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Udder and teat morphometry and its relationship with occurrence of intramammary infections in dairy cattle

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Abstract

Bovine mastitis, an inflammatory disease of the mammary gland, is often caused by bacterial infection and is a major health problem on dairy farms. A cross-sectional study of four herds was conducted to assess udder and teat-related risk factors for the development of intramammary infections in 243 lactating cows. Udder and teat morphometric parameters were determined through visual appraisal method. Intramammary infection was assessed using California mastitis test (CMT), Somatic cell count (SCC) and bacteriology of milk samples. The frequencies of udder conformations were 65.8%, 18.9%, 7.4% and 7.8% for desirable, pendulous, round and goaty-shapes, respectively. Teat conformation revealed that 76.0%, 5.3%, 6.6%, 4.5% and 7.5% for desirable, short, funnel, bottle and conical-shapes, respectively. There was a significant (p<0.01) effect of udder shape and teat-end shape on SCC level and intramammary infections. The mean SCC and mastitis infection levels for undesirable udder was significantly (p<0.05) higher as compared to the desirable shaped udder. Similarly, significant (p<0.05) higher level SCC was found in undesired teats compared to desirable teats. The overall proportion of quarter mastitis infection in all four farms revealed 1.23% for clinical and 36.52% for subclinical mastitis. Herd-wise subclinical mastitis prevalence showed 42.8%, 35.07%, 33.09% and 2.94% for farm A, C, B and C, respectively. The most frequently isolated bacterial species were Staphylococcus aureus (56.34%) followed by Escherichia coli (22.55%), Streptococcus agalactiae (9.86%) and Bacillus spp. (8.45%). In conclusion, undesirable udder and teats conformations were more susceptible to intramammary infection and these traits must be considered accordingly while selecting dairy animals for future milk production. Observed high occurrence of subclinical Staphylococcal mastitis needs improvements in milking hygiene, mammary gland health, regular screening of animals, and use of biosecurity as an intervention tactic to limit the spread of pathogens at dairy farms.

Keywords: dairy cattle; mastitis; Udder, Teat, morphology

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Introduction

Udder and teat conformation traits in cattle are highly inheritable and can be categorized according to their shape as undesirable or desirable (Guarín *et al.,* 2017). Cows with undesirable shapes of udder and teats are more susceptible to injury and are in high risk of infection by pathogens responsible for mastitis Received: 09/12/2022 Accepted: 06/03/2023 Published: 29/03/2023

(Bhutto *et al.*, 2010; Singh *et al.*, 2014). Similarly, udder and teat conformation characters are related to resistance to intramammary infections (Klein *et al.*, 2005; Sharma *et al.*, 2011). Higher incidence of subclinical mastitis has been reported in cows with pendulous shaped udder than in trough-shaped udders (Uzmay *et al.*, 2003; Kamboj *et al.*, 2008). In addition, animals with smaller teat are more prone to mastitis than

animals with medium and large teats because the shorter teat allow microbes to move up without difficulty compared to animals with larger teat canal (Danish *et al.*, 2018). Therefore, udder and teat conformation qualities can be used for improving udder health (Nakov *et al.*, 2014).

Somatic cells, the milk-secreting epithelial cells from the lining of the gland, are response to injury or infection (Sharma et al., 2011). Increased Somatic cell count (SCC) in milk (including 75% of leukocytes and 25% of epithelial cells) is indicative of intramammary infection (Condas et al., 2017; Alhussien and Dang, 2018). Although, animal's age, stage of lactation, parity, seasons, stress and management practices cause variation in SCC (Alhussien and Dang, 2018; Kline et al., 2018), the presence of pathogens in the quarter is usually considered to be the main factor associated with the elevation of SCC in milk (Malik et al., 2018; Sumon et al., 2017). Somatic cell count in individual cow's milk or bulk milk is routinely used worldwide as direct indicator of udder health in dairy animals (Dufour and Dohoo, 2013; Stocco et al., 2020) and indirectly monitors the quality of milk (Alhussien et al., 2018). However, the normal composition of milk somatic cells varies with the type of secretion or lactation cycle. Normally, in a healthy mammary gland, the somatic cell count (SCC) in milk is lower than 1×10⁵ cells/mL, while during bacterial infection SCC can increase to above 1×10⁶ cells/mL (Bytyqi et al., 2010). More studies have revealed positive correlation between milk anatomical /morphological SCC with characteristics of teats and udder (Sharma et al., 2017; Guarín et al., 2017; Bhutto et al., 2010).

