and G.M. Dunny. 1983. Effects of antibiotic treatment of nonlactating dairy cows on antibiotic resistance patterns of bovine mastitis pathogens. Antimicrob. Agents Chemother. 24:771-776.

3. Brown, M.B., and A.E. Scasserra. 1990. Antimicrobial resistance in streptococcal species isolated from bovine mammary glands. Am. J. Vet. Res. 51:2015-2018.

4. Constable, P.D., and D.E. Morin. 2002. Use of antimicrobial susceptibility testing of bacterial pathogens isolated from the milk of dairy cows with clinical mastitis to predict response to treatment with cephapirin and oxytetracycline. JAVMA 221:103-108.

5. Costa, E.O., N.R. Benites, J.L. Guerra, et al. 2000. Antimicrobial susceptibility of *Staphylococcus* spp. Isolated from mammary parenchymas of slaughtered dairy cows. J. Vet Med B. Infect Dis Vet Public Health 47:99-103.

6. Craven, N., J.C. Anderson, and T.O. Jones.1986. Antimicrobial drug susceptibility of *Staphylococcus aureus* isolated from bovine mastitis. Vet. Rec. 118:290-291.

7. Davidson, J.N. 1980. Antibiotic resistance patterns of bovine mastitis pathogens. Proc 19th Annual Meeting Natl. Mastitis Council, p.181.

8. De Oliveira, A.P., J.L. Watts, S.A. Salmon, and F.M. Aarestrup. 2000. Antimicrobial susceptibility of *Staphylococcus aureus* isolated from bovine mastitis in Europe and the United States. J. Dairy Sci. 83:855-862.

9. Devriese, L.A., F. Haesebrouck, J. Hommez, et al. 1997. A 25-year survey of antibiotic susceptibility testing in *Staphylococcus aureus* from bovine mastitis in Belgium, with special reference to penicillinase. Vlaams Diergeneeskundig Tijdschrift 66:170-173.

10. Erskine, R.J. 1996. Why do antibiotic residues in milk happen? Michigan Dairy Review 1:16.

11. Erskine, R.J., R.D. Walker, C.A. Bolin, et al. 2002. Trends in Antibacterial Susceptibility of Mastitis Pathogens During a Seven-Year Period. J. Dairy Sci. 85:1111-1118.

12. W. Fang and S. Pyorala. Mastitis-causing *Escherichia coli*: serum sensitivity and susceptibility to selected antibacterials in milk. J. Dairy Sci. 1996 79: 76-82.

13. Franklin, A. and M. Horn af Rantzien. 1983. Antimicrobial drug susceptibility of *Staphylococcus aureus* strains isolated from bovine milk. Nord. Vet. Med. 35:460-464.

14. Gentilini, E., G. Denamiel, P. Liorente, et al. 2000. Antimicrobial susceptibility of *Staphylococcus aureus* isolated from bovine mastitis in Argentina. J. Dairy Sci. 83:1224-1227.

15. Griggs, D.J., M.C. Hall, Y.F. Jin, and L.J.V. Piddock. 1994. Quinolone resistance in veterinary isolates of *Salmonella.* J. Antimicrob. Chemother. 33:1173-1189.

16. Hiramatsu, K., L. Cui, M. Kuroda, and T. Ito. 2001. The emergence and evolution of methicillin-resistant *Staphylococcus aureus*. Trends in Microbiol. 9:486-493.

17. Jones, A., T.M. Higgs, F.K. Neave, et al. 1967. The sensitivity of bovine staphylococci, streptococci and corynebacterium to cloxacillin and various other antibiotics. J. Dairy Res. 34:249-255.

18. Kaneene, J.B., R. Miller. 1992. Description and evaluation of the influence of veterinary presence on the use of antibiotics and sulfonamides in dairy herds. J. Am. Vet. Med. Assoc. 201:68-76.

19. Livermore, D.M. 2000. Antibiotic resistance in staphylococci. Int. J. Antimicrobial Agents. 16:S3-S10.

20. Mackie, D.P., E.F. Logan, D.A. Pollack, and S.P. Rodgers. 1988. Antibiotic sensitivity of bovine staphylococcal and coliform mastitis isolates over four years. Vet. Rec. 123:515-517.

21. Makovec, J.A., and P.L. Ruegg. 2003. Antimicrobial resistance of bacteria isolated from dairy cow milk samples submitted for bacterial culture: 8,905 samples (1994-2001). JAVMA 222:1582-1589.

22. McDonald, J.S., T.J. McDonald, and D.R. Stark. 1976. Antibiograms of streptococci isolated from bovine mammary infections. Am. J. Vet. Res. 37:1185-1187.

