

Milk hygiene and udder health in the periurban area of Hamdallaye, Niger

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Abstract The prevalence of intra-mammary infections in dairy herds was studied in Hamdallaye, Niger. A total of 956 milk samples were collected in 2007 from 239 lactating cows of four local breeds in eight traditional herds; the first sampling was undertaken in

the dry season at morning milking, and the second in the rainy season at evening milking. *Staphylococcus aureus*, Coagulase-Negative Staphylococci (CNS) and environmental microorganisms were detected in significantly ($p < 0.05$) more samples in the rainy season, 55.2%, than in the dry season, 27.1%. Statistically significant ($P < 0.05$) differences in prevalence were observed among herds and according to lactation number. Infections were assigned to four classes, according to the major pathogen, and the respective mean somatic cell counts during the dry season were: *S. aureus*, 775×10^3 cells/ml; CNS, 447×10^3 cells/ml; environmental microorganisms, 407×10^3 cells/ml; and non-infected, 262×10^3 cells/ml. Most of the tested strains were sensitive to antibiotics, and selected strains of *S. aureus* ($n=15$) were negative to the multiplex PCR tests for production of enterotoxins.

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Introduction

Mastitis is a very common disease, especially in intensively managed dairy herds of northern countries, but has never been considered among the most important health issues in less productive tropical bovine breeds (Rege et al. 1994). Dairy cows in tropical regions producing on average 500 kg of milk per lactation rarely experience clinical mastitis (Gambo and Agnem Etchike 2001). Nevertheless, in

recent years, milk producers in developing countries have noted the presence of intra-mammary infections: with no evidence of symptoms or clear milk alteration occurs, but damage in terms of animal production and public health are remarkable. In Niger, the proportion of milk refused by collection centres for hygienic reasons is estimated to be between 17 and 30% (Ruppel et Dan Gomma 2000). Many of the production units are in distant sites, precluding the possibility of routine surveys on milk quality (Marichatou et al. 2005) to evaluate udder health, determine infections prevalence in the herds and to regulate the quality of milk for consumer. The objective of this study was to quantify, for the first time, intra-mammary infections in eight traditional herds that provide raw milk to the collecting centre of Hamdallaye, recently instituted by PSEAU, to determine potential risks for public health and to propose control measure fit to local situation.

Materials and methods

Study areas

The rural municipality of Hamdallaye (Fig. 1), in Niger, is 30 km from Niamey and its geographical coordinates are: 2°37' of Longitude East; 2°08' of Longitude West; 13°51' of Latitude North and 13°33' of Latitude South. Its climate is Sahel-type, characterized by two distinct seasons: a rainy season lasting 3–4 months and a dry season lasting 8–9 months. Annual average rainfall is 400 mm, but can vary considerably from year to year.

The 8 studied sites were chosen among 16 sites organized in the Union of Dairy Producers Cooperative of Rural Municipality of Hamdallaye "UPROLAIT", created by PSEAU (Project of Safety of Niamey Periurban Farming and Agriculture) and have access to a recently established collection centre for raw milk.

The criteria for inclusion of sites in the study were: a distance from Hamdallaye of <20 km, the availability of the farmers to collaborate and a declared daily production of at least 250 litres.

Sampling and sample processing

Before the study, farmers were surveyed to collect general information about husbandry procedures. Then, 952 quarter milk samples were collected from

239 lactating cows in two different seasons, 114 and 125 cows, respectively, in the dry and rainy seasons.

All samples were frozen, and then transported to the Laboratory of Department of Veterinary Pathology, Hygiene and Public Health of the University of Milan. Briefly, 10 µl of each milk sample was spread on blood agar plates, incubated aerobically for 24 h at 37° C and then evaluated for identification of bacteria. For each sample from the dry season, SCC was determined by using an automated fluorescent microscopic somatic cell counter (Bentley Somacount 150, Bentley Instruments, Dublin, Ireland). Strains of *S. aureus*, *CNS*, *Streptococcus uberis* and *Pseudomonas spp.* were tested for antibiotic resistance with the disk diffusion Kirby-Bauer methods. Finally, fifteen strains of *S. aureus* by the dry season samples were tested with multiplex PCR test for production of enterotoxins A, C, D, G, H, I, J and L.

