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# Individual animal risk factors for clinical mastitis in meat sheep in Norway

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#### Abstract

An *m:n* matched case–control study was conducted to identify risk factors for ovine clinical mastitis (CM). Data were from a national sheep registry and only ewes that lambed in the spring of 2004 were included. Eligible cases (n = 2857) and controls (n = 76,716) from 1056 flocks of meat sheep were matched on flock and conditional logistic regression was used for analysis of the data. CM risk was associated with age of the ewe and whether or not assistance at lambing was needed owing to dystocia; however, the effects of both these factors were modified by the number of lambs born. In ewes with 1 lamb, increasing age was associated with increased odds of CM (OR = 1.2 for each 1-year increase), while only a slight numerical increase in the odds was observed in ewes with >1 lamb. Dystocia was associated with increased odds of CM in ewes with 1 lamb (OR = 1.7) or 2 lambs (OR = 1.4), while no association was observed in ewes with >2 lambs. The odds of CM increased markedly with increasing number of lambs born to the ewe. For example, odds for 2-year-old ewes without dystocia were 6.7 times greater for those with >3 lambs than for those with 1 lamb. Compared with ewes of old Norwegian breeds, ewes of other breeds were more likely to experience CM (OR = 1.7). Ewes treated for CM at least once during the preceding 3 years had 4.0 times greater odds of CM compared with ewes without a CM history. It is likely that the effect estimates from this study, which are adjusted for breed and unaffected by inter-flock variations, are valid also for other meat sheep populations.

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Keywords: Ewe; Clinical mastitis; Risk factors; Case-control study

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# 1. Introduction

Mastitis is the cause of great economic losses in sheep production worldwide. Surveys in various populations have demonstrated high prevalences of subclinical intramammary infections, ranging from 12% (Watkins et al., 1991) to 29% (Kirk et al., 1996) on the individual animal level. Annual incidences of clinical mastitis (CM) around 2% seem to be common (Quinlivan, 1968; Lafi et al., 1998); however, an incidence as high as 7% has been reported (Larsgard and Vaabenoe, 1993). Severe clinical cases are not uncommon and a fatality rate of 4% for ewes with acute CM has been found (Onnasch et al., 2002). Acute CM is a painful condition; thus, ovine mastitis is also an important animal welfare issue.

While subclinical ovine mastitis has been studied in several countries, investigations on CM are relatively few (Bergonier et al., 2003). For both mastitis forms, aetiological studies have mainly focused on causal organisms. There are few reports on the epidemiology of and risk factors for ovine mastitis, and these are partly based on observations in a single flock or a limited number of flocks.

Dairy sheep dominate the sheep industry in many countries. In other countries, including Norway, sheep are kept for meat and wool production. Differences in management practices between these production systems are likely to be associated with some differences in mastitis risk factor exposures.

There are somewhat diverging observations regarding the distribution of cases of CM in relation to the time of lambing. An Irish study in meat sheep found that one third of ewes with CM developed the disease during the first week postpartum (Onnasch et al., 2002). A study in Norway found that only 15% of the cases occurred in that period, while more than 60% occurred between 2 and 5 weeks postpartum (Indrebø, 1991). According to a study of dairy ewes in Greece, clinical cases in the dry period are not uncommon; a 5% cumulative incidence of mammary abnormalities (mainly CM) was recorded in that period (Saratsis et al., 1998).

In general, both individual animal and environmental factors are likely to affect the risk of mastitis. A tendency for increased risk of subclinical mastitis with increasing age of the ewe has been observed (Lafi et al., 1998; Al-Majali and Jawabreh, 2003). However, results from studies on CM are conflicting; some did not find any age effect (Quinlivan, 1968; Larsgard and Vaabenoe, 1993), whereas one study found primiparous to be at smaller risk than were older ewes (Indrebø, 1991).

Uniparous ewes have been found to be at a smaller risk of CM or subclinical mastitis as compared with those with two or more lambs (Gross et al., 1978; Indrebø, 1991; Lafi et al., 1998). In contrast, Torres-Hernandez and Hohenboken (1979) did not find any association between the number of lambs being suckled and risk of subclinical mastitis.

Breed differences in susceptibility to mastitis have been reported (Torres-Hernandez and Hohenboken, 1979; Watson et al., 1990), clearly suggesting genetic effects on the resistance to the disease. Heritability estimates, adjusted for breed effects, also suggest genetic influence (Larsgard and Vaabenoe, 1993). Moreover, mastitis resistance might be expected to be influenced by nutrition, and there are indications that a poor body condition increases the risk of the disease (Onnasch et al., 2002).

