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Prevalence and incidence of subclinical mastitis in dairy ewes and goats

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ABSTRACT

The prevalence and incidence of bacterial mastitis have not been determined for small milking ruminants in Vermont, USA. Milk samples were collected for bacteriology from each gland of 110 goats (6 herds) and 153 sheep (3 herds) within 24 hours of parturition and approximately 40 days later. The prevalence, incidence rate of new infection and the spontaneous cure rate were determined. Goats had a significantly higher prevalence of infection than sheep ($P<0.05$). Prevalance at parturition and 40 days later was 27.3% and 25.5% and 15.0% and 9.1% for goats and sheep, respectively. Prevalence of mastitis increased with age in goats ($P = 0.08$) but not sheep and differed among herds ($P<0.05$). It is concluded that there is variation among species and herds in the prevalence and incidence of mastitis.

Keywords: mastitis; goats; sheep.

INTRODUCTION

Mastitis control depends on minimising new infections and reducing the duration of existing infections (Bramley and Dodd, 1984). Understanding the rate of new infections, the duration of existing infections and the spontaneous cure rate would enable research to focus on periods of high risk of new intramammary infections (IMI) and enhance development of management practices that may reduce losses associated with mastitis.

Coagulase negative staphylococci (CNS) are the most prevalent pathogens of the mammary gland of sheep and goats (Poutrel and Lerondelle, 1983; Maisi *et al.*, 1987; de la Cruz *et al.*, 1994; Fthenakis, 1994; Contreras *et al.*, 1995; Kirk *et al.*, 1996; Burriel, 1997). In cattle, CNS were shown to be contagious and to be controlled by use of postmilking teat disinfection (Lam *et al.*, 1996). However, the dynamics of CNS mastitis in goats and sheep are not well documented. Many studies have defined the number of infected glands at one point of lactation, that is the prevalence of infection. Prevalence of mastitis is dependant on the rate of new intramammary infections (IMI; i.e. the incidence of new infections; Thrusfield, 1996) and the duration of existing infections, as effected by the spontaneous cure rate and cure rate following treatment (Lam *et al.*, 1996). However, few studies have quantified the incidence rate of new IMI or the spontaneous cure rate in small ruminants.

The aim of the present study was to determine the prevalence of IMI at parturition and 40 days later and to quantify the incidence of new IMI and the spontaneous cure rate over this period.

MATERIALS AND METHODS

Goats (n = 110) and sheep (n = 155) from 6 and 3 herds and flocks, in Vermont, USA, were enrolled in the study. All enrolled animals underwent parturition between February and April 1999. Goat herd size ranged from 15 to 180 and between 8.8% and 73.3% of does within herds were enrolled. For the sheep, the herd size ranged from 130 to 145 and between 35.2% and 41.2% of ewes were enrolled.

All animals were housed over winter. The sheep flocks lambed seasonally with all lambings occurring between March and May. Lambs were left on the ewes for approximately 4 weeks after lambing until weaning in April or May. The ewes were then machine milked twice daily. All goat herds had multiple kidding periods throughout the year, with some goats in milk at all times. The goat herds remained housed throughout the study. The kids were removed within 24 hours of kidding and does were then machine milked twice daily.

All sheep herds and five of the six goat herds machine milked the animals. The goats in herd 'A' were hand milked. Teats were prepared before milking by spraying or dipping with disinfectant followed by drying with individual towels in 6 of the 9 herds or flocks, with 2 herds using iodine-soaked towels to prepare udders before milking and one doing no pre-milking teat preparation (Table 2). An iodine-based teat disinfectant was applied in all herds or flocks following milking.

Herdowners were asked to collect milk from both glands within 24 hours of kidding or lambing. Herdowners were given sterile tubes, alcohol and swabs and instructed in collection of milk samples aseptically. Briefly, the teat-end was prepared for milking by the herdowner following the normal herd protocol, then scrubbed with a cotton wool pledget wetted with 70% alcohol. After the teat end had dried, the first strip of milk was discarded and then approximately 10 ml of milk was expressed into a sterile test tube, the tube was capped and labelled with herd, animal's number or name and gland and then placed on ice. The samples were frozen at –20 °C for up to 1 month before culture.

On one day for each herd, approximately 6 weeks (average = 40.5 ± 8.1 days, mean ± standard deviation) after parturition, a milk sample was drawn from each gland of each enrolled animal for bacterial culture. Milk samples were transported to the laboratory on ice and refrigerated at 4 °C.