Statistical analysis

The data were analysed statistically by the SPSS version 16.0 (*Statistical Package for the Social Sciences*) using the Pearson Chi-square test ($P < 0.05$).

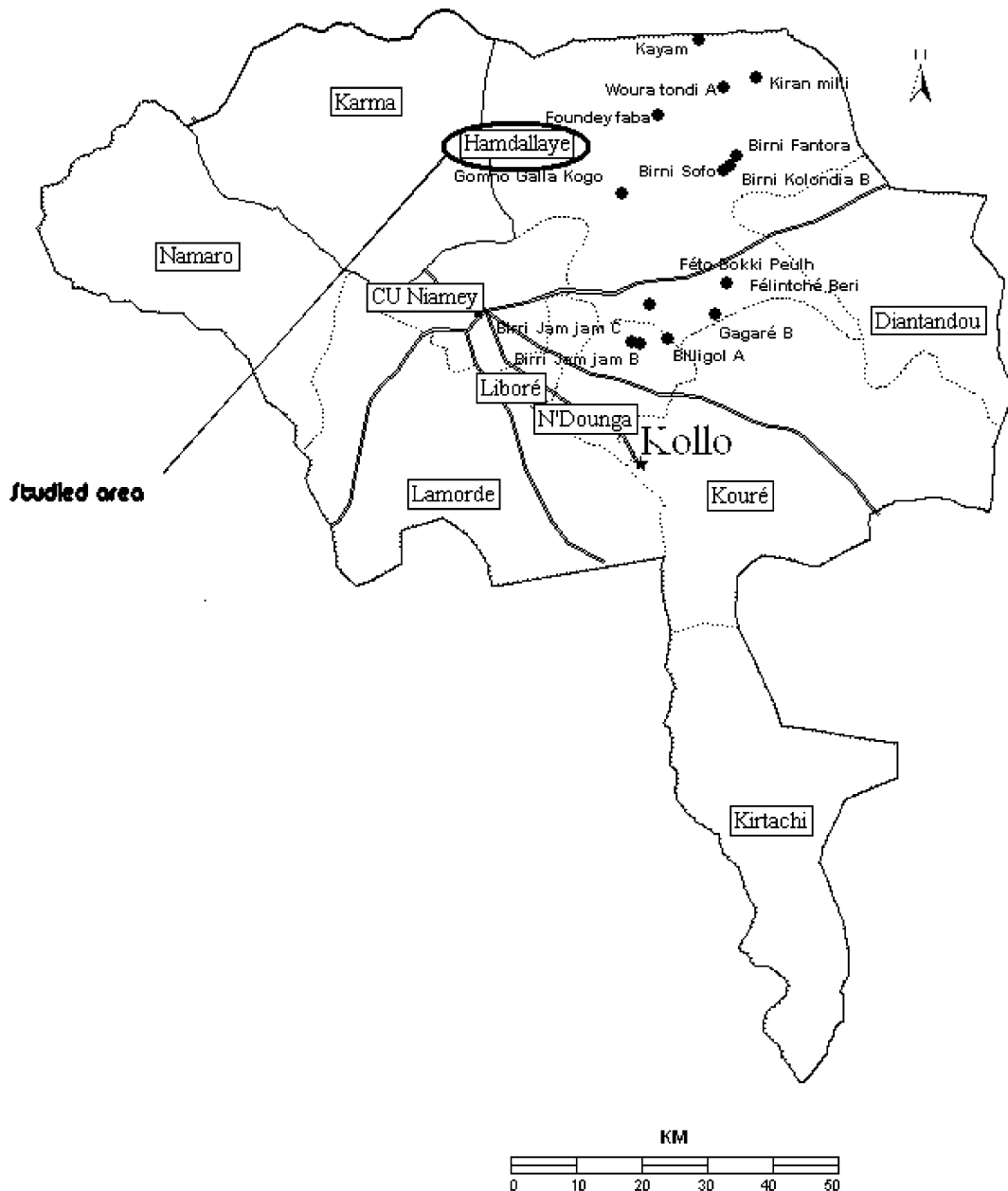
Results

Survey results

Ninety-one farmers were interviewed for general information about livestock practices. The cows represented the following local breeds: Bororo (37%), Azawak (32%), Djelli (18%), Goudali (2%) and their crossbreeds (11%). Cows are milked manually by the farmer, twice daily: in the morning for sale and in the evening for home-consumption. Milking takes place in the open air, nearby the rest of the herd and pumpkins are used as containers in most cases (the others use plastic bins); 55% of farmers hold the container during milking, while 45% set in on the ground.