Ewes that have suffered CM are usually culled before the next breeding season and one would expect only those considered completely cured be kept in the flock. Whether or not such ewes are at increased risk of experiencing CM later does not seem to have been studied. An experimental study, in which milk samples were collected repeatedly from ewes, showed that subclinical mastitis occurred more frequently in certain ewes than what would be expected if cases occurred

at random (Torres-Hernandez and Hohenboken, 1979). Whether these findings were due to persistent infections or repeated new-infections was not clear.

The very few reports addressing risk factors for ovine CM were mainly based on univariable analyses of a relatively small number of cases from certain regions or flocks. Thus, more comprehensive studies are warranted in order to estimate, with reasonable precision, the multivariable-adjusted effects of various potential risk factors.

In Norway, individual ewe data, which include disease records, are available from a national sheep registry. We used data from this registry to identify risk factors for ovine CM, focusing mainly on individual animal characteristics suspected to be associated with CM. The distribution of CM cases in relation to the time of lambing was also examined.

#### 2. Materials and methods

#### 2.1. Data source

Data were collected from the databases of the Norwegian Sheep Recording System (NSRS). The NSRS was established in 1962. In 2004, 4402 flocks, comprising 24.8% of all sheep flocks in Norway, were enrolled and the total number of ewes included was 309,573. Each animal has a unique identification composed by numbers for county, municipality within county, flock within municipality and animal within flock.

Disease recording was introduced as part of the NSRS in 1995. Clinical cases in a flock are recorded on a standardized health card, identifying the particular animal and using specific code numbers for various diseases. Due to regulations regarding the sales and use of veterinary drugs, disease cases are usually recorded on the health cards by veterinary surgeons. CM is recorded as either "severe or moderate" or "mild" in accordance with definitions given for bovine mastitis (International Dairy Federation, 1999). In this study, we did not distinguish between these two diagnoses and used the diagnosis CM.

Initially, we identified all CM cases in 2004 in the disease record file. A disease episode is the unit of observation in this file and one particular animal may occur as one or more observations depending on the number of disease episodes experienced. Additional data were collected from a second file, in which each lambing of each ewe is the unit of observation (containing variables characterizing the particular lambing), and a third file in which each animal is the unit of observation (containing variables describing characteristics and events concerning the animal from birth until slaughter or death from other reasons than slaughter).

#### 2.2. Study design and animals and variables included

We conducted an *m:n* matched case–control study using flock as the matching factor. In 2004, a total of 3018 CM episodes were recorded in 2935 ewes in the NSRS. For the 83 ewes for which CM had been recorded twice, only the first episode was considered. One criterion for being included as a case was that the ewe had lambed between March 1 and June 15 in 2004. The main lambing season in Norway is in April and May, i.e., shortly before grass pasture is available. A lambing date prior to March 1 or later than June 15 is considered highly inconvenient and was recorded for only 0.3% of the ewes in the flocks participating in the NSRS in 2004. Cases of abortion are recorded using a specific code number and ewes that aborted in 2004 were not included.

As matched controls we included all ewes without any record of CM in 2004 that belonged to the same flocks as the cases. Ewes that experienced abortion and those that lambed prior to March 1 or later than June 15 were not included. In the NSRS, the date of and reason for death or culling are recorded. Some of the candidates for being controls in this study died or were culled within a few weeks of lambing. Based on results of previous studies (Indrebø, 1991; Onnasch et al., 2002), we expected most cases of CM to occur within 3 months of lambing. To ensure that the controls had been alive and thus at risk for CM during the most important part (from a CM risk perspective) of the observation period (2004), we decided not to include ewes that died or were culled within 90 days of lambing. "Slaughtered because of udder or teat damage" is one of the alternatives in the NSRS for coding the reason for death or culling, and some ewes were culled for that reason in 2004 without having a CM record that year. It could not be ruled out that some of these ewes had experienced a CM episode that was not recorded in the health card. Therefore, we decided not to include as controls ewes without a CM diagnosis that were culled because of udder or teat damage.

To represent the number of lambs per ewe we decided to use the total number of lambs born, regardless of whether or not any of the lambs were stillborn. In the NSRS, there are 21 different code numbers for breed, including a separate number for the Texel  $\times$  Norwegian White crossbreed and one for other crossbreeds or unknown breed. The course of lambing is recorded as either "normal", "assistance, but not dystocia" or "assistance, dystocia".

To establish the CM history of the ewes for the previous 3 years, records for the years 2001–2003 were collected from the disease record file in the NSRS and linked with the other data for our cases and controls. Ewes with one or more CM episodes during these years were considered "CM history positive" and those without a CM record were considered "CM history negative". The subgroup of ewes suitable for analyzing the potential effect of CM history, i.e. ewes that were at least 2 years old in 2004, included 2481 cases and 57,448 matched controls from 1005 flocks.

