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PREVALENCE OF SUBCLINICAL MASTITIS AND ITS EFFECTS ON REPRODUCTIVE PERFORMANCE IN DAIRY COWS DURING THE POSTPARTUM PERIOD IN GASABO **DISTRICT, RWANDA**

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ABSTRACT

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Keywords

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Mastitis is one of the most important diseases affecting production and reproductive efficiency of dairy cows. The objective of this study was to estimate the prevalence of subclinical mastitis (SCMA) and its effects on reproductive performance in postpartum dairy cows. In a cross-sectional survey, sample cows (n = 200) within their 10-40 days in milk from smallholder farms (n = 159) were selected and tested for SCMA using California Mastitis Test. They were classified into two groups: cows with SCMA (SCMA group, n = 101) and cows without SCMA (WSCMA group, n = 99). Reproductive performance including calving-to-first-oestrus interval (CFOI), calvingto-first service interval (CFSI), calving-to-conception interval (CCI), cows that remained non-pregnant, postpartum anoestrus, and total number of services per conception were recorded for 150 days in milk. The overall prevalence of SCMA recorded was 50.5%. The mean number of days for CFOI was significantly longer (p<0.05) in cows with SCMA $(87.6\pm4.8 \text{ days})$ than in cows WSCMA $(72.5\pm4.3 \text{ days})$. The mean days for CFSI was significantly longer (p<0.05) in cows with SCMA (97.2±5.6 days) than in cows WSCMA (78.9±4.9 days). The difference between the CCI of the SCMA positive cows (107.8±7.4 days) and that of the negative ones (62.5±4.5 days) was statistically significant (p<0.001). Cows with SCMA required more breeding services (p<0.05) to conceive $(1.9\pm0.1 \text{ services})$ than cows WSCMA $(1.2\pm0.1 \text{ services})$. Results indicate that SCMA had significant negative effect on fertility performance. Therefore, improving management interventions to prevent and control SCMA is essential for reducing its adverse effects in dairy cows.



Contribution/Originality: This study is one of very few studies which have investigated the occurrence of subclinical mastitis and its effects on subsequent reproductive performance of zero-grazed dairy cows under existing smallholder farming conditions. The study has also contributed to the existing literatures concerning mastitis infections in dairy cows.

1. INTRODUCTION

Mastitis in dairy cows is considered as one of the most important diseases resulting in poor production and reproductive efficiency (Ahmadzadeh et al., 2009; Halasa, Huijps, Østerås, & Hogeveen, 2007; Komba & Kashoma, 2020) and affecting herd profitability and sustainability (Dolecheck, García-Guerra, & Moraes, 2019). Mastitis is an inflammation of mammary glands and may be clinical or subclinical mastitis (SCMA). The former is often characterized by visible changes in milk composition that may be associated with inflammation signs of the udder of the cow. In contrast, the latter is not accompanied by visible signs thus, milk yield appears to be normal (Fox, 2009). SCMA is considered to be 15 to 40 times more prevalent than clinical mastitis (Henri & Christine, 2003) because of its asymptomatic nature, making it potentially and extremely costly to dairy farms and the dairy industry as a whole. This may result in absence of sustainability of cow-calf operations.

The amount of milk sold per cow per day of life is a proxy of dairy herd efficiency (Birmani, Nawab, Cun, Ye, & Mei, 2019). It was previously reported that the reproductive performance of dairy cows is the primary determinant of dairy herd efficiency (Albaaj, Foucras, & Raboisson, 2017). This is because of its associations with the number of calves born per year, milk yield, and culling rate. But, mastitis has been shown to have negative effects on reproductive performance of dairy cows (Ahmadzadeh et al., 2009; Fuenzalida, Fricke, & Ruegg, 2015). In a dairy herd, cows with SCMA lead to suboptimal reproductive performance parameters: prolonged calving-to-first-oestrus interval, longer periods to the first service, longer calving-to-conception interval, a greater proportion of cows that remain non-pregnant in a specified period, and the high number of services per conception compared to cows without SCMA (Birmani et al., 2019; Schrick et al., 2001). This is an indication that poor reproductive performance due to post-calving conditions impacts cow's productivity, nutrition and therapeutic costs (Komba & Kashoma, 2020). Also, Asaduzzaman, Bhuiyan, Rahman, and Bhattacharjee (2016) observed that when a cow repeatedly fails to conceive, it becomes costly for dairy farmers. This means that the combined productive and reproductive cycle in dairy cows must be managed in a way that the calving interval does not exceed 12 months (Nyabinwa, Kashongwe, Hirwa, & Bebe, 2020; Rukundo, Gachuiri, & Wanyoike, 2018).

