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REGULAR ARTICLES

Prevalence of bovine subclinical mastitis and antibiotic susceptibility patterns of major mastitis pathogens isolated in Unguja island of Zanzibar, Tanzania

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Abstract A cross-sectional study was conducted between January and July 2014 in Unguja island of Zanzibar to establish prevalence of subclinical mastitis (SCM) in smallholder dairy cows and patterns of antibacterial susceptibility of major mastitis pathogens isolated. A total of 416 dairy cows from 201 farmers were randomly selected from three districts of Unguja Island to participate in the study. Questionnaire interview, field observation, individual cow examination, California Mastitis Test (CMT) and bacteriological examination were carried out. Kirby-Bauer disc diffusion technique was used to test drug sensitivity for common bacteria isolated. Based on CMT results, the overall prevalence of SCM was 28.6, 48.8 and 64.7% at quarter, cow and farm level, respectively. Prevalence of bacterial infection was recorded at 42.9, 70.9 and 78.6% at quarter, cow and farm examined, respectively. The common bacteria isolated included *Staphylococcus aureus* (36.8%), *Pseudomonas aeruginosa* (17.8%), *Staphylococcus epidermidis* (16.1%), *Klebsiella* spp. (9.5%), *Micrococcus* spp. (6.3%) and *Escherichia coli* (4.9%). In conclusion, findings of this study demonstrated high level of subclinical mastitis at farms, cows and quarters levels with both contagious and environmental bacterial pathogen involved. Therefore, efforts should be directed to the decreased subclinical mastitis by improving sanitary measures and proper milking practice.

Keywords CMT · Intra-mammary infection · Teat injury · Drug resistance · Hygiene

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Introduction

Bovine mastitis is the most important disease in dairy industry around the world (Girma et al. 2012) which can be classified into contagious and environmental forms (Bradley 2002). Contagious mastitis is caused by pathogens which can cause infection and can be transmitted between animals. Environmental mastitis is caused by opportunistic pathogens living within cow's environment that can invade udder through teat orifices when proper milking techniques are not observed. Based on the clinical manifestation of affected cows, mastitis could also be classified as either clinical mastitis (CM) or subclinical mastitis (SCM) (Radostits et al. 2000). Clinical mastitis is characterised by physical changes such as swelling, hotness, pain and indurations of the udder or chemical changes which include discolouration, presence of clots in the milk of infected quarter which can be easily observed by farmer. In contrary, SCM cannot be physically observed but can be detected by increased Somatic Cell Counts (SCC) to more than 200,000 cells/ml or by isolation of microbes that caused the disease.

Economic losses from CM include reduced milk production, increased treatment costs, milk discarded and culling of chronic cases. These can be observed easily by the farmer compared to SCM. In SCM a farmer cannot realise the economic loss, which is relatively higher due to the prolonged decrease of milk produced from individual cow, together with

the decreased quality of produced milk as a result of increased number of both somatic cell count and pathogens (Halasa et al. 2009). The public health risk associated with bovine mastitis cannot be under estimated where antibiotics are used, especially when the withdrawal period is not observed. In addition, a wide range of pathogens can be harmful to human when raw milk is consumed (Jones 2009).

Occurrence of both types of CM and SCM has been previously demonstrated in different studies conducted in other parts of Tanzania (Swai et al. 2006; Karimuribo et al. 2006) with high prevalence of SCM compared to CM. In those studies, different species of mastitis pathogens were isolated and various risk factors associated with the occurrence of both CM and SCM were identified. Quarter level prevalence of SCM was previously described by Suleiman et al. (2013) in Pemba island showing a high prevalence of 34.4 and 35.7% as defined by CMT and bacteria isolation respectively.