#### 2.3. Statistical methods

For descriptive purposes, crude odds ratios (OR) were calculated for the various regions of location of the ewes. Two dummy variables were created for modeling the course of lambing, using "normal lambing" as the reference. Three dummy variables indicating calendar time of lambing were formed, using lambing time prior to April 16 as the reference. Box–Tidwell transformation was used to test whether the ewes' age and number of lambs were linear in the log odds of experiencing CM (Hosmer and Lemeshow, 1989). Pearson chi-square test was used when comparing proportions of CM cases within different time periods.

Univariable and multivariable analyses were performed using conditional logistic regression. The likelihood function for the conditional logistic regression model is identical to the partial likelihood function for the Cox proportional hazards model (Allison, 1999) and we used the PHREG procedure in SAS (SAS Institute Inc., Cary, NC, USA). Due to the *m*:*n* matching design we specified the discrete logistic model, which uses the exact partial likelihood, to fit the data. Each flock constituted a stratum. As required by the PHREG procedure, dummy survival times were created; all cases were given the same event time value (=1) and all controls were censored at some arbitrarily fixed later time (=2).

Due to the small number of explanatory variables relative to the number of observations, all variables were included in the initial multivariable model. Potential interaction was assessed by adding two-factor interaction terms to the main effects multivariable model, using the likelihood ratio test for testing significance. When significant interaction was present, odds for each

subgroup were calculated by entering parameter point estimates from the final model together with subgroup specifications into the model and taking the antilog of the resulting logit. Subgroup OR values, adjusted for the effects of the other variables in the final model, were calculated after selecting a suitable reference category.

The *P*-values of the parameters were based on the Wald chi-square statistic. P < 0.05 was considered significant.

#### 3. Results

Of the 2935 ewes for which one or more CM episodes were recorded in 2004, 44 were excluded because they were nonpregnant or aborted and 34 were excluded because they lambed before March 1 or later than June 15. Thus, 2857 ewes met the eligibility criteria for cases; 2402 (84.1%) of which were diagnosed as "severe or moderate" CM and the remaining as "mild" CM. Of the control candidates that did not experience abortion and met the inclusion criterion regarding lambing date (n = 77,201), 440 were not included because they died or were culled within 90 days of lambing, and an additional 45 were not included because they were eligible as controls was 76,716. Characteristics of the cases and controls are shown in Table 1.

Cases and controls were from 1056 flocks. The number of cases within a flock varied from 1 to  $31 \pmod{8}$  (median = 2) and the number of controls from 8 to 542 (median = 61) (Table 2).

Fig. 1 shows the distribution of ewes with CM according to number of days between the times of lambing and treatment. Those treated earlier than 10 days prior to lambing (n = 70) or later than 100 days after lambing (n = 143) are not shown in the figure. Of the total number of cases, 175 (6.1%) were reported to occur prior to lambing, 1054 (36.9%) during the first 7 days postpartum, 927 (32.4%) between days 8 and 28 postpartum and 701 (24.5%) later than 28 days postpartum. The numbers of cases observed the second, third and fourth week postpartum were 304, 317 and 306, respectively. The relative proportion of cases was significantly smaller the second week than the first week postpartum (P < 0.001). Both the fifth and sixth week postpartum the relative proportions of cases were significantly smaller than that for the preceding week (P < 0.01).

One criterion for being included as a control was that the ewe was alive for at least 90 days after lambing, and 93.6% of the cases occurred before day 90 postpartum. The relative percentages of cases recorded in July, August, September, October, November and December were 1.3%, 3.5%, 1.5%, 1.1%, 1.0% and 0.2%, respectively. Of the controls, 9745 (12.7%) were culled in 2004. Of those culled, 27 (0.3%) were culled in July, 1433 (14.7%) in August, 3955 (40.6%) in September, 2506 (25.7%) in October, 1563 (16.0%) in November and 261 (2.7%) in December. The reasons for being culled were "slaughtered" (96.9%), "died" (1.1%), various diseases (0.3%), "sold as live animal" (which is a separate "culling" code) (0.5%), "accidents" (0.2%) and various other reasons (1.1%).

The incidence of CM cases (number of cases/total number of ewes included) in each of the 1056 flocks varied between 0.003 (1 case/299 ewes) and 0.381 (8 cases/21 ewes) (Table 2). The median incidence was 0.033 and the mean 0.042. The distribution of the flocks by CM incidence is shown in Fig. 2.