Approximately 27% of farmers practice fore-stripping to check for abnormal milk and filtration is practised by 54% of farmers. Knowledge about quality and safety of milk are quite deficient. Nevertheless, they are able to recognize aspects of quality based on a drop of milk in terms of consistency (too liquid), colour or composition (low fat content).

Finally, with regard to udder health, 53% of farmers were able to identify lesions on the udders, 40% lesions



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Fig. 1 Geographical localization of the municipality of Hamdallaye, Department of Kollo, Niger

and inequality of the teats and 33% mastitis. Traditionally, mastitis is cured with application of plant materials.

Laboratory analysis

According to the bacteriology, the prevalence of subclinical mastitis in Hamdallaye varied from 27.1% to 55.2% ($P < 0.05$), respectively, between

dry and rainy seasons. Mastitis infections were classified into three groups: 1) *S. aureus*, 2) CNS and 3) environmental microorganisms and reported across seasons and sites in Table 1. An association between bacteria and lactation number ($P < 0.05$) was observed (Table 2) no differences were detected among breeds for mastitis infection ($P > 0.05$).

Table 1 Prevalence of subclinical mastitis in 8 herds in the Hamdallaye area

Period	Bacteria	Herds								Total	
		Louga	Gagare	Félintche	Bartawal	Féto-bokki	Toka-binekani	Toulwey	Toulware		
Dry season	Negative	N	40	112	67	73	62	25	38	417	
		%	66.7	68.3	79.8	91.3	81.6	56.8	59.4	72.0	
	<i>S. aureus</i>	N	9	29	3	3	10	1	11	66	
		%	15	17.7	3.6	3.8	13.2	2.3	17.2	11.5	
	SCN	N	11	14	9	3	4	17	15	73	
		%	18.3	8.5	10.7	3.8	5.3	38.6	23.4	12.8	
	Environmental	N	0	9	5	1	0	1	0	16	
		%	0	5.5	6	1.3	0	2.3	0	2.8	
Rainy season	Negative	N	6	7	10	12	7	5	2	7	56
		%	33.3	38.9	76.9	80	53.8	38.5	13.3	35	44.8
	<i>S. aureus</i>	N	11	8	0	2	3	3	0	13	40
		%	61.1	44.4	0	13.3	23.1	23.1	0	65	32
	SCN	N	0	0	0	0	2	1	1	0	4
		%	0	0	0	0	15.4	7.7	6.7	0	3.2
	Environmental	N	1	3	3	1	1	4	12	0	25
		%	5.6	16.7	23.1	2.7	7.70	30.8	80	0	20

The SCC ranged from 10^3 to 10^7 cells/ml of milk. Infections were assigned to four classes, according to the major pathogen, and the respective mean SCC (<0.05) were: non-infected (262×10^3 cells/ml); *S. aureus* (775×10^3 cells/ml); CNS (447×10^3 cells/ml) and environmental microorganisms (407×10^3 cells/ml).

Antimicrobial susceptibility and multiplex PCR

Sensitivity of bacteria (*S. aureus*: n=16; *Streptococcus uberis*: n=1; CNS: n=1 and *Pseudomonas* spp.: n=1) was tested against 25 antibiotics and combinations of antibiotics. *Streptococcus uberis* and CNS showed a good sensitivity to all the antibiotics tested. In all herds, *S. aureus* were resistant to Cefquinome and

Tetracycline in both dry and rainy seasons and *Pseudomonas* spp., found in only two herds (Toulwey and Gagaré), was resistant to all the antibiotics.

Finally, all of the 15 strains of *S. aureus* tested were negative for enterotoxins, according to multiplex PCR.