The adverse effects of SCMA on reproductive performance demonstrates that the disease is an underlying cause of the prevalent suboptimal reproductive parameters of cows (Birmani et al., 2019) yet accurate estimates of the effects are rare, especially for smallholder dairy cows. In their studies, Moore, Cullor, Bondurant, and Sischo (1991) and Barker, Schrick, Lewis, Dowlen, and Oliver (1998) reported that the negative correlation between mastitis and reproduction efficiency is related to the alteration of the intervals between estrus cycle and decreased length of the luteal phase. Furthermore, Birmani et al. (2019) reported that mastitis pathogens may induce production of prostaglandins $F2\alpha$, which subsequently may cause luteolysis. So far, the prevalence of SCMA and its effects on reproductive performance of postpartum dairy cows of Gasabo district in Rwanda is not known. The hypothesis was that subclinical mastitis (SCMA) would negatively impact reproductive performance. Therefore, the current study was carried out to determine the prevalence and the effects of SCMA on reproductive performance of postpartum smallholder dairy cows under field conditions in Gasabo district.

2. MATERIALS AND METHODS

2.1. Study Area

The study was conducted in Gasabo district located within the Rwanda medium rainfall medium-altitude livestock production zone $(1^{\circ}52' \text{ S}, 30^{\circ}06' \text{ E})$ (Figure 1). The district is one of the three districts of Kigali City, with

15 administrative sectors, 73 cells, 501 villages, and covers an area of 430.30 Km2 of which 90.0% represent the rural zone (DDP, 2013). The topography is characterized by the mixture of high mountains with an average altitude of 1,800m mainly located in rural zone, sloping basins and valleys. It has a temperate tropical highland climate with two major climatic seasons in a year, the dry and rainy seasons. The average annual rainfall ranges between 900 and 1500 mm with the mean annual temperature of 22°C (Rwanda Livestock Master Pan, 2017). The district was chosen because of its highest concentration of smallholder dairy zero-grazing production in the country (MINAGRI, 2015).

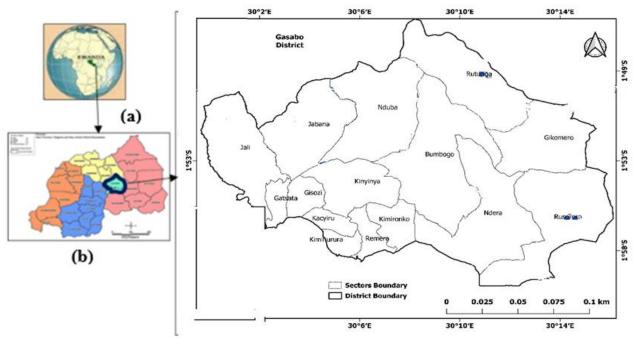


Figure-1. Map indicating the location of study areas (a) Map of Africa, (b) Map of Rwanda and Map of Gasabo district. Source: DDP (2013).

2.2. Study Design

A cross-sectional study was used to determine the prevalence of SCMA. Thereafter, sample cows were followed in a prospective study design within four months of post-SCMA diagnosis to evaluate the impact of SCMA on reproductive performance outcomes. One hundred and fifty-nine (159) smallholder farmers were selected through snowball sampling technique from November 2019 to February 2020. This was initiated with a list of some farmers provided to the researchers by sector animal resources officers. These initial participating farmers were recruited based on (i) having at least one cow within 10–40 days in milk, (ii) being identified by his/her peer, (iii) willingness to participate, (iv) keeping records of reproductive data, and (v) physical accessibility of the herd during the study period.

The sample size was determined by using the formula stated by Dohoo, Martin, and Stryhn (2009) as follows:

$$n = \frac{Z^2 P(1-P)}{d^2}$$

Where

n = sample size.

Z = 1.96 = z-score value at 95% confidence level.

P = Expected prevalence of SCMA.

d = precision level set at 0.06 for a 95% confidence interval.

Therefore, the sample size was determined at 95% confidence interval with an expected prevalence of 76.2% from a previous study in the same region (Ndahetuye et al., 2019) thus yielding a minimum sample size of 194 dairy cows.