Crude analysis of the data, with western Norway as the reference region, showed that the OR for CM was 0.872 for eastern Norway, 0.736 for southern Norway, 0.619 for northwest Norway and 0.782 for northern Norway.

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Variable	Levels	Number		
		Cases	Controls	
Number of lambs born	1	204	14,506	
	2	1314	41,033	
	3	1140	18,786	
	>3	197	2,273	
	Missing	2	118	
Age of ewe (year)	1	376	16,709	
	2	636	18,685	
	3	620	14,915	
	4	505	11,342	
	5	388	7,709	
	6	222	4,674	
	>6	110	2,682	
Course of lambing	Normal	1966	57,539	
	Assistance, not dystocia	315	8,323	
	Assistance, dystocia	576	10,851	
	Missing	0	3	
Location within Norway <sup>a</sup>	Eastern Norway	1038	25,897	
	Southern Norway	510	15,055	
	Western Norway	497	10,828	
	Northwest Norway	316	11,123	
	Northern Norway	496	13,813	
Time of lambing	March	47	1,073	
	April 1–15	421	9,764	
	April 16–30	1147	29,501	
	May 1–15	1048	29,617	
	May 16–31	180	6,262	
	June	14	499	
Breed	Old Norwegian breeds <sup>b</sup>	283	10,862	
	Other breeds <sup>c</sup>	2574	65,854	
Mastitis history	Mastitis 2001, 2002 or 2003	66	315	
	Not mastitis 2001–2003	2415	57,133	
	Irrelevant <sup>d</sup>	376	19,268	

Table 1
Characteristics of the 79,573 ewes included in the study (2857 cases and 76,716 controls)

<sup>a</sup> Eastern Norway: Oppland, Hedmark, Akershus, Østfold, Vestfold, Buskerud and Telemark counties; *southern* Norway: Aust-Agder, Vest-Agder and Rogaland counties; *western Norway*: Hordaland and Sogn og Fjordane counties; *northwest Norway*: Møre og Romsdal, Sør-Trøndelag and Nord-Trøndelag counties; *northern Norway*: Nordland, Troms and Finnmark counties.

<sup>b</sup> Including Spael (n = 10,037), Furbearing Sheep (n = 874), Blazed Sheep (n = 126), Old Norwegian Sheep ("Wild Sheep") (n = 104), Fuglestad Pied (n = 63), Grey Trønder (n = 20).

<sup>c</sup> Including Norwegian White (n = 34,898), Dala (n = 17,033), Steigar (n = 11,043), Rygja (n = 3099), Texel (n = 1078), Cheviot (n = 1065), Suffolk (n = 335), Merino (n = 121), Finnish Landrace (n = 29), Oxford Down (n = 27), Blackface (n = 2), various crossbreeds (n = 104).

<sup>d</sup> Ewes that were 1 year old in 2004 (n = 17,085) and additional original controls that did not belong to flocks with cases >1 year old (n = 2559).

Number of cases per flock	Number of flocks	Number of controls per flock			
		Median	Minimum	Maximum	
1	419	54	8	299	
2	256	54	11	292	
3	146	64.5	12	542	
4	81	78	17	275	
5	52	89.5	27	186	
6	33	84	28	202	
7	15	95	37	264	
8	14	81.5	21	171	
9	9	95	49	218	
10	8	107	51	320	
11	6	88.5	29	200	
12	5	110	100	295	
13	2	71.5	48	95	
14	2	81.5	52	111	
15	2	75.5	63	88	
16	1	-	80	80	
17	2	101	97	105	
18	1	-	174	174	
19	1	-	149	149	
31	1	_	203	203	

Table 2 Distribution of clinical mastitis cases and controls on flocks

In most flocks, ewes were bred for the first time when they were around 7 months old. Thus, age (in years) is identical to parity for most of the ewes. In only 80 (7.6%) of the flocks, with a total of 3923 ewes (4.9% of the ewes in the study), there were no ewes that lambed for the first time by 1 year of age.

The Box–Tidwell transformation did not reveal any significant nonlinear relation between age and the logit of CM ( $\chi^2 = 2.17$  for the age  $\times$  log age term when added to the multivariable model;

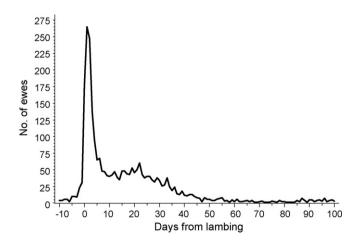


Fig. 1. Daily number of ewes treated for clinical mastitis between 10 days before and 100 days after lambing (n = 2644) by number of days from lambing to treatment (70 ewes treated earlier than 10 days prepartum and 143 ewes treated later than 100 days postpartum not shown).