Association of udder morphology with the occurrence of mastitis has already been established worldwide (Bhutto et al., 2010; Klaas et al., 2004; Bharti et al., 2015). Nevertheless, there is paucity of information elucidating the association between the udder and teat morphology with intramammary infections in dairy cattle reared under the tropical condition, as compared to the currently available data for cows maintained on temperate conditions. In light of these facts, the objective of the present study was to evaluate the relationships between udder and conformations teat with intramammary infection in lactating dairy cows reared under tropical climate of Tanzania.

Materials and methods

Ethical clearance

Visual inspection of animals and collection of milk samples were performed in accordance with animal welfare guidelines without harming the animals. Farm data were collected with the prior consent of the farm manager.

Study design and location

A cross-sectional study was conducted to collect data from 243 lactating cows on four parastatal dairy farms in Tanzania (designated as farms A, B, C, and D). Farm A (Kitulo Livestock Multiplication Unit) is located at an altitude of 2630-2820 meters above sea level in the Makete district of the Njombe region. Kitulo farm has a semi-temperate climate with maximum and minimum temperatures of 4°C to 8°C and 14.5°C to 18.5°C, respectively. Kitulo area receives unimodal rainfall ranging from 1200 to 1600 mm per year. The wet season in Kitulo area begins in October and ends in May, followed by a cool, dry period that runs from June through August. Farms B (Mazimbu Dairy), C (Magadu Dairy) and D (Animal Research Unit) all belong to Sokoine University of Agriculture and are located in the same climate zone in Morogoro Municipality. Geographically, the municipality of Morogoro lies at an altitude of 500-600 meters above sea level and experiences a mixture of warm and cold temperatures ranging from 27-33.7 °C in the dry/warm season and 14.2-21.7 °C in cool/wet season. Morogoro municipal has a sub-humid tropical climate with a bimodal precipitation pattern (annual average of 600–900 mm) characterized by two precipitation seasons per year, with short rains (October to December) during the dry season. A moderate rainfall of 148 mm resulted in long rains (falling from March to May/June) with an average of 472 mm of precipitation (Meteorological Center, Sokoine Agricultural University). The drought peaks in September and lasts about six months.

Animal Management

In this study, two genotypes of dairy cattle were involved; pure Friesian cattle kept at farm A, and crossbred (crosses of *Bos taurus* and *Bos indicus*) cattle reared at farms B, C and D. Farm A was planted with temperate pastures consisting primarily of Lolium perenne, Lolium multiflurium, and *Infolium repens* in a well-maintained paddock and animals allowed to graze for 24 hours. Animals were given water ad libitum in pasture and during milking. Milking cows and heavily pregnant animals were given supplements twice a day (during milking hours) of farm-made concentrates comprising of maize and rice bran concentrates (60-70%), sunflower seed cake or cotton seed cake (25%), mineral supplements 2% and 1% salt. In farm B and C, animals were allowed to graze on mixture of established pasture (Cenchru sciliaris, Brachiaria brizantha, Chloris gayana and Panicum maximum) and natural pastures for about 8 hours and fed with hay or green fodder as well as ad libitum water after returning to the housing pallor in the afternoon. The floor of the house was made up of rough concrete and having an adequate slope for better drainage. The houses were cleaned twice a day, in the morning from 6.30 AM to 8.00 AM and evening from 3.30 to 5.00 PM. Lactating and heavily pregnant cows were supplemented with farm-made concentrate during milking hours to complete the nutrient requirements. Animals managed on farm D were grazed on natural pastures for 8 hours a day (9.00 am to 2.30 pm and 4.00 to 6.30 pm) with provision of *ad libitum* water in animal sheds. Only lactating and heavily pregnant cows were supplemented with farmmade concentrate during milking hours. Farms A, B and C practiced machine-milking twice a day during the morning from 6.00 to 8.00 AM and evening from 3.00 to 5.00 PM. While farm D used hand milking twice a day during 7.00 - 8 AM and evening from 3.00 to 5.00 pm. Animal in all farms were routinely vaccinated against common diseases (Brucellosis, Contagious Bovine Pleuropneumonia, Anthrax and Black Quarter diseases), regularly dewormed and dipped against external parasites.