Most of the time, treatment of mastitis begins before knowing the causative agent involved or without proper antibiotic testing. This may lead to the use of antibiotics which are not effective or the use of antibiotics in cases where bacteria are not involved as causative agents. Improper use of antibiotics may increase the percentage of resistance among bacteria in both animal and human populations (Padol et al. 2015). Although antibiotic resistance in bovine mastitis pathogens has been reported elsewhere (Awandkar et al. 2013), similar work has never been conducted in Zanzibar. The current cross-sectional study was therefore designed to (a) investigate the occurrence of SCM in Unguja Island, (b) identify important risk factors associated with SCM, (c) isolate common bacterial mastitis pathogen and establish their antimicrobial susceptibility profiles.

Material and methods

Study area and sample size

The Unguja Island lies between latitudes 05° 72″ and 06° 48″ South of Equator and longitude 39° 30″ and 39° 51″ East of Greenwich about 45 km from the east cost of Tanzania Mainland. The island has a tropical climate with temperature ranging from 24 to 32 °C, experiencing two rainy seasons in March to May and September to November and two dry seasons of November to March and May to September. This study used the formula derived from Bennett et al. (1991) to estimate disease proportion using cluster sampling as shown.

$$n = \frac{p(1-p)Dz^2}{e^2 b}$$

Where n is a sample size (number of households); p = estimated prevalence of SCM at farm level in previous study which was 0.67 according to Suleiman et al. (2013); e^2 is precision which is equal to absolute estimated error at 5% (0.05); b is number of lactating dairy cows to be sampled from each household (2); z is a confidence level at 95% (1.96); D = design effect which can be calculated using the following formula D = 1 + (b-) roh = 1.2; roh is a rate of homogeneity which was estimated at 0.2. A total of 201 dairy farmers were selected to participate in the study.

Sampling technique and sample frame

A multi-stage sampling procedure was adopted to select participating dairy farmers. Initially, purposive sampling of three out of six districts of Unguja Island based on the population of dairy cattle in the districts was made. The selected districts included North B, West and Central. In the second stage, a total of 28 *Shehias* (lowest administrative level of local government authority in Zanzibar) were randomly selected from a list of 59 *Shehias* with dairy farming activities. In the third stage, a total of 201 dairy farming households were randomly selected from an established sampling frame of all dairy farmers in the selected *Shehia* using a simple random selection procedure. For the dairy farmer to be included in the sample frame, the only criterion was the possession of at least one lactating dairy cow. Finally, each household was considered as a cluster and all lactating dairy cows were included where 416 lactating dairy cows were selected.

Data collection procedure

Each participating household was interviewed by using a simple structured questionnaire. The aim of this interview was to collect information about the farmers' awareness, knowledge and practices, and compare them with the outcome variable of occurrence of SCM in dairy cows. Field observation was used to assess environmental condition, management procedure and individual cow assessment in terms of hygiene and physical soundness in relation to the occurrence of mastitis where general condition of cow, udder and individual teats were assessed for observable clinical signs of mastitis. Several farm and cow risk factors were collected during the field visit, among them were use of calf for milk let-down during milking (yes, no), hygienic condition of the drainage system (dirty, clean), use of towels to dry the teats before milking (yes, no), characteristic of dairy farmers to wash hands before milking (yes, no), number of animals in the farm, lactating stage of each sampled cow, amount of milk produced in litres, number of milkings per day (twice, once), number of parturition, water availability for cleaning (yes, no), milking technique used if teat stripped or hand fist. All the observed risk factors were compared with the occurrence of SCM based on CMT or bacteriology results.