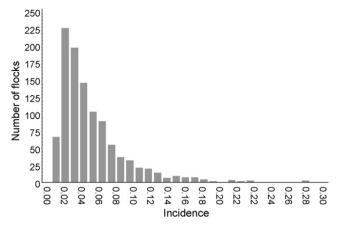


Fig. 2. Distribution of 1056 sheep flocks by incidence of clinical mastitis (number of cases/total number of ewes) grouped in intervals of 0.01 (figures are upper limits of intervals) (flocks in the Norwegian Sheep Recording Scheme, 2004).

P = 0.14), while a highly significant nonlinear component was present in the relation between number of lambs and the logit of CM ( $\chi^2 = 15.06$ ; P < 0.001). Therefore, age was kept as a continuous variable in the models, while number of lambs was treated as a categorical variable (grouped as 1, 2, 3 and >3 lambs), using dummy variables with 1 lamb as the reference group.

Univariable conditional logistic regression analyses of the potential risk factors under study showed that, with the exception of "assistance at lambing, not dystocia", all variables were significant (Table 3).

We included all explanatory variables in the multivariable model and the results are shown in Table 3. The odds of CM increased with increasing age of the ewe and with increasing number of lambs born to the ewe, and were greater in ewes assisted at lambing due to dystocia than in ewes that lambed without assistance. However, the interaction terms number of lambs × age and number of lambs × course of lambing (dystocia versus not dystocia) were significant, which had to be taken into account when calculating OR for the various levels of these variables. The interaction between number of lambs and age is displayed in Fig. 3. There was a greater increase in log odds of CM with increasing age in ewes with 1 lamb than in ewes with >1 lamb. A 1-year increase in those with 1 lamb was associated with 19.2% increase in the odds of CM, while a corresponding increase in the age of those with >1 lamb was associated with between 3.5 and 6.6% increase in the odds of CM. Fig. 4 shows that the effects of different courses of lambing on log odds of CM depended on the number of lambs born. The effect of dystocia on the risk of CM was largest for ewes with 1 lamb (point estimate for OR, 1.72), somewhat smaller for those with 2 lambs (OR = 1.39) and close to unity for those with 3 lambs (OR = 0.97) or >3 lambs (OR = 1.05), using normal lambing as reference.

The effect of number of lambs was large. As an example, accounting for the additional effects of age and dystocia, the odds of CM were 9.24 times greater for a 5-year-old ewe with dystocia and >3 lambs compared with a 1-year-old ewe that gave birth to 1 lamb without dystocia.

Ewes of Spael or other old Norwegian breeds had smaller odds of CM as compared with ewes of other breeds (Table 3). Two-factor interaction terms including breed together with each of the main effect variables for age, number of lambs, course of lambing and time of lambing were not significant when added separately to the final multivariable model (likelihood ratio tests; 0.1 < P < 0.8). The observed proportions of cases within the various breeds lent support to our

Table 3

Variable	Univariable			Multivariable adjusted <sup>c</sup>		
	OR	95% CI	Р	OR	95% CI	Р
Number of lambs born						
1	1.00			1.00		
2	2.26	1.94-2.62	< 0.01	3.19	2.42-4.21	< 0.01
3	4.37	3.74-5.09	< 0.01	5.80	4.29-7.85	< 0.01
>3	6.25	5.07-7.69	< 0.01	8.84	5.21-15.0	< 0.01
Age of ewe (year, continuous)	1.15	1.13-1.18	< 0.01	1.19	1.11-1.28	< 0.01
Course of lambing						
Normal	1.00			1.00		
Assistance, not dystocia	1.04	0.91-1.19	0.61	1.14	1.00-1.31	0.06
Assistance, dystocia	1.43	1.29-1.59	< 0.01	1.72	1.15-2.58	< 0.01
Time of lambing						
Before April 16	1.00			1.00		
April 16–30	0.84	0.73-0.96	0.01	0.92	0.80 - 1.05	0.22
May 1–15	0.76	0.64-0.89	< 0.01	0.86	0.73-1.02	0.08
After May 15	0.57	0.45-0.72	< 0.01	0.69	0.54-0.88	< 0.01
Breed						
Old Norwegian breeds	1.00			1.00		
Other breeds	1.58	1.26-1.98	< 0.01	1.69	1.34-2.13	< 0.01
Interactions						
Dystocia $\times$ 2 lambs				0.80	0.52-1.24	0.32
Dystocia $\times$ 3 lambs				0.56	0.37-0.86	< 0.01
Dystocia $\times >3$ lambs				0.61	0.37-1.02	0.06
Age $\times$ 2 lambs				0.88	0.81-0.95	< 0.01
Age $\times$ 3 lambs				0.89	0.83-0.97	< 0.01
Age $\times >3$ lambs				0.87	0.76-0.99	0.03

Univariable and multivariable conditional logistic analyses <sup>a</sup> of potential risk factors for clinical ovine mastitis in Norwa	y
(2004) <sup>b</sup>	

<sup>a</sup> Conditioned on flock (the matching factor).