Evaluation and classification of udder and teat conformation

Udder and teat conformations from 243 cows were evaluated through visual examination and categorized accordingly (Danish *et al.*, 2018; Basavaraj *et al.*, 2019). Udder conformation was evaluated and classified into four types: pendulous, round, goaty or desirable/normal udder. Similarly shape of teats was examined grossly and categorized as desirable; short; funnel-shaped; bottle-shaped and cone-shaped as per the visual appraisal method. In addition, the dimensions (teat length and teat diameter) were measured with Vernier calipers in each quarter of individual animal. Briefly, the teat length (in millimeters) was measured as the distance from the teat base to the end of the teat, and the teat diameter was measured at the central part of the teat.

Screening for Intramammary Infection

Presence of clinical or subclinical mastitis was revealed by examining for the cardinal signs of the disease. For clinal mastitis, animals were examined for presence of udder inflammation together with or without systemic reactions whereas the subclinical mastitis was checked using California Mastitis Test (CMT) protocol as previously described by Mpatswenumugabo et al., (2017). Briefly, milk samples (approximately 3 mL) from each quarter / teat were put in marked CMT paddle cups (marked A, B, C and D to correspond to individual teats/quarter) and mixed with equal amount (about 3 mL) of CMT solution. The paddle was rotated in a circular motion for about 10 seconds to mix the milk and CMT solution thoroughly. The test was read quickly, as the visible reaction of gel formation grading of sub-clinical and clinical mastitis. CMT score ranging from 0 to 4 was used, where 0 is negative result (no gel formation), 1 is traceable (possible infection), and 2 or 3 indicates a positive result and 4 has the thickest gel formation. A sample was defined as positive to SCM when one or more quarters with $CMT \ge 2+$ were detected.

Collection of milk samples

Proper history on the previous occurrence and treatment of mastitis was taken prior to milk sample collection for bacteriology. Before milk sample collection, the udder of cows was thoroughly washed using clean water, dried with clean towel and teats were sprayed with 70% ethanol. Few milliliters of milk (2-3 streaks) were poured off, and milk sample were aseptically collected in sterile bottles. Collected milk samples were portioned into two; one part for somatic cell count and second part for bacteriological culture. Milk samples for bacteriological culture were packed in cool box packed with ice packs to maintain cold chain environment while transported to the Microbiology laboratory at the College of Veterinary Medicine and Biomedical Sciences of Sokoine University of Agriculture (SUA).

Somatic Cell Count

Milk samples for SCC were prepared using the microscopic slides method as previously prescribed (Bharti et al., 2015). Briefly, dried milk smears were prepared using 10µL milk and stained with modified Newman-Lampert stain for 1-2 minutes, then the stain was drained and the smears were washed gently with tap water and dried again. Stained dry milk smears were examined under a microscope oil immersion lens (x100). Thirty different backgrounds were observed per smear and the average number of somatic cells per background was calculated. The average number of cells per background was then multiplied by the microscopic factor of the microscope which is 240807, to obtain the number of cells per milliliter of milk.