Calibrated, disposable inoculating loops (Difco Laboratories, Detroit, MI) were used to streak 0.01 ml of milk onto a gland plate of 5% trypticase washed-sheep-blood agar (TBA; Micro-Diagnostics, Inc., Lombard, IL). Plates were incubated at 37°C for 48 h. The colonies were provisionally speciated as *Staphylococcus aureus* (SA),

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CNS, other streptococci, Coliforms (CO) or *Corynebacterium bovis* (Cb) on morphology and haemolysis pattern. Where >4 colony forming units (CFU's) per plate of a single isolate were found, representative colonies were subcultured on TBA. Gram's stain were performed on all significant colonies (i.e., > 5 CFU). Gram-positive cocci isolates were tested for catalase and coagulase production and Gram-negative colonies were further analysed with MIO medium (motility, indole, ornithine; Difco Laboratories, Detroit MI). An IMI was diagnosed when > 4 CFU/plate of each of 1 or 2 colony types were isolated. Samples from which >2 colony types were isolated or where <5 colonies of any pathogen were isolated were regarded as contaminated or as having 'minor' infections, respectively. A new IMI was diagnosed when a pathogen was isolated from a previously uninfected gland. Spontaneous cure was defined as having occurred when a pathogen isolated at the first sample was not isolated at the second sampling. Prevalence was defined as the number of glands (or animals) from which a pathogen was isolated divided by the total number of glands (or animals) sampled at each time period. The incidence of new IMI was calculated as the total number of new IMI divided by the total number of days between samplings for glands with no infection at the initial sampling.

The effect of age (coded as 1 + 2, 3 + 4 or > 4 years), herd, species, day of year at parturition (Julian date) and days postpartum at second sampling on the prevalence at parturition and 40 days later was examined by logistic regression. As herd and species were confounded, two sets of models were run, one with herd and the other variables and one with species and the other variables. Forward stepwise analysis was used with log likelihood as the selection methodology.

RESULTS

More goats than sheep were infected at parturition and at 40 days postpartum (P < 0.05; Table 1). No change was determined in the total number of goats infected at parturition compared to 40 days postpartum (P>0.3). The number of sheep with infection tended to decline with time postpartum (P = 0.12) and the number of glands of sheep infected declined significantly postpartum (P < 0.01; Table 1).

TABLE 1: The percentage of animals with 0, 1 or 2 glands with infections at parturition and 40 days later

	Goats		Sheep	
	0¥	40¥	0¥	40¥
no. animals	110	110	153	132
No growth (% animals)	50.0	66.4	52.9	69.7
1 gland (% animals)	19.1	15.5	11.1	9.1
2 glands (% animals)	8.2	10.0	3.9	0.0
% animals infected	27.3	25.5	15.0	9.1
% glands infected	35.5	35.5	19.0	9.1
Cs + minors* (% animals)	22.7	9.1	32.0	21.2

¥ Days postpartum

* Contaminated sample (>2 colony types) or minor infection (i.e. < 5 CFU/plate)

Herds differed significantly in prevalence of IMI at parturition (P<0.005) and 40 days later (P < 0.005; Table 2). The number of infected glands increased with age group in goats (6.8%, 19.4% and 31.6% of glands from 1 + 2, 3 + 4 and > 4 year old animals; P < 0.05) but not sheep (4.7%, 4.3% and 0% of glands from 1 + 2, 3 + 4 and > 4 year old animals, respectively).

TABLE 2: The prevalence and incidence (within herd or flock) of glands with positive bacterial culture at calving and 40 days postpartum

				Prevalence		Incidence		
				Day 0	Day 40			
Herd	Spp.	Teat prep*.	No. glands sampled	% infected	% infected	No. new glands	Total years	Cases/ year at risk
A	Goat	Dip	22	22.7	36.4	2	0.82	2.44
B	Goat	Spray	40	42.5	40.0	1	2.02	0.50
C	Goat	Spray	22	0.0	0.0	0	2.81	0.00
D	Goat	None	66	6.1	4.5	1	6.55	0.15
E	Goat	Towel	42	19.0	7.1	0	0.53	0.00
F	Goat	Dip	32	18.8	28.1	3	1.78	1.69
G	Sheep	Towel	108	8.3	4.6	3	4.76	0.63
H	Sheep	Towel	102	3.9	1.0	1	8.75	0.11
I	Sheep	Towel	100	15.0	6.0	3	2.86	1.05

* premilking teat preparation method

The proportion of infected animals at 40 days postpartum was affected by herd (P < 0.05) and age and species (P < 0.05; with herd removed from the analysis) but not by parturition date or days postpartum at second sampling.

A total of 14 new IMI in 14 separate animals were detected, 7 each in sheep and goats. Incidence was 0.92 cases/animal/year or 0.44 cases/gland/year. Goats and sheep did not differ in incidence rate (0.47 and 0.42 cases/gland/year for goats and sheep, respectively). Between zero and three new IMI were detected in each herd with the gland incidence rate varying among herds from 0 to 2.4 cases/gland/year (Table 2). Animals in which one gland was infected at parturition were more likely to have the second gland infected at 40 days postpartum than animals which did not have a pathogen in either gland at parturition (5/15 vs. 7/106; Odds Ratio = 7.6, P < 0.01).

Spontaneous cure occurred in 26 of the 38 (68.4%) glands infected at parturition. Goats had a lower self-cure rate than sheep (11/22 (50.0 %) vs. 15/16 (93.8 %), respectively, Odds Ratio = 0.06; P < 0.05).

Coagulase negative staphylococci were the most common isolates from both goats and sheep and at parturition and 40 days later (Table 3).