<sup>b</sup> Multivariable analysis included 2855 cases (no. of lambs missing for 2 ewes) and 76,597 controls (number of lambs and/or course of lambing missing for 119 ewes).

<sup>c</sup> Adjusted for the effects of the other variables in the table.

choice for grouping of the breeds. Of the breeds represented by at least 10 cases in the study, the proportions of cases within each of the old Norwegian breeds (<0.0027) were smaller than the proportions of cases within each of those classified as other breeds (>0.0034).

Using ewes that lambed prior to April 16 as the reference group, multivariable analysis revealed that OR for those that lambed later than May 15 was significantly smaller than 1 (Table 3). In contrast to the results of univariable analyses, the ORs estimated from the multivariable model for ewes that lambed between April 16 and May 15 were not significantly different from 1. Analysis of a subset of the data, including only cases treated within 30 days of lambing (n = 2260) and their matched controls (n = 72,279) and using the conditional logistic model that was fitted to the entire dataset, yielded a nonsignificant OR for those treated later than May 15 (OR = 0.83, P = 0.16). The other variables that were significant when analyzing the entire dataset were significant also for the subset (data not shown).

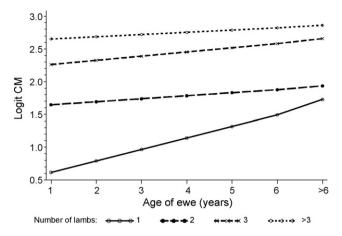


Fig. 3. Log odds of clinical mastitis by age for ewes giving birth to 1, 2, 3 or >3 lambs based on estimates from the multivariable model.

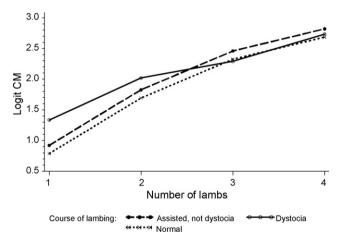


Fig. 4. Log odds of clinical mastitis by age for ewes that had a normal lambing or were assisted at lambing, with or without dystocia, based on estimates from the multivariable model.

Table 4

Univariable and multivariable conditional logistic analyses<sup>a</sup> of association between clinical mastitis (CM) history in the period 2001–2003 and odds ratio for experiencing CM in 2004 (Norway)<sup>b</sup>

Mastitis history	Univariable			Multivariable adjusted <sup>c</sup>		
	OR	95% CI	Р	OR	95% CI	Р
No mastitis 2001–2003	1.00			1.00		
Mastitis 2001, 2002 or 2003	4.17	3.13-5.57	< 0.01	3.96	2.95-5.33	< 0.01

<sup>a</sup> Conditioned on flock.

<sup>b</sup> Including ewes that were at least 2 years old in 2004 (2481 cases and 57,448 controls matched on flock).

<sup>c</sup> Adjusted for the effects of age, number of lambs, course of lambing, time of lambing, breed and the interaction between dystocia and number of lambs.

Adjusted for the effects of other significant predictors, ewes with at least one CM episode in 2001, 2002 or 2003 had greater odds of experiencing CM in 2004 than those without any CM record in that time period (Table 4). In addition to mastitis history, the significant terms in the multivariable model fitted to this subset were the same as those in the model fitted to the entire dataset, with the exception of the interactions between age and number of lambs (0.5 < P < 0.8).