Bacteriological examination of milk samples

At the laboratory, samples were inoculated onto nutrient agar, blood agar, and Chapman's agar plates. Plates were incubated at 37°C under aerobic conditions for 24-48 hours before final observation. At least one colony forming unit (CFU) was required for the pathogens (Bacillus spp., Streptococcus agalactiae, Escherichia coli and Staphylococcus aureus) to be considered positive bacterial growth. Positive isolates were firstly characterized based on colony morphology, hemolytic properties, Gram reaction, catalase production, and biochemical examination. A catalase test was performed on Gram positive (+) isolates to distinguish between staphylococcal and streptococcal species. Catalase-negative tentatively identified colonies were as Streptococcus spp., while coagulase-positive colonies were regarded as *S. aureus. Bacillus* spp. were identified based on colony morphology and Gram staining. Gram (-) isolates were inoculated onto eosin methylene blue (EMB) agar and MacConkey agar and incubated at 37 °C for 24 h to identify *E. coli*. A greenish metallic isolate on EMB agar and a lactose-positive pink isolate were tentatively identified as *E. coli* after an IMVIC (indole, methyl red, Voges-Proskauer, Citrate) Reaction. Samples were said to be contaminated if three or more types of bacteria were isolated from one milk sample and no growth of the specified major pathogens was detected. Cultures were regarded negative if no growth was seen after 48 hours of incubation.

Statistical analysis

Information collected in this research were compiled and cleaned in Microsoft Excel® and imported into SPSS Statistical Package version 17 for analysis. Estimates of the prevalence of pathogens commonly isolated on dairy farms were determined using standard formulae (Number of positive animals/samples divided by total number of animals/samples tested). Descriptive statistics were calculated to determine the frequency of intramammary infection. A statistically significant relationship between variables was considered to be present if the p-value was less than 0.05. Figures were generated in Microsoft excel®.

Results

Udder and teat morphometrics

The occurrence of various udder and teat conformation in Friesian and crossbred cows kept in four farms under different management systems are as presented in Table 1.

Table 1. Frequency of different morphological types of udder in Friesian and crossbred dairy cattle

	Farm A		Fa	Farm B		Farm C		Farm D		Total	
Odder conformation types	AF	RF	AF	RF	AF	RF	AF	RF	AF	RF	
Desirable	76	60.8	45	67.2	26	76.5	13	76.4	160	65.8	
Pendulous	28	22.4	13	19.4	3	8.8	2	11.8	46	18.9	
Round	9	7.2	3	4.5	4	11.8	2	11.8	18	7.4	
Goaty	12	9.6	6	8.9	1	2.9	0	0	19	7.8	
TOTAL	125	100	67	100	34	100	17	100	243	100	

AF: Absolute Frequency; RF: Relative Frequency

The observation revealed high percentage of cows had desirable udder (65.8%) and teat (76.0%) shapes. Among the undesirable udder shapes, pendulous type udder had higher frequency (18.9%), followed with goaty (7.8%) and round (7.4%). The frequencies of undesirable teat shapes were 5.3%, 6.6%, 4.5% and 7.5% for short, funnel, bottle and conical shaped teats, respectively (Table 2). It was further observed that the percent occurrence of undesirable udder types was significantly higher (P<0.05) in farm A which had pure breed Friesian cows than in the other three farms keeping crossbred cows. Similarly, the study revealed that the frequencies of undesirable teat shapes was significantly higher (P<0.03) in machinery milked cows (32.4%) than in hand milked animals (17.6%). Among the machinery milked cows, pure breed (Friesian) had higher occurrence of undesirable teat shapes (38.2%) than crossbred cows (29.7%). The results of teat measurements (Table 2) showed variation in teat length from 49.0 mm to 72.0 mm with a mean of 49.31 ± 0.28 mm. The average teat diameter was 16.73 ± 0.15 mm, ranging from 13.0 mm to 28.5 mm. Pure Friesian cows had higher average teat length (52.84±0.36) and diameter (20.56±0.25) than 48.13±0.26 and 15.45±0.10 for average teat length and diameter, respectively, observed crossbreed cows.