Cow side and laboratory test

California Mastitis Test (CMT) was used to screen lactating cows for SCM. In this test one or two strips of milk (about 2 mls) from each teat was milked directly into the CMT pad after discarding the first two strips. Each milk sample in the CMT pad was mixed with equal amount of CMT reagent and the result recorded according to the standard procedure (Varatanovic et al. 2010) depending on the reaction formed. Quarter milk samples from both CMT positive and negative quarters were aseptically collected directly from each teat into a 20-ml sterile and unique labelled universal bottle according to the procedure described by Quinn et al. (1994). The samples were stored in a cool box containing ice pack with a temperature of 4 °C and transported to the Maruhubi Veterinary Investigation Centre for bacteria isolation and identification. In the laboratory, the samples were cultured on the same day or deep frozen at − 20 °C waiting for culture within 72 h. Each sample was cultured in duplicate using MacConkey and blood agar, one set incubated aerobically and another an-aerobically. Bacterial growth was observed after 24 and 48 h where if no growth observed, the quarter was considered negative. For positive growth, colony morphology, pigmentation and haemolytic reaction were observed. Gram stain was used to distinguish between gram positive and negative bacteria and to study the microscopical features of the isolated bacteria. Different biochemical tests as described by Vashist et al. 2013a were used to identify genus and species of isolated bacteria. Modified oxidase test was used to differentiate between gram positive cocci bacteria, those with positive results were identified as *Micrococcus* spp. while those with negative results were subjected to catalase and coagulase tests, isolates with positive results in both tests were identified as *S. aureus*. Isolates that tested positive to catalase and negative to coagulase were subjected to oxidase test and those with negative results were confirmed to be *S. epidermidis* (non-haemolytic) or *S. haemolyticus* (haemolytic). Rod shape gram positive bacteria were exposed to catalase and oxidase tests, negative results for both tests confirmed the presence of *Trueperella pyogenes* while isolates that were positive to catalase and urease were identified as *Corynebacterium* spp. Based on culture characteristics on MacConkey agar, Gram negative bacteria were categorised into lactose fermenters with pink colonies and non-lactose fermenters (colourless colonies). Lactose fermenters were subjected to catalase, urease, indole tests and citrate fermentation, isolates with positive results for catalase and indole with negative results for urease and citrate fermentation were identified as *E. coli* and those with citrate fermentation, catalase and urease positive and indole negative were identified as *Klebsiella* spp. Non-lactose fermenters were subjected to catalase and oxidase tests, isolates with positive results to both tests were identified as *Pseudomonas aeruginosa*, isolates with catalase positive and oxidase negative were identified as *Proteus* spp.

Testing for antibiotic sensitivity

A total of eight antibiotics belonging to four groups were used to assess susceptibility profiles of major mastitis bacteria isolated. The antibiotic selection was based on their availability in the local market and those commonly used to treat mastitis in Zanzibar. Susceptibility was tested using Kirby-Bauer disc diffusion technique as described by Vashistet al. 2013b. A portion of pure colony of bacteria isolated was transferred in a tube with 5 ml of nutrient broth and incubated at 37 °C for 24 h. The mixture was then transferred into a Muller Hinton agar plate and spread using sterile glass tube. The antibiotic discs were then placed on the plate using sterile forceps and incubated for 24 h at 37 °C. The antibiotic discs used, their concentration and used breakpoints are shown in Table 1.

Data processing and analysis

Data storage and analysis was done using Epinfo® version 7 for window where descriptive statistic and logistic regression analysis was performed. Graphic presentation was prepared in Microsoft Excel 2007. The outcome variable in this study was the prevalence of SCM as defined by CMT or bacteria culture at farm, cow and quarter levels. A cow's quarter was considered positive for CMT if the score was positive one and above while a cow was regarded positive if at least one quarter was CMT positive, where as a farm was considered positive if at least one cow in that farm was CMT positive. Based on culture results, a quarter was considered culture positive if bacteria were isolated from the sample collected while a cow was considered culture positive if bacteria were isolated from at least one quarter, and a farm was positive if at least one cow from that farm was positive. Tests for significance of proportions as defined by SCM prevalence in different categorical variables were performed using chi square (x^2) test. Simple regression analysis was used to screen categorical and explanatory variables that significantly influenced the occurrence of SCM as defined by CMT and Bacteria isolation. Variables that scored P value equal to or less than 0.2 during simple regression (14 and 19 risk factors as defined for CMT and bacteria isolation respectively) were forwarded to multiple regression analysis and only variable with P value ≤ 0.05 were included in the final model of risk factors that influenced the prevalence of SCM.