## 4. Discussion

This population-based case-control study identified and quantified the effects of several individual animal risk factors for ovine CM. The total number of lambs born to the ewe, including stillborn, had a major impact on the odds of suffering CM; increasing number of lambs was associated with increasing odds. Age of the ewe and whether or not assistance at lambing was required due to dystocia also affected the odds of CM. Ewes assisted at lambing due to dystocia had greater odds of CM than had those with an uncomplicated lambing, and increasing age tended to be associated with increasing odds of CM. However, the effects of both age and the course of lambing were modified by the number of lambs born. Considering all three factors together, and controlling for the effects of the other significant variables, we found that the odds of CM were more than 9 times greater for a 5-year-old ewe with >3 lambs that was assisted at lambing because of dystocia, as compared with a 1-year-old ewe with single birth and unassisted delivery. Previous studies have also found ewes with multiple births to be more susceptible to CM than were ewes with single birth (Indrebø, 1991; Larsgard and Vaabenoe, 1993). It has been suggested that teat irritation from vigorous sucking may occur more frequently in multiparous ewes, particularly those nursing more than two lambs, and teat lesions increase the risk of CM (Indrebø, 1991). However, due to the pronounced peak in CM occurrence very close to lambing, other factors might be suspected to be important. It has been shown that resistance to bovine mastitis is decreased at parturition (Kehrli et al., 1989), and one may speculate whether periparturient resistance is impaired to a greater extent in ewes with multiple births than in those with single births. Teat injuries caused by sucking lambs would be expected to occur more frequently after teeth have erupted and some previous studies have reported a second peak in CM occurrence approximately 3-5 weeks after lambing (Indrebø, 1991; Onnasch et al., 2002). The pattern of CM occurrence in our study clearly suggests that apart from the risk associated with lambing and the immediate postpartum period, there is a proportionally greater risk during the first 5 weeks postpartum compared with the subsequent weeks. Whether there are differences between risk factors for CM cases occurring close to lambing and those for cases occurring a few weeks postpartum should be studied more closely.

We found that the effect of age on the odds of CM differed between ewes with 1 lamb and those with >1 lamb; in the latter, increasing age was associated with only a modest numerical increase in the odds, whereas a more pronounced increase was observed in ewes with singles (Fig. 3). One possible explanation is that milk yield of ewes increases with increasing age (Ruiz et al., 2000), and a single lamb will not empty both glands of a high-producing ewe. As has been shown experimentally in cows (Thomas et al., 1972), poor emptying of a gland will increase the risk of CM. Some studies of subclinical mastitis have found the risk to increase with age (Watson et al., 1990; Watkins et al., 1991; Lafi et al., 1998; Al-Majali and Jawabreh, 2003), while previous studies of the association between age and CM are conflicting. One study found a tendency to a higher CM frequency in older ewes (Indrebø, 1991), while others did not find any relationship (Kvitrud and Lysne, 1959; Quinlivan, 1968; Larsgard and Vaabenoe, 1993). Most of these studies used univariable analyses; therefore, the results might have been influenced by confounders.

Moreover, the highly significant interaction between age and number of lambs must be taken into account when addressing age effects.

Assisted lambing because of dystocia increased the odds of CM. Epidemiologic studies have also found an association between dystocia and mastitis risk in cows (Peeler et al., 1994). There was an interaction between dystocia and number of lambs born; the adverse effect of dystocia was most pronounced in ewes with singles (Fig. 4). Fetopelvic disproportion is a major cause of dystocia in ewes with single lambs (Arthur et al., 1996) and is often associated with prolonged straining. In such cases, increased susceptibility to postpartum intramammary infection is conceivable. In ewes delivering 2 lambs, the effect of dystocia on CM risk was less pronounced than in ewes with 1 lamb, while no associated with multiple birth, successful obstetric delivery is often achieved with less difficulties because of the smaller fetuses.

We found differences between breeds in the risk of CM. Ewes of several different breeds were included in the study; however, most breeds were represented by only a few cases and it was not possible to estimate with reasonable precision the effects of each single breed. We therefore chose to treat animals of some old Norwegian breeds as one group and the other Norwegian breeds together with foreign breeds as a second group. The latter group had 1.7 times greater odds of experiencing CM as compared with the old Norwegian breeds, of which ewes of the Spael breed comprised almost 90%. A previous study also found Spael ewes to be at a smaller risk of CM as compared with ewes of some other Norwegian breeds (Larsgard and Vaabenoe, 1993). Other researchers have also reported breed differences in ovine mastitis risk. In Australia, Watson et al. (1990) observed a higher prevalence of intramammary infection in Border Leicester  $\times$  Merino crosses than in Merino ewes and Border Leicester  $\times$  Booroola Merino crosses.

In Norway, it is common practice that sheep-owners check the udder of ewes before breeding and those with visible or palpable udder abnormalities are culled. If not routinely culled, it is likely that ewes known to have suffered from CM are examined particularly thoroughly before the subsequent breeding season, and those without any clinical abnormalities may be considered cured and not culled. According to the health card records, some of the cases and controls in this study had experienced CM at least once during the previous 3 years. Analysis of data on ewes >1 year showed that ewes with a CM history were at increased risk of suffering a new CM episode; adjusted for the effects of other important risk factors the odds of such ewes experiencing CM were four times the odds of ewes without a CM history. Thus, there are good reasons to recommend that ewes that have suffered from CM be culled the subsequent autumn, regardless of whether or not signs of udder abnormalities are present.