Table 2. Classification of morphological shapes and biometrics of cow teats from four dairy farms

	Farm A		Fai	Farm B		Farm C		Farm D		otal
	AF	RF	AF	RF	AF	RF	AF	RF	AF	RF
Type of teats shape										
Desirable	373	74.6	209	78.0	101	74.3	56	82.4	739	76.0
Short	24	4.8	18	6.7	6	4.4	4	5.9	52	5.3
Funnel	34	6.8	16	6.0	12	8.8	2	2.9	64	6.6
Bottle	26	5.2	8	3.0	8	5.9	2	2.9	44	4.5
Cone	43	8.6	17	6.3	9	6.6	4	5.9	73	7.5
TOTAL	500	100	268	100	136	100	68	100	972	100
Teats biometrics										
Average Length	52.84±	0.36	48.42±	0.26	51.74±	0.36	44.24±	0.15	49.31±	0.28
(millimeter)										
Average diameter	20.56±	0.25	15.49±	0.09	15.87±	0.15	14.98±	0.07	16.73±	0.15
(millimeter)										

AF: Absolute Frequency; RF: Relative Frequency

Milk somatic cell count

Results showed (Figure 1) that there were significant differences (P<0.01) between cattle genotypes on SCC and the analysis revealed that the effects of intramammary infections were also statistically significant (P<0.01). The overall SCC means for normal udder, subclinical mastitis and clinical mastitis were 3.5 million cells, 4.7 million cells and 7.7 million cells, respectively. Further analysis revealed high correlation between SCC and udder/teat shapes. The mean \pm SE of SCC for

desirable, pendulous, round and goaty shaped udders was 3.110±0.024, 6.235±0.027, 4.962±0.021, and 4.728±0.014, respectively. Similarly, mean ± SE of SCC for desirable, short, funnel, bottle and conical-shaped teats was 3.189±0.105, 6.850±0.068, 5.989±0.195, 4.899±0.168, and 5.030±0.145, respectively. Furthermore, analysis showed a significantly (p<0.01) positive correlation between SCC and teat diameter as well as teat length with correlation coefficient of 0.443 and 0.419, respectively.



SCM = subclinical mastitis Figure 1. Relationship between SCC and intramammary infection status in cows kept in four dairy farms.

Intramammary infection

Nine hundred and seventy-two quarters from 243 lactating dairy cows (125 Friesian and 118 crossbred) were screened for the presence of mastitis in the study. Out of the 972 quarters examined, normal, clinical and sub-clinical abnormalities were detected in 605 (62.24%), 12 (1.23%) and 355 (36.52%), respectively. The prevalence of subclinical mastitis in cows at farm A, B, C and D were 42.8%, 35.07%, 33.09% and

2.94%, respectively (Figure 2); and there was a significant difference (P<0.01) in the prevalence between machine milked farms (A, B & C) and hand milked animals (farm D) (Figure 1). Most of subclinical mastitis cases were observed affecting one quarter (30.06%) followed with two quarters (24.18%) and, three and four quarters (22.88%) (Table 3).



SCM = subclinical mastitis Figure 2. The intramammary status of cows in relation to prevalence of mastitis in four dairy farms

Farm	Total number of cows affected				
	One quarter	Two quarters	Three quarters	Four quarters	
Farm A	24	11	8	6	49
Farm B	4	3	4	3	14
Farm C	3	1	1	2	7
Farm D	0	0	0	1	1
TOTAL	31 (43.66%)	15 (21.13%)	13 (18.31%)	12 (16.90%)	71 (100%)

Table 3. Distribution of intramammary infection in cows' quarters in the four herds

The most prevalent microorganisms isolated from the intramammary infected animals (clinical and subclinical mastitis) included *Staphylococcus aureus* (56.34%), *Escherichia coli* (22.55%), *Streptococcus agalactiae* (9.86%), *Bacillus* spp. (8.45%), and none typed microorganisms (2.82%) as shown in Table 4.

Table 4.	Frequency	of bacterial	isolates	from a	cases of	^c mastitis	in	four	dairy	farms
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	-		Number	Number of isolates				
Bacterial isolates						isolates		
	Farm A	Farm B	Farm C	Farm D	Total	-		
<i>Bacillus</i> spp.	4	1	1	0	6	8.45		
Escherichia coli	8	5	2	1	16	22.55		
Staphylococcus aureus	28	8	4	0	40	56.34		
Streptococcus agalactiae	7	0	0	0	7	9.86		
Others	2	0	0	0	2	2.82		

Relationship between udder/teat Morphometry and incidence of intramammary infection