Table 1 Concentration and susceptibility break points of antimicrobial used in antimicrobial sensitivity testing of major bacterial pathogen isolated from subclinical mastitis

Antibacterial	Concentration	Bacteria group	Inhibition zone in diameter (mm) for sensitivity disks		
			Resistant	Intermediate	Susceptible
Amoxicillin	30 μg	Staphylococci	≤ 19		≥ 20
		Other bacteria	≤ 13	14–17	≥ 18
Cephalexin	30 μg	All	≤ 14	15–17	≥ 18
Gentamycin	10 μg	All	≤ 12	13–14	≥ 15
Kanamycin	30 μg	All	≤ 13	14–17	≥ 18
Neomycin	30 μg	All	≤ 12	13–16	≥ 17
Tetracycline	30 μg	All	≤ 14	15–18	≥ 19
Penicillin G	2 IU	Staphylococci	≤ 28		≥ 29
		Other bacteria	≤ 14		≥ 15
Streptomycin	10 μg	All	≤ 11	12–14	≥ 15

Derived from provided datasheets attached to the sensitivity kits from HiMedia laboratory PVT ltd, Mumbai India

Results

Screening for mastitis

Most farmers (62.6%) were aware about clinical mastitis and its clinical manifestation, 39.4% of the interviewed farmers reported having at least one clinical case of bovine mastitis in their farm for the period of 1 year before the study but none of the farmers had knowledge about subclinical mastitis. A total number of 1664 quarters were physically screened for CM and related lesions, out of which 16 (1%) were blind, 14 (0.8%) were injured and 1634 (98.2%) were normal. Milk samples from 1648 quarters were subjected to CMT screening and laboratory culture. No milk samples were obtained from blind teats.

Prevalence of subclinical mastitis based on CMT and bacteria isolation

The quarter level prevalence of SCM as defined by CMT and bacteria isolation showed an overall prevalence of 28.6 and 42.9% for CMT and bacteria isolation respectively. There was no statistical difference in the prevalence among quarters based on their position ($P > 0.05$). Quarter level prevalence for both CMT and bacteria isolation were significantly higher in the Central district compared to the West and North B districts ($P < 0.05$) as shown in Table 2. SCM prevalence at cow and farm level in each study district is shown in Table 2 with an overall prevalence of 48.8 and 64.7% for CMT and 70.9 and 78.6% for bacteria isolation at cow and farm level, respectively. During this study, bacteria were isolated from 75.6% out of 472 milk samples that tested positive to CMT and 28.8% out of 1176 milk samples that tested negative for CMT. The results described that it was more likely to isolate bacteria from CMT positive compared to CMT negative (OR = 7.3, 95% CI = 5.7–9.4). Although 50% of injured teats were CMT positive compared to 28.5% normal teats and bacteria were isolated from 57.1% of injured teats compared to 42.8% of normal quarters but the difference was not significant in both scenarios ($P = 0.07$ and 0.27) for CMT and bacteria isolation respectively.

Bacteria isolated

Out of 1648 sample cultured, 35.4% yielded a single bacteria species, 7.5% produced two bacteria species, 1.1% samples yielded more than two bacteria species which were considered as contaminants and were not included in the final positive results. A total of 19 species of bacteria were isolated, the majority of which were aerobic as shown in Fig. 1. Antibiotic susceptibility patterns of each of the common bacteria isolated in this study are shown in Table 3.

Potential risk factors associated with SCM

As shown in Table 4, the final model of categorical and continuous variables associated with the occurrence of SCM as defined by CMT at quarter level included water availability, use of calf for milk let-down, lactation stage and herd size. Based on bacteria isolation, the risk factors included drainage, milking per day, milking technique, uses of towel, hand wash before milking, number of parity and milk produced.