Discussion

One important finding from the survey undertaken was the complete ignorance by farmers of well-accepted (in industrialized countries) hygiene practices such as the cleaning and disinfection of teats before and after milking. Failure in cleaning exposes the udder and milk to microbiological contamination. Mean prevalence of intra-mammary infections in Hamdallaye dairy cattle was 42.2% (27.1% and 55.2% in the dry and rainy seasons, respectively), whereas the farmers estimated the prevalence to be around 33%. Major mastitis causing pathogens isolated were classified according to their origin and route of transmission into two groups: contagious, whose origin is the skin, and are transmitted from cow to cow, mainly during milking (*S. aureus*, CNS) and not contagious (*Streptococcus uberis*, *Pseudomonas* spp.), whose origin is bedding, and transmitted from the environment to the cow (Andrews et al. 2004). In this study contagious microorganisms, with a signif-

Table 2 Lactation number and isolation of bacteria in Hamdallaye dairy cattle

Lactation number	Bacteria				
	Negative	<i>S. aureus</i>	CNS	Environmental	
1	N	19	13	0	5
	%	51.4	35.1	0	13.5
2	N	19	4	1	9
	%	57.6	12.1	3	27.3
≥ 3	N	18	23	3	11
	%	32.7	41.8	5.5	20
Total	N	56	40	4	25
	%	44.8	32	3.2	20

icant predominance of *S. aureus*, were more prevalent (24.30–35.20% for herd) than environmental microorganisms (2.80–20.00%). These results agree with those in other aetiological reports on infectious mastitis in urban and periurban areas of Niamey (Issa Ibrahim 2005) and other African countries Burkina Faso, Tanzania, Mali and Ethiopia (Sidibé et al. 2004; Kivaria et al. 2004; Bonfoh et al. 2003; Almaw et al. 2008). Significantly greater numbers of microorganisms were isolated in the rainy season than in the dry season (Table 1), particularly for environmental microorganisms.

No statistically significant differences among breeds were observed in the prevalence of microorganisms. This result was probably related to the similarity in environment, breeding methods and low production levels in all sites and a lack of a strong genetic and breed-related effect.

With regard to lactation number, CNS and environmental microorganisms increased significantly from parity 1 to parity ≥ 3 (respectively from 0.00% to 5.50% and from 13.50% to 20.00%,) (Table 2). Cattle in first and third or greater lactation cows were more infected by *S. aureus* 35.10% and 44.80%, respectively, than in lactation 2. Our results are consistent with previous studies in Cameroon that demonstrated a significant difference ($p < 0.01$) in subclinical mastitis prevalence between primiparous and multiparous cows (Gambo et al. 2001). In Ethiopia, Demelash et al. (2005) found that older cows (> 10 years) were more affected by subclinical mastitis (38.6%), than younger cows (23.6%), in which clinical mastitis was predominant. The increase in mastitis risk, relate with parity was also described from Kerro Dego and Tareke (2003), Rakotozandrindrainy et al. (2007) and Karimuribo et al. (2008). A significant relationship was found between somatic cell count and the presence of mastitis causing pathogens: *S. aureus* (775×10^3 cells/ml), CNS (447×10^3 cells/ml) and environmental microorganisms (407×10^3 cells/ml). *S. aureus* was the most frequently isolated major pathogen from subclinically infected quarters and was also responsible for the largest increases in SCC observed.

In the present study, sensibility tests showed two types of *S. aureus* in Hamdallaye herds: *S. aureus* resistant to Cefquinome and *S. aureus* resistant to Tetracycline. Resistance to Tetracycline (15%) and cefquinome (24.7%) has been reported by other

researchers in Kenya (Shitandi and Mwangi 2004) and Turkey (Kirkkan et al. 2005). Pseudomonas strains copiously isolated in two sites (Toulwey and Gagaré) were resistant to all antibiotics tested.

The negativity of the 15 strains of *S. aureus* tested to multiplex PCR for enterotoxins is a positive result. Nevertheless, pasteurization of milk is still recommended, when possible for these farmers and their customers and the research of enterotoxigenic strains should continue.

To conclude, this work suggest that the farmers in the area surveyed need to improve hygienic practices, with particular regard to milking, milk conservation and manipulation. This aim could be achieved by implementation of professional training for farmers and animal caretakers to increase their awareness about proper sanitation for human and animal health.

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