The udder and teats are the first line of defense against intramammary infection. Among various shape of the udder examined in the present study, pendulous shaped udder was having highest incidence of mastitis (76.10%) followed by round (50.0%) and goaty (47.37%) shaped udder (Table 5). Similarly, the present study revealed that teat shapes may be a risk factor for intramammary infection as we found higher incidence of mastitis in undesirable teats (73.08%, 50.0%, 43.75% and 41.10%) in short, funnel, bottle and conical shaped teats, respectively) than in desirable teat shapes (Table 5). Furthermore, our results indicated an increase in the degree of intramammary infection with a decrease in teat length and teat diameter as log SCC and mastitis were found to be significantly positively correlated with teat morphometry.

Table 5. Incidence of mastitis in dairy cows with respect to different udder and teat morphology

Type of udder and teat	Farm A	Farm B	Farm C	Farm D	Total
morphological shape	% (n)	% (n)	% (n)	% (n)	% (n)
UDDER					
Desirable udder	14.47% (11)	8.87% (4)	11.54% (3)	0 (0)	11.25% (18)
Pendulous udder	85.71% (25)	53.85% (7)	66.67% (2)	50% (1)	76.10% (35)
Round udder	66.67% (6)	33.33% (1)	50% (2)	0 (0)	50% (9)
Goaty udder	58.33% (7)	33.33% (2)	0 (0)	0 (0)	47.37% (9)
TEATS					
Desirable teat	5.90% (22)	2.39% (5)	1.98% (2)	0 (0)	3.92% (29)
Short teat	75.0% (18)	55.56% (10)	50.0% (3)	100% (4)	73.08% (35)
Funnel teat	52.94% (18)	43.75% (7)	25.0% (3)	0 (0)	43.75% (28)

Bottle teat	57.69% (15)	62.5% (5)	25.00% (2)	0 (0)	50.00% (22)
Conical teat	48.84% (21)	41.18% (7)	22.22% (2)	0 (0)	41.10% (30)

Discussion

Intramammary infection is an imperative menace affecting the dairy sector with udder and teat conformation being a risk factors for intramammary infection (Bhutto et al., 2010; Singh et al., 2014). Cows with undesirable shaped udders and teats are more susceptible to lesions and contamination by mastitis-causing pathogens, which increase the risk of mastitis (Singh et al., 2014). Therefore, the udder conformation traits with strong arguments can be used to improve udder health (Nakov et al., 2014). In this study, the overall frequencies of desirable (65.8%), pendulous (18.9%), goaty (7.8%), and round shaped udder (7.4%) were recorded. The recorded undesirable udder shapes (pendulous, goaty and round shaped udder) are in agreement with the findings reported elsewhere (Modh et al., 2017; Basavaraj et al., 2019). Teat shape plays an important role in milk flow of milk from the udder, which also helps in selection of high milk vielding cattle and the different shapes of the teat might be its genetic heritability. In the present study, teats from examined animals exhibited the following frequencies: 74.6% (373/500) teats of desirable shape, 7.5% (73/500) conical-shaped teats, 6.6% (64/500) funnel-shaped teats, 5.3% (52/500) short teats, and 4.5% (44/500) bottleshaped teats. These data are consistent with the findings reported by Okano et al., (2015) in Holstein cows in two dairy farms in Brazil. However, our findings are different from those observed in dairy cattle kept in United Kingdom (Rathore, 1976), India (Bharti et al., 2015) and Bangladesh (Islam et al., 2020). The dimension (length and diameter) of teats recorded in this study ranged from 44.24±0.15 to 52.84±0.36 and 14.98±0.07 to 20.56±0.25 in pure Friesian and crossbreed cows, respectively. Pure Friesian cows had longer (length) and larger (diameter) teats than crossbreed cows. Observed teat measurements from crossbred cows are comparable with findings reported by Islam et al., (2020) in crossbred dairy cows in Bangladesh. Similarly, Friesian teat dimension findings reported here are in line with other studied conducted previously in the different parts of the world including Turkish Holstein-Friesian farms