Discussion

The prevalence of SCM as described by both methods of CMT and bacteria culture at farm, cow and quarter levels were high. The high prevalence of SCM within East African region were also described in previous studies conducted before

Table 2 Prevalence of SCM at quarter, cow and farm level based on CMT and bacteria isolation in each of the study district

	North B *n* (%)	Central *n* (%)	West *n* (%)	*P* value
CMT screening				
Quarter level	73/288 (25.3)	169/496 (34.1)	230/864 (26.6)	0.005
Cow level	36/73 (49.3)	67/125 (53.6)	100/218 (45.9)	0.385
Farm level	29/43 (67.4)	44/67 (65.7)	57/91 (62.6)	0.844
Bacteria isolation				
Quarter level	135/288 (46.9)	277/496 (55.8)	295/864 (34.1)	0.000
Cow level	48/73 (65.8)	117/125 (93.6)	130/218 (59.6)	0.000
Farm level	29/43 (67.4)	59/67 (88.1)	70/91 (76.9)	0.371

(Abrahmsen et al. 2014; Nkoroi and Maitho 2014). SCM prevalence was significantly higher in the Central district compared to the West and North B districts (Table 2). Poor sanitary measures in the Central district as demonstrated before (Suleiman et al., 2016) could explain such higher prevalence. Findings of this study indicated that it is more likely to isolate bacteria from CMT positive cases of bovine subclinical mastitis compared to CMT negative (OR = 7.3). CMT positive results without bacteria isolation observed may be due to the presence of other microorganisms rather than bacteria that trigger cow immunological response and hence increased number of somatic cell counts in the milk samples, pathogens like fungi, algae, mycoplasma, mycobacteria etc. that require special culturing media and methods. This may also be explained by the occurrence of infections of short term nature that were already cleared during samples collection depending on cow immunological status as observed before (Abdel-Rady and Sayed 2009). While CMT negative samples that harboured bacteria may be explained by the presence of bacteria that do not trigger the immunological response of the infected quarter resulting in a limited increase in the somatic cells count (Gitau et al. 2014). This is what can be identified as intra-mammary infection (IMI) where bacteria were isolated without somatic cell count change of the infected quarter (Mdegela et al. 2009). Likewise, contaminants occurring in the milk samples despite hygienic precautions may lead to false positive detections. Teat injuries increased the risk of quarters to be infected with SCM for about two and half folds (RR = 2.49). Presence of wounds near the teat orifice may be accompanied with the presence of opportunistic bacteria nearby that may invade the teat canal during or immediately after milking. This was also described by Madut et al. 2009.

The predominant bacteria species isolated from this study was *Staphylococcus aureus* with making about36.8% of the bacteria isolated; similar results were also reported by other researchers (Ondiek et al. 2013; Elbably et al. 2013). The majority of bacteria species isolated in this study were also described in other studies conducted in Tanzania or other East African countries. Very few samples (0.1%) harboured *Clostridium* spp. as a potential cause of SCM. Involvement of *Clostridium* spp. was also mentioned in other part of Africa (Osman et al. 2009; Odongo et al. 2012). In contrast to what

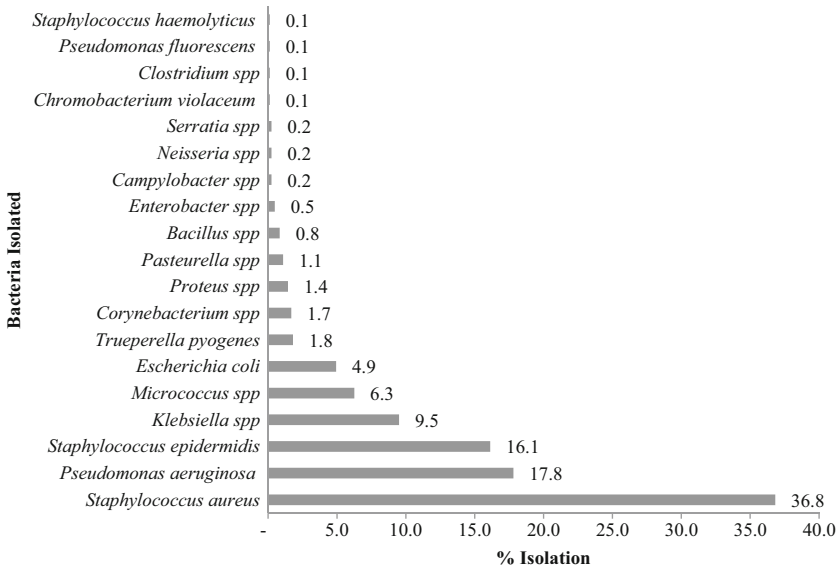

Fig. 1 Bacteria species isolated from milk samples tested (n = 831)