23. McDonald, J.S., T.J. McDonald, and D.R. Stark. 1977. Antibiotic sensitivity of aerobic Gram-negative rods isolated from bovine udder infections. Am. J. Vet. Res. 38:1503-1508.

24. Moore, G.A. and L.E. Heider. 1984. Treatment of mastitis. Vet. Clin. North Am. Large Anim. Practice. 6:323-331.

25. Myllys, V., T. Honkanen-Buzalski, P. Huovinen, M. Sandholm, and E. Nurmi. 1994. Association of changes in the bacterial ecology of bovine mastitis with changes in the use of milking machines and antibacterial drugs. Acta Vet. Scand. 35:363-369.

26. Myllys, V., K. Asplund, E. Brofeldt, et al. 1998. Bovine mastitis in Finland in 1988 and 1995- Changes in prevalence and antimicrobial resistance. Acta Vet. Scand. 39:119-126.

27. Owens, W.E., and J.L. Watts. 1988. Antimicrobial susceptibility and beta-lactamase testing of staphylococci isolated from dairy herds. J. Dairy Sci. 71:1934-1939.

28. Owens W.E., J.L. Watts, B.B. Greene, et al. 1990. Minimum inhibitory concentrations and disk diffusion zone diameter for selected antibiotics against streptococci isolated from bovine intramammary infections. J. Dairy Sci. 73:1225-1231.

29. Owens W.E., C.H. Ray, J.L. Watts, et al. 1997. Comparison of success of antibiotic therapy during lactation and results of antimicrobial susceptibility tests for bovine mastitis. J. Dairy Sci. 80:313-317.

30. Piddock, L.J.V. 1996. Does the use of antimicrobial agents in veterinary medicine and animal husbandry select antibiotic-resistant bacteria that infect man and compromise antimicrobial therapy. J. Antimicro. Chemother. 38:1-3.

31. Rossitto P.V., L. Ruiz, Y. Kikuchi, et al. 2002. Antibiotic susceptibility patterns for environmental streptococci isolated from bovine mastitis in central California dairies. J. Dairy Sci. 85:132-138.

32. S. A. Salmon, J. L. Watts, F. M. Aarestrup, J. W. Pankey, and R. J. Yancey, Jr. Minimum inhibitory concentrations for selected antimicrobial agents against organisms isolated from the mammary glands of dairy heifers in New Zealand and Denmark J. Dairy Sci. 1998 81: 570-578.

33. Schaeren, W., and M. Schaellibaum 1987. Drug resistance of the principal pathogens of bovine mastitis. Schweiz. Milchw. Forschung 16:69-70.

34. Schaeren, W., and M. Schaellibaum 1993. Bestimmung der minimalen Hemmkonzentrationen bei Mastitiserregern. Report of the Federal Dairy Research Station 18: 1-31.

35. Schifferli, D., Schaellibaum, M., J. Nicolet 1984. Bestimmung der Minimalhemmkonzemtration bei Mastitiserregern beim Rind. Schweiz. Arch. Tierheilk. 126: 23-34.

36. Sears, P.M. 1986. Antibiogram information collected by the NYSMCP for the period of 02/75-07/85. Quality Milk Promotion Services Newsletter, Volume 1, No. 3, Winter, 1985-1986.

37. Singh, M., M.A. Chaudry, J.N Yadava, and S.C. Sanyal. 1992. The spectrum of antibiotic resistance in human and veterinary isolates of *Escherichia coli* from 1984-1986 in northern India. J. Antimicrob. Chemother. 29:159-168.

38. Teuber, M. 2001. Veterinary use and antibiotic resistance. Current Opinion in Microbiology. 4:493-499.

39. Watts J.L., S.A. Salmon, R.J. Yancey, Jr, et al.. 1995. Antimicrobial susceptibility of microorganism isolated from the mammary glands of dairy heifers. J. Dairy Sci. 78:1637-1648.

40. Watts J.L., and S.A. Salmon. 1997. Activity of selected antimicrobial agents against strains of *Staphylococcus aureus* isolated from bovine intramammary infections that produce Beta-lactamase. J. Dairy Sci. 80:788-791.