(Bardakcioglu *et al.*, 2011) and Belgian Holstein-Friesian cows (Zwertvaegher *et al.*, 2012). Nevertheless, teat diameters are dependents of the breed of cows (Zwertvaegher *et al.*, 2011), parity (Seker *et al.*, 2009), stage of lactation (Tilki *et al.*, 2005) and quarter position (Zwertvaegher *et al.*, 2012)

Somatic cells are part of the udder's innate immune system and consist of 75-85% white blood cells (macrophages, polymorphonuclear neutrophils (PMNs), lymphocytes) and 15-25% epithelial cells (Barrett, 2002). Determining the number somatic cells in milk is internationally recognized as the gold standard for diagnosing subclinical mastitis in ruminants and humans (Hunt et al., 2013; Bharti et al., 2017; Malik et al., 2018). This study observed higher level of somatic cell count in pure breed Friesian cattle than in crossbreeds. Similarly, it has been reported that high-milk producing cattle breeds such as Brown Swiss and Holstein Friesian have a higher presence of SCC/mL in milk than crossbreed cow breeds (Alhussien et al., 2016; Alhussien and Dang, 2018). Further analysis vielded higher level of somatic cell count in undesirable udder and teats in comparison to desirable shaped udder teats. Several researchers have reported SCC variation with respect to various breeds of cows (Ahlawat et al., 2008; Koc and Kizilkaya, 2009; Alhussien and Dang, 2018). The relationship of the teat conformation and size on SCC observed in the current study is similar to report of Sharma et al., (2016) that SCC in milk is usually higher in short teats with a larger width of teat canal. However, other researchers have not found the relationship of udder and teat shapes and dimensions on SCC (Coban et al., 2009; Orban et al., 2009). Other researchers had also stated a significant impact of udder shape and reported that cows with pendulous udders have higher SCC compared to regular udder shapes (Ahlawat et al., 2008). Correspondingly, this study revealed a good correlation between intramammary infection and teat dimensions (length and diameter). Several researches have reported similar trend of presence of higher value

of somatic cell counts in shorter teats (Nemcova *et al.*, 2007) and thin teat diameter (Orban *et al.*, 2009). Nevertheless, others have reported no effects of teat length and thickness on SCC (Juozaitiene *et al.*, 2006). Other researchers have however reported that long and thick teats are potential risk factor for occurrence of intramammary infection in cows (Haghkhah *et al.*, 2011; Singh *et al.*, 2014; Bharti *et al.*, 2015).

This study also reported that, the quarter level prevalence for subclinical mastitis was 30.06%, 24.18% and 22.88% for one guarter, two guarters, and three and four quarters, respectively. These findings are almost similar to that reported in Uruguay (Gianneechini et al., 2002), Zimbabwe (Katsande et al., 2013) and Bangladesh (Sumon et al., 2017). However, quarter SCM prevalence of 16.90% to 43.66% are higher than previously reported in Riverine buffaloes (Gagandeep et al., 2018) and Dairy cattle (Milesa et al., 2019). Difference in SCM between herds can be associated with farmers' awareness about the disease and adaption toward the use different control measures for maintaining good hygiene around animals' udder and teats. Several researchers have highlighted udder and teat shape as risk factors for intramammary infection in dairy cows. This is because cows with undesirable udder shapes and long, thick teats are more prone to lesions and contamination with mastitis-causing organisms, increasing the risk of mastitis (Bharti et al., 2015; Bhutto et al., 2010; Singh et al., 2014). Results regarding the occurrence of intramammary infection in relation to udder and teat morphometrics reported here are comparable to those observed elsewhere (Kamboj et al., 2008; Uzmay et al., 2003; George et al., 2007; Danish et al., 2018). It is hypothesized that animals with pendulous udder have positive correlation with high incidence of mastitis because long and pendulous udders get in contact with ground more frequently causing injuries and thus predisposing the animals to pathogens that are associated with mastitis.