TABLE 3: Bacterial species isolated from glands of goats and sheep at parturition and 40 days later

	Goats				Sheep			
	Day 0		Day 40		Day 0		Day 40	
	n	%	n	%	n	%	n	%
Corynebacterium spp.	2	5.0	3	7.7				
Staphylococcus aureus	2	5.0	4	10.3	4	33.3	2	7.1
CNS*	35	87.5	26	66.7	8	66.7	22	78.6
Others¥	1	2.5	6	15.6	0	0.0	4	14.4
Total	40		39		12		28	

* Coagulase negative staphylococci

¥ Including Coliforms (n = 1), *Enterococcus faecalis* (n = 2), *Lactococcus garvieae* (n = 1), *Pasteurella sp.* (n = 1), *Pseudomonas aeruginosa* (n = 1), *Stomatococcus mucilaginosus* (n = 1), *Streptococcus gallolyticus* (n = 1), *Other streptococcoci* (n = 1).

DISCUSSION

The prevalence of glands with IMI was 35.5% and 19.0% in goats and sheep at parturition. Prevalence of gland infections in goats ranged from 9% to 65% in earlier studies (reviewed by Contreras *et al.*, 1995). Prevalence of bacterial infection of ewe udder glands is reported to range between 4% and 26% (Fthenakis, 1994; de la Cruz *et al.*, 1994). This range of prevalence among studies and herds may reflect differences in bacteriological techniques, time of sampling within a lactation, milking management (e.g., hand milked vs. machine milked, teat spraying, etc.), physical environment (housing vs. pastured), mastitis management programmes, nutrition, milk production or antibiotic use among herds.

Prevalence declined in sheep but not goats by days postpartum. A decline in prevalence with time postpartum was reported in ewes (Fthenakis, 1994). Differences in management between the ewes and does included time of weaning, seasonality of parturition and differences among herdowners in management practices. All goats in the present trial were machine or hand milked from within 24 hours after parturition, whereas the lambs were left on the ewes until about 30 to 35 days postpartum. The sheep herds were all seasonal lambing and all used pasture grazing for more than 50% of the year. The goats were all housed for the majority of the year and kidded year round. The housing of the goats may have been more heavily contaminated than that of the sheep due to the continual use of the facilities. Additionally, as there were does lactating at all times of the year, the potential for ongoing transmission of infection from late to early lactation goats during milking or in the barns was always present. In contrast, all sheep were non-lactating at the same time for some months, which may have reduced the potential for ewe to ewe transmission of pathogens.

Prevalence of IMI increased with age in goats in agreement with other studies (Sanchez *et al.*, 1999). Increasing prevalence with age may be due to increased length of exposure to pathogens in older compared to younger animals. The higher prevalence of IMI in older animals may also be explained by the long duration of infection with CNS, the most common pathogen in small ruminants (Maisi and Riipinen, 1991; Contreras *et al.*, 1995), combined with the spontaneous cure rate being lower than the incidence of new IMI. Hence, the prevalence of IMI will increase with time. Prevalence in ewes did not increase with age as has been previously reported (Fthenakis, 1994). The high spontaneous cure rate found in the ewes in the present trial may be the explanation for this.

Incidence of new bacterial IMI was similar for goats and sheep in the present study and averaged 0.92 cases/gland/year. This is similar to the incidence of 1%/ewe/week for ewes pre-weaning reported by Hueston *et al.* (1986). Incidence of new IMI does not appear to have been reported for dairy goats. Incidence of IMI for CNS in housed dairy cattle is reported as 1.58 cases/1000 quarter days or about 0.5 cases/quarter/year (Lam, 1996). The goats and sheep appear to have a higher incidence rate than that reported for cattle. However, incidence in the present study was calculated only over the first 40 days of lactation. Incidence of new clinical infections was shown to be higher in early, compared to late, lactation in cattle (Hogan *et al.*, 1989; Lam, 1996). Hueston *et al*, (1986) noted an increase in incidence rate of new IMI following weaning from 1%/ewe/week preweaning to 9.7%/ewe/week post-weaning. In that study ewes were not machine-milked following weaning, so the increased incidence of IMI was associated with involution of the mammary gland.

In the present study, 15 of 16 sheep with pathogens at lambing underwent spontaneous cure compare to only 50% of goats. Spontaneous cure in sheep was reported as 9.5% and 21% between 2 and 8 weeks post-weaning and lambing to 6 weeks postpartum (Fthenakis, 1994; Kirk *et al.*, 1996). The high frequency of suckling and the large litter size (average of > 2 lambs/ewe) may have resulted in the ewes being frequently milked dry which may have resulted in the high spontaneous cure rate observed.

Prevalence of IMI is dependent on the duration of infection, the incidence of new IMI and the spontaneous cure rate (Lam, 1996). The higher prevalence of IMI at 40 days postpartum in sheep relative to goats may be explained by the higher prevalence at parturition in goats and the lower spontaneous cure rate over the days prepartum.

It is concluded that there are significant differences in the epidemiology of mastitis between goats and sheep and that different mastitis control measures may be required for the two species

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