41. White, D.G. 1999. Use and misuse of antimicrobials in veterinary medicine. Proc 38th Annual Meeting Natl. Mastitis Council, p.9. Table 1- Summary of studies using MIC data for *S. aureus.*

Table 1. Summary of MIC data for *S. aureus* from dairy cows
MIC_{90} (µg/ml)

	Ref (29) 1997	Ref (14) 2000	Ref (17) 1967	Ref (13) 1983	Ref (8) 2000	Ref (6) 1986	Ref (40) *β-lact* (+) 1997	Ref (40) *β-lact* (-) 1997	Ref (35) 1984	Ref (33) 1987	Ref (34) 1993
Ampicillin	---	---	---	0.5	1.0	---	4.0	0.5	---	---	---
Ceftiofur	2.0	---	---	---	1.0	---	1.0	1.0	---	---	---
Cephalothin*	0.25	0.75	---	---	1.0	---	0.5	0.25	---	---	1.21
Cloxacillin	0.5	---	0.07	---		0.5	---	---	0.25	0.40	0.22
Erythromycin**	0.5	0.75	---	0.5	.5	0.5	64.0	0.5	3.28	3.46	3.21
Neomycin	---	---	---	8.0	2.0	1.0	---	---	0.41	0.55	0.39
Novobiocin	0.5	---	1.25	---		0.5	---	---	---	---	---
Oxacillin	---	0.5	---	0.25	1.0	---	1.0	0.25	---	---	---
Penicillin	0.125	1.5	> 100	0.25	.5	---	16.0	0.25	(47.8%)#	(33.3%)#	(10.1%)#
Pirlimycin	1.0	---	---	---	1.0	---	64.0	0.5	---	---	---
Streptomycin	---	---	5.0	8.0		4.0	---	---	---	---	---
Tetracycline	---	---	0.6	2.0		0.5	---	---	---	---	---

* Or cephapirin /cefoperazone

** or spiramicin

Beta-lactamase positive

Table 2. Summary of MIC data for *S. aureus* from heifers

MIC_{90} (µg/ml)

	Ref (32) 1998 Denmark	Ref (32) 1998 New Zealand	Ref (39) 1995
Ampicillin	0.5	---	---
Ceftiofur	1.0	2.0	1.0
Cephalothin	0.5	0.5	0.5
Cloxacillin	---	0.5	0.5
Erythromycin	0.5	0.5	0.5
Novobiocin	---	1.0	0.5
Oxacillin	0.5	---	---
Penicillin	---	---	0.13
Pirlimycin	0.5	1.0	0.5
Enrofloxacin	---	0.25	0.5
Tetracycline	---	---	0.6

Table 3. Summary of studies using disk diffusion for *S. aureus*

% of Isolates Resistant

	Ref (27)	Ref (36)	Ref (9)	Ref (9)	Ref (9)	Ref (5)	Ref (14)	Ref (25)		Ref (21)	Ref (11)
	1988	1986	1971	1982-83	1996	2000	2000	1988	1995	1994-2001	1994-2000
Ampicillin	7.0	54.0				83.0	---	---	---	34.9	49.6
Ceftiofur	---	---				---	---	---	---	---	0.2
Cephalothin*	0	1.0	3.5**	0**	0**	0	0	0.3	2.0	0.1	0.2
Cloxacillin	---	20.0				---	---	---	---	6.8	---
Erythromycin	0.3	6.0	3***	9***	12***	17.0	11.6	4.1	2.6	14.9	
Gentamicin	0	1.0				---	3.4	---	---	---	1.1
Novobiocin	0	7.0	0	3	1	---	---	---	---	21.8	---
Oxacillin	0	---				42.0	0	1.5	0	---	0.6
Penicillin	7.0	57.0	38**	75**	51**	75.0	40.0	31.8	50.7	32.6	40.6
Pirlimycin	---	---				---	7.7	---		13.6	2.1
Streptomycin	6.0	36.0	26	---	---	---	---	---	---	---	---
Tetracycline	0	8.0	21	8	9	17.0	---	7.0	11.7	22.6	8.5

* Or cephapirin

** Represents resistance as reported for all beta-lactamase-labile (penicillin and ampicillin) and stable (cloxacillin and cephalosporins) antibacterial drugs