Table 3 Patterns of antimicrobial resistance of major bacteria isolated from quarters with subclinical mastitis

Antimicrobial agent	Percentage of resistance and sensitivity													
	E. coli (*n* = 13)		*Klebsiella* spp. (*n* = 57)		*Micrococcus* spp. (*n* = 35)		*P. aeruginosa* (*n* = 82)		*S. aureus* (*n* = 217)		*S. epidermidis* (*n* = 58)		*T. pyogenes* (*n* = 14)	
	R	S	R	S	R	S	R	S	R	S	R	S	R	S
Amoxicillin	92.3	7.7	50.9	42.1	40.0	60.0	70.7	22.0	47.0	48.8	29.3	65.5	42.9	57.1
Cephalexin	69.2	30.8	21.1	63.2	40.0	51.4	54.9	36.6	29.5	65.4	3.4	87.9	42.9	57.1
Gentamycin	15.4	84.6	0	100.0	2.9	94.3	2.4	86.6	5.1	90.3	1.7	94.8	0	100.0
Kanamycin	53.8	0	1.8	75.4	37.1	51.4	32.9	32.9	11.1	65.4	8.6	87.9	0	57.1
Neomycin	0	69.2	7.0	59.6	14.3	48.6	31.7	32.9	10.1	65.0	10.3	81.0	0	28.6
Penicillin G	100.0	0	100.0	0	74.3	20.0	91.5	8.5	88.0	9.2	70.7	27.6	100.0	0
Streptomycin	15.4	46.2	17.5	75.4	28.6	60.0	30.5	59.8	17.5	74.7	6.9	89.7	0	100.0
Tetracycline	30.8	46.2	3.5	89.5	28.6	48.6	26.8	51.2	16.6	71.4	19.0	79.3	0	71.4

R resistance, *S* sensitive, percentage of isolate with intermediate results is not shown in this table

has been demonstrated in other studies (Mdegela et al. 2009; Nkoroi and Maitho 2014), this study could not manage to demonstrate the involvement of *Streptococcus* spp. in the occurrence of SCM.

During this study, more risk factors were significant when defining SCM by bacteria culture compared to CMT screening. The SCM prevalence was higher in barns with dirty drainage system (OR = 1.21) because both, environmental and contagious bacteria have favourable conditions to hide waiting to invade the uninfected udder (Abunna et al. 2013). Water availability plays a vital role in the cleaning of barn, milking area, cow udder and hands before milking and hence the prevalence of SCM decreased with water availability (P = 0.007). Proper cleaning of hand before milking have positive effects on reducing the prevalence of SCM (OR = 0.56), the quantity of opportunistic bacteria which may invade the udder when proper milking hygiene is not practiced (Shittu et al. 2012). Although all farmers participated in the study use hand milking but the way they milk their cows have significant effect on the increased prevalence of SCM (OR = 0.63). The prevalence seems to be lower in farms used stripped method of milking compared to those who used hand fist. Use of calves for milk let-down was found to have positive impact in reducing the prevalence of SCM, probably by facilitating complete withdrawal of milk from the udder (Sedano et al. 2010). In contrast, some authors described a negative impact of use of calves for milk let-down (Shittu et al. 2012) since calves may transmit pathogens from one quarter to another. The SCM prevalence increased with the increased herd size in the farm (OR = 1.072). This situation has also been mentioned in previous study in Pemba (Suleiman et al. 2013). Prevalence of SCM was less in farms

Table 4 Risk factors associated with the prevalence of SCM based on CMT and bacteria isolation