The literature on ovine mastitis contains very little information regarding effects of flock characteristics on CM risk. It is very likely that environmental factors and management practices, shown to be important risk factors for bovine mastitis (Barnouin et al., 2005), strongly affect the risk of ovine CM. Such factors vary among flocks. The distribution of individual animal risk factors under study may also vary among flocks and flock is therefore a potential confounder. The mean number of cases and controls per flock in this study was 2.7 and 72.6, respectively, and within-flock correlation in the data would be expected to be present to some extent. If not accounted for in the analysis this could lead to incorrect variance estimates. Furthermore, potential differences in routines for disease recording among flocks can introduce bias. Therefore, to ensure valid estimates for our individual animal risk factors, we decided to use a matched case–control study design with flock as the matching factor. This design also eliminated bias that might have been caused by different geographic location, a factor shown to affect the risk of ovine CM (Quinlivan, 1968). Crude analysis of our data demonstrated variation between

regions within Norway; however, this study was not designed to explore such relationships and these results should be considered merely suggestive.

Misclassification of ewes is a possibility. Usually, the diagnosis CM is straightforward and it is very unlikely that controls were misclassified as cases. However, failure to detect or record mild cases of CM might have occurred among ewes included as controls. Farmers are usually quite aware of udder health of the ewes, particularly around the time of lambing and the subsequent weeks. Later, usually in the end of June, many flocks in Norway are moved to distant pastures, often in the mountains. Cases of CM may then occur without being noticed. In most such cases, chronic udder changes would likely be the result. These would normally be detected when the ewes return home in the autumn, at least upon udder examination before the subsequent breeding season. However, there might be some doubt whether all such chronic CM cases are recorded in the health cards. Our final model identified time of lambing as a risk factor; ewes that lambed later than May 15 had smaller odds of having a CM diagnosis as compared with those that lambed before April 15. However, when analyzing a subset of the data, limited to cases treated within 30 days of lambing and their matched controls, time of lambing was not significant. The explanation for this discrepancy could be that, in many flocks, the period postpartum under careful observation on home pastures was shorter for ewes that lambed late in spring and CM episodes occurring on mountain pastures might have passed without being registered. Some ewes without a CM diagnosis were culled because of udder or teat damage, some of which might have had CM. However, these were not included as controls. Nevertheless, some ewes that had suffered from CM might have been misclassified as CM negative controls in our study. Misclassification of cases as controls would tend to produce smaller risk estimates than the real ones.

We selected cases of CM during 2004. Ideally, all controls should have been alive and thus at risk for CM throughout the entire 2004, which was not the case. Nearly 13% of the controls were culled in 2004. We did not include as controls ewes that died or were culled within 90 days postpartum, which means that all controls were at risk during the pre- and postpartum period where the great majority (94%) of the CM cases occurred. Although some of the controls that were culled later than 90 days postpartum were hypothetically misclassified ewes because they might have developed CM if they had remained alive throughout 2004, the number of such ewes would likely be very small. Letting the overall proportion of CM cases in the study (2857/79,573 = 0.036) represent the CM incidence risk for the ewes in the flocks in 2004 and considering the small proportion of cases that occurred during the final months of the year, very few CM cases would be expected to occur among the controls that were culled in 2004.

Various pathogens are involved in ovine mastitis and, depending on their reservoirs and pathogenesis, mastitis caused by different organisms would be expected to be associated with some risk factor differences. In Norway, *Staphylococcus aureus* is the predominant bacterial cause of ovine CM and was in a recent comprehensive study isolated from 65% of affected glands (Mørk et al., 2007). Studies of ovine CM in other countries also have found *S. aureus* to be an important causal organism (Bergonier et al., 2003). Thus, it is likely that the risk estimates from the current study, which are adjusted for breed effects and not influenced by inter-flock differences in environmental risk factor exposures, are valid also for meat sheep populations in other countries than Norway.

## 5. Conclusions

We found that the odds of CM in meat sheep increased markedly with increasing number of lambs born to the ewe, a fact which should be taken into consideration in sheep breeding programmes. In ewes with 1 lamb, increasing age was associated with increased odds of CM; in those with more than 1 lamb there was only a slight numerical increase in OR with increasing age. Dystocia was associated with increased odds of CM in ewes with 1 or 2 lambs. Ewes of old Norwegian breeds, mainly Spael, had smaller odds of CM than had ewes of other breeds. Compared with ewes without a CM history, those that had such a history had considerably greater odds of experiencing CM. Thus, it is reasonable to recommend that ewes that have suffered CM be slaughtered before the subsequent breeding season, even if there are no signs of chronic udder changes.

Because more than 40% of the CM cases occurred between 7 days before and 7 days after lambing further studies that specifically address risk factors in the periparturient period should be undertaken.

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