Bovine mastitis is one of the main diseases affecting dairy herds worldwide and inadequate milking practices or poorly maintained milking equipment (Capuco *et al.,* 1994; Schukken *et al.,* 2003) are among the risk factors for the occurrence of this intramammary infection. In

this study, the prevalence of subclinical mastitis was higher (farms average of 36.99%) in machinery milked cows than in hand milked cows (2.94%). Similar findings have been reported in Cuba (Ruiz et al., 2014) and Dutch dairy herds (Barkema et al., 1999). The milking machine, especially when not functioning properly, is implicated in the occurrence of mastitis by transmitting infections between cows or quarters, or adversely affecting udder health through damaging or changing the resistance of the cow's first line of defense. In addition, it is hypothesed that milking machine that lack enough vacuum, inappropriate milking practices, poor udder cleaning prior to milking are among of risk factors of occurrence of mastitis (Oliveria et al., 2015).

Most of the intramammary infections were caused by Staphylococcus aureus (56.34%) which suggests that the prevention of spread of this contagious bacterium during milking was not effective. High prevalence of S. aureus in mastitic cattle has been reported previously in Tanzania (Mdegela et al., 2004; Mdegela et al., 2005; Mdegela et al., 2009; Kashoma et al., 2015), Rwanda (Mpatswenumugabo et al., 2017; Ndahetuve et al., 2020), Algeria (Saidi et al., 2013), Italy (Moroni et al., 2006), Brazil (Dittmann et al., 2017), Morocco (Bendahou et al., 2008), Ethiopia (Daka et al., 2012; Zeryehun and Abera, 2012) and (Shitandi 2004). Kenva and Sternesjö, Stapphylococcus aureus are the most prevalent pathogen on the skin of udder of clinically healthy cattle and animal environment (Kashoma et al., 2015), and has the capacity to penetrate into the tissue, producing deep seated foci protected by a tissue barrier (Barkema et al., 2009; Ranjan et al., 2010; Rall et al., 2013). The reported high frequency of staphylococcal mastitis is considered to be due to poor milking hygiene and lack of proper attention to the health of the mammary gland in general. Thus, hygiene at milking is of paramount importance in control of these infections because the bacteria are likely to be spread during the milking process. In this study, the result of isolation of Escherichia coli was almost similar to that reported elsewhere (Kivaria et al., 2007; Mpatswenumugabo et al., 2017) but lower than that reported in Bangladesh (Kavesh et al., 2014; Sumon et al., 2017). Variations in E. coli isolation may be attributable to poor

cleanliness, drainage and manure disposal in farms as well as poor milking practice. Even though contagious *Streptococcus agalactiae* (9.86%) were ranked in third position in the present study, the bacteria have been stated as the most famous bacteria in cow mastitis in some countries (Hegde et al., 2013; Östensson et al., 2013). The prevalence rate of S. agalactiae revealed in the present study agrees with preceding findings observed elsewhere (Mdegela et al., 2009; Persson et al., 2011). Bacillus spp., were present in 8.45% of cases in the present study. Bacillus spp. has also been recognized as important bacteria in both CM and SCM in earlier studies (Nieminen et al., 2007; Amer et al., 2018). However, these findings disagree with other studies showing that mastitis caused by *Bacillus* spp is uncommon in dairy cows (Sori et al., 2005; Abera et al., 2012). The presence of Bacillus spp. could be associated with poor hygienic conditions of milkers as the bacteria are broadly dispersed in dairy environment, including on teat skin, milkers' skin, and farm floors, which signify reservoirs of bacteria related with intramammary infections (Amer et al., 2018; De Visscher et al., 2014).

Conclusion

The present study revealed the association between some udder and teat morphometric traits and poor udder health in lactating dairy

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cows. Most of cows with pendulous udder and funnel shaped teats were more susceptible to intramammary infections. Among the animals, Friesian cows which were machinery-milked cows had high prevalence of undesirable udder and teat shapes as well as intramammary infection than cross-bred cows maintained under hand milking management. High prevalence of contagious Staphylococcus aureus in mastitic cattle was among of observation. Therefore, udder and teat conformation traits should be considered will selecting animals for milk production. Similarly, high prevalence of Staphylococcus aureus, the contagious pathogens, and well spread via milking machine components, hands of milking personnel, and through washcloths, thus calls for hygienic improvement during milking practices.

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