*** Represents resistance reported for all macrolide drugs

Table 4. Summary of studies using MIC data for Streptococci MIC_{90} ϕ

	Ref (3) 1990	Ref (29) 1997	Ref (31) 2002	Ref (32) 1998 Ψ	Ref (35) 1984	Ref (33) 1987	Ref (34) 1993
Ampicillin	---	**---**	**0.5,** (0.6)	**2.0**	---	---	---
Ceftiofur	---	**0.5,** (1.0)	**2.0,** (0.25)	**1.0**	---	---	---
Cephalothin*	**---**	**0.25,** (0.25)	**1.0,** (1.0)	**8.0**	---	---	**0.58**
Cloxacillin	---	**4.0,** (0.5)	---	---	**0.25**	**0.87**	**0.45**
Erythromycin**	**≥ 256,** (4), 2	**1.0,** (0.25)	**8.0,** (0.12)	**4.0**	**0.27**	**0.16**	**0.34**
Gentamicin	**---**	**---**	---	**---**	**---**	**3.13**	**3.33**
Novobiocin	---	**4.0,** (2.0)	---	---	**---**	**---**	**---**
Oxacillin	**---**	---	**1.0,** (1.0)	**16.0**	**---**	**---**	**---**
Penicillin	**≤ 1,** (≤ 1), ≤1	**.125,** ≤ (0.06)	**0.25,** (0.6)	**2.0**	**0.07**	**0.25**	**0.11**
Pirlimycin	---	---	**8.0,** (0.25)	**4.0**	**---**	**---**	**---**
Tetracycline	**32,** (8), 0	**---**	**16.0,** (16.0)	**---**			
Chloramphenicol	---	---	---	---	**3.27**	**2.50**	**3.30**

ϕ Values listed are for ***Str. uberis*****,** (*Str. dysgalactiae)*, and *Str. agalactiae*, respectively

* Or cephapirin/cefoperazone

** Or spiramycin

Ψ Isolates collected from mammary glands from heifers in Denmark

Table 5. Summary of studies using disk diffusion data for Streptococci

% Resistant ϕ

	Ref (22) 1976	Ref (28) 1990	Ref (31) 2002	Ref (11) 2002
Ampicillin	---	**0,** (2)	**7.5,** (0)	**2.1,** (0.8), 2.6
Ceftiofur	---	---	**6.8,** (0.7)	**0,** (0), 0
Cephalothin*	**0,** (0), 0	**1,** (0)	**2.8,** (0)	**0.2,** (0.3), 0
Cloxacillin	**2,** (2), 0	---	---	---
Erythromycin	**3,** (0), 0	**8,** (1)	**6.6,** (6.6)	**31.9,** (18.0), 15.4
Gentamicin	**87,** (22), 100	**47,** (3)	---	**34.2,** (3.2), 76.9
Novobiocin	---	**6,** (3)	---	---
Oxacillin	**2,** (0), 0	---	**1.3,** (1.3)	**41.7,** (1.9), 3.8
Penicillin	**0,** (0), 0	**3,** (2)	**49.6,** (2.0)	**5.5,** (5.5), 3.9
Pirlimycin	---	---	**39.1,** (7.9)	**20.1,** (11), 7.1
Streptomycin	**100,** (100), 100	**62,** (26)	---	---
Tetracycline	**2,** (63), 0	**24,** (9)	**72.9,** (1.7)	**45.2,** (60.2), 46.2

ϕ Values listed are for ***Str. uberis*,** (*Str. dysgalactiae*), and *Str. agalactiae*, respectively

* Or cephapirin

Table 6. Summary of studies using MIC data for coliforms

MIC_{90}

	Ref (35) 1984	Ref (33) 1987	Ref (34) 1993
Ampicillin	2.26	3.22	2.42
Cefoperazon	---	---	0.48
Polymycin B/E	1.53	1.88	1.01
Neomycin	1.15	1.11	0.45
Gentamicin	0.63	0.52	0.45
Chloramphenicol	5.63	5.33	2.58
Cotrimoxazole	1.42	2.87	0.64
Streptomycin	3.51	---	---
Tetracycline	1.81	---	---
Norfloxacine	---	---	$\leq$ 0.07

Table 7. Summary of studies using disk diffusion data for coliforms

% Resistant ϕ

	Ref (23) 1977	Ref (36) 1986	Ref (11) 2002	Ref (21) 2003
Ampicillin	---	**34.0** (90.0)	**15.7** (98.5)	**21.9** (89.1)
Ceftiofur	---	---	**4.6** (14.1)	**---**
Cephalothin*	**40.0** (20.0)	**21.0** (23.0)	**25.5** (4.2)	**27.9** (12.1)
Cloxacillin	**---**	**97.0** (98.0)	---	**99.4** (99.1)
Erythromycin	**---**	**91.0** (97.0)	**---**	**99.2 (**99.0)
Gentamicin	**0** (1.0)	**2.0** (2.0)	**2.0** (0)	**---**
Novobiocin	---	**96.0** (90.0)	---	---
Penicillin	**98.0** (99.0)	**98.0** (99.0)	**---**	**99.6** (99.5)
Streptomycin	**30.0** (23.0)	**46.0** (52.0)	---	---
Tetracycline	**85.0** (45.0)	**33.0** (35.0)	**33.2** (33.0)	**37.4** (30.0)

ϕ Values listed are for ***E. coli*** and (*Klebsiella)*, respectively

* Or cephapirin