For each recruited farmer, an explanation of the objective of the study was presented, and informed consent was given prior to starting data collection. Therefore, 200 dairy cows from 159 farms were enrolled in this study. Their breed distribution was 54.0% dairy crossbreds, 31.0% dairy pure breeds, and 15.0% indigenous cattle. The local breed was Ankole longhorn, and crossbreds were Ankole longhorn crossed with Jersey, Friesian, Sahiwal, and Brown Swiss. The sample cows were grouped into two groups: (i) group SCMA and (ii) group without SCMA (WSCMA) after the screening for SCMA. Cows were kept on predominantly fed on the cut- and- curry feeding system with natural grasses, crop residues, stems and leaves of banana and Napier grass (*Pennisetum purpureum*). On the first day of the farm visit, general information on farmer and herd management were collected through direct observation, and interviews using a pre-tested structured questionnaire. Cows were examined for SCMA and followed up for four months for reproductive data recording. Body condition score (BCS) was determined using a one- to five-point scale with an increment of one (Edmondson, Lean, Weaver, Farver, & Webster, 1989) and, therefore, was collapsed into binary variables: BCS≥3 meant cow in good condition and BCS<3 indicated cow in poor condition.

2.2.1. Screening for Subclinical Mastitis

Milk samples were collected and tested for SCMA using California Mastitis Test (CMT) on dairy cows at 25.8 ± 0.6 days in milk. The CMT followed the procedures described previously (Dingwell, Leslie, Schukken, Sargeant, & Timms, 2003; Leach et al., 2008). In brief, milk samples from each quarter were collected in a clean CMT paddle. Two millilitres of milk per quarter were collected in each shallow cup after foremilk was removed. This is the quantity of milk that was left in each cup if the CMT paddle was held nearly vertical. Equal millilitre of CMT reagent was added to each cup in the paddle. The milk - CMT reagent mixture was rotated in a circular motion for 15 seconds to thoroughly mix the contents with the presence of gel or slime being recorded for each shallow cup (McGill, 2018). Reaction results were read quickly (within 5 seconds), scored visually and given as gel formation (Dingwell et al., 2003; McGill, 2018). The more gel formation, the higher the score. The CMT was repeated twice for each sample to have the accuracy of the test results. The reaction was scored by a single trained observer on a 5-point ordered categorical scale, ranging from 1 = Mixture remains unchanged to 5 = almost-solid gel forms (using 1-point increment) (Table 1). Any reaction ≥ 2 indicated that the quarter had SCMA (Leach et al., 2008). Therefore, a cow was recorded as positive for SCMA if it had at least one quarter tested positive for SCMA.

Category	Score value	Descriptions of reaction	Interpretation of infection
Negative	1	The mixture remains	No infections
		unchanged and homogenous	
Trace	2	Slight thickening of the	Positive for SCMA
		mixture	
Weakly positive	3	Distinct thickening of the	Positive for SCMA
		mixture, but no tendency to	
		form a gel	
Distinctly positive	4	Distinct gel formation	Positive for SCMA
Strongly positive	5	Strong gel formation, which	Positive for SCMA
		tends to adhere to paddle	

|--|

Note: *SCMA = subclinical mastitis.

2.2.2. Reproductive Performance Parameters

After SCMA screening, a four-month prospective observational study was used to collect subsequent reproductive performance data under existing herd management conditions practiced by the farmers. During the

monthly visits, the reproductive performance data were recorded from cows positive for SCMA as well as negative cows. These included data on first estrus detection date, first breeding service date, date of return on heat, number of services per conception, cows pregnant at end of the data collection period (150 dpp), and cows not seen in heat during the first two months postpartum. All cows were serviced based on visual estrus detection carried out daily by the farmer in the morning (05:00 - 06:00 a.m.) and afternoon (17:00 - 18:00). These fertility measures were recorded by the farmer and subsequently confirmed by a researcher. A cow calendar was used for proper reproductive performance data recording. These reproductive parameters were calving to first oestrus interval (CFOI), calving to first service interval (CFSI), calving to conception interval (CCI), cows that remained non-pregnant (NPC), postpartum anoestrus (PPA), and total number of services per conception (NSC).

The data collected were used to calculate the indicators considered in this study (CFOI, CFSI, CCI, NPC, and PPA) to measure the fertility performance in postpartum dairy cows. Moreover, pregnancy status assessment was conducted using transrectal palpation procedures and occurred at a mean of $74.4\pm$ 0.4 (range: 60 to 90) days following service. A cow not detected in oestrus during the first two months after calving was classified as being in postpartum anoestrus. The definitions of the reproductive performance indicators were obtained from the literature (Birmani et al., 2019; Nyabinwa et al., 2020).

2.3. Statistical Analyses

The prevalence of SCMA was calculated as the number of SCMA positive cows divided by the sample size. The quarter SCMA prevalence was computed as the number of quarters with SCMA divided by the total number of quarters examined. Descriptive statistics were generated using frequency procedures and cross-tabulation.