Term	OR	95%	C.I.	Coefficient	S. E.	*Z*-statistic	*P* value
I. Risk factors based on CMT							
Water available (yes/no)	0.176	0.05	0.622	− 1.736	0.644	− 2.695	0.007
Calf milk let-down (yes/no)	0.244	0.1	0.601	− 1.407	0.459	− 3.070	0.002
Herd size (number)	1.072	1.01	1.139	0.070	0.031	2.284	0.022
Lactation stage (number)	1.239	1.084	1.417	0.214	0.069	3.134	0.001
II. Risk factors based on bacteria isolation							
Drainage (dirty/clean)	1.211	1.005	1.459	0.191	0.095	2.016	0.043
Milking day (twice/once)	0.567	0.446	0.722	− 0.566	0.123	− 4.603	0.000
Milking technique (stripped/hand fist)	0.6332	0.502	0.799	− 0.457	0.119	− 3.843	0.000
Dry towel (yes/no)	1.594	1.094	2.323	0.466	0.192	2.427	0.015
Hand wash (yes/no)	0.562	0.412	0.766	−0.576	0.158	− 3.640	0.000
Parturitions (number)	1.116	1.033	1.207	0.110	0.04	2.772	0.005
Milk production (number)	0.947	0.915	0.981	− 0.053	0.018	− 3.035	0.002

that milked their cows twice a day compared to those milked their cows once a day. Milking a cow twice per day may reduce the volume of pathogens in the udder and hence reduce prevalence. Moreover, the complete removal of the milk during milking may reduce nutrients for the pathogens to grow. The prevalence increased with increased number of parity ($P = 0.005$) and stage of lactation ($P = 0.001$). This may be caused by decreased cow immunity in older animals compared to young cows, or weakness in sphincter muscle leaving the teat orifice open after milking. Similar findings were observed by other researchers (Karimuribo et al. 2008; Moges et al. 2011). Farmers using towels to dry their cow before and after milking had more infected cows ($P = 0.015$) probably due to the use of the same towel for all cows in the shed which results in the distribution of infection from one cow to another. The prevalence as defined by bacteria isolation seem to be reduced as the milk production increase (OR = 0.94), probably due to the low concentration of pathogen in the milk.

Multi-drug resistance is threat to the control and treatment of mastitis since it reduces choice of drug to be used. Most farmers in the study area use penicillin, streptomycin and tetracycline to treat their animals and a combination of penicillin and neomycin or kanamycin for intra-mammary infusion. Major bacterial pathogens isolated were resistant to those common antibiotics used to treat bovine mastitis and other bacterial diseases in the study area. Some drugs such as penicillin are no longer appropriate for the treatment of mastitis since 87.6% of the pathogens isolated were resistant to penicillin. High prevalence of antimicrobial resistance that found in this study does not differ much with what has been previously described in other parts of Africa (Kasozi et al. 2014; Ibrahim et al. 2014). Multi-drug resistance in veterinary medicine may raise a huge public health concern since the same drugs are used to treat infections in the human population. The threat may be by introducing antibiotic residue into the human food chain that may leads to the direct or indirect toxicity or allergic reaction; or by failure therapy to the common clinical diseases (Vishnuraj et al. 2016). Most farmers use drugs to treat their animals without proper advice from qualified veterinarians and do not abide to the withdrawal period indicated as described before (Tilahun and Aylate 2015).

Conclusion

Bovine mastitis has long been considered to be a disease of economic importance in the dairy industry. A high percentage of farmers reporting cases of clinical mastitis (CM) and a high prevalence of SCM observed during this study are the evidence of the problems' extent in Zanzibar. CMT screening test provides useful information about the status of mastitis under field circumstance. Bacteria isolation could provide detailed information about the status and nature of pathogen involved. Antimicrobial testing could also be useful tool to diagnose which antibacterial should be used in each case of mastitis. Based on the finding of this study, various farm and cow level risk factors could play a role in the increased occurrence of SCM.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest and this document is the original research work conducted at Maruhubi Veterinary Investigation centre in Unguja Island of Zanzibar, Tanzania, as part of post graduate study of the corresponding author, and no part of it is submitted anywhere else for publication or conference.

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