Data on DFO, DFMA, and DNP were tested for normality using a Shapiro-Wilk normality test, and the appropriate transformation was conducted to fulfil the parametric assumptions (Manikandan, 2010). Therefore, they were log10 transformed before analysis, but for ease of interpretation, the results are reported in the original scale (Scheid, Schiavon, Gastal, Timm, & Lucia Jr, 2012).

Data on CFOI, CFSI, CCI, NPC between cows positive and negative for SCMA were analyzed using Independent-Samples T-Test. The CCI at 150 days in milk was evaluated using time-to-event analyses (Austin, 2017). Furthermore, data on PPA for cows positive and negative for SCMA were compared by Pearson's chi-Square Test. Analyses were performed using SPSS software version 22.0 for windows (SPSS, 2013). Ninety-five percent confidence interval (95% CI) for each SCMA prevalence estimate were computed by using the EpiTools software (AusVet Animal Health Services, 2015). In all analyses, the level of significance was set at p<0.05.

3. RESULTS

Variables	Levels	Number	Frequency (%)
Gender (%)	Male	90	56.6
	Female	69	43.4
Educational level (%)	Informal	63	39.6
	Primary	69	43.4
	Secondary	23	14.5
	University	4	2.5
Cattle breed	Local	30	15.0
	Crossbreds	108	54.0
	Pure	62	31.0
Herd size	1–3 cows	196	98.0
	≥4 cows	4	2.0
Parity	Primiparous	70	35.0
	Multiparous	130	65.0

Table-2. Characteristics of respondents (N = 159) and herds in Gasabo district, Rwanda.

3.1. Study Farm Characteristics and Management

Table 2 presents the characteristics of farmers and herds in the study area. The mean \pm SE number of cows sampled per farm was 1.2 ± 0.0 (median =1), with a range across all farms from 1 to 3 cows. The average days in milk at the sampling of 25.8 ± 0.6 days (median = 26.0). 35.0% of cows sampled were primiparous and 65.0% were multiparous. More than a third (39.6%) of sample farmers were without formal education, and males were dominating (56.6%) dairy farming. The sampled farms had an average herd size of 1.2 ± 0.1 (median =1.0) lactating cows with a range of 1 to 5 cows on small farm holding of 4.8 ± 0.2 (median = 4.9) acres. Daily milk yield per cow was 7.3 ± 0.2 (median = 6.5) litres and lactation length of 255 days corresponding to 1 861.5 litres of milk/cow/lactation length (Table 3).

Variables	Number	Mean±SE*	Median	Minimum	Maximum
Age of farmers (years)	159	40.6 ± 0.2	39.0	23.0	55.0
Land size (acres)	159	4.8 ± 0.2	4.9	0.6	8.6
Herd size (number)	159	1.2 ± 0.1	1.0	1.0	5.0
Dairy farming experience (years)	159	10.3 ± 0.5	10.0	1.0	25.0
Milk yield (mean litres/cow/day)	200	7.3 ± 0.2	6.5	1.5	18.0
Lactation length (months)	200	8.5 ± 0.1	5.0	4.0	10.0
Lactation milk yield*		1 861.5	1 275.0	1 020.0	2 550.0
(litres/cow/lactation length)					

Table-3. Mean estimates for respondents and herd characteristics.

Note: *SE = standard error, *predicted from milk yield/cow/day and lactation length.

3.2. Distribution and Association of Subclinical Mastitis with Different Variables

The distribution and association of SCMA with different variables are presented in Table 4. The overall cowlevel prevalence of SCMA was 50.5% (95% CI: 43.4–57.6). The prevalence of SCMA was found significantly higher (p<0.001) among pure dairy breeds (22.0%, 95% CI: 16.5–28.4) than among dairy crossbreeds (20.5%, 95% CI: 15.1– 26.8) and indigenous local breeds (8.0%, 95% CI: 4.6–12.7). The prevalence of SCMA was higher in rear quarters (81.2% and 95.0%) than in front quarters (50.5% and 53.5%).

Parameter	Levels	Cows	Affected	SCMA	95%	95% CI		
		examined (n)	cows (n)	prevalence (%)	Lower limit	Upper limit	square statistic	
Cow breed	Local	30	16	8.0	4.6	12.7	18.9***	
	Crossbreds	108	41	20.5	15.1	26.8		
	Pure	62	44	22.0	16.5	28.4		
	Total	200	101	50.5	43.4	57.6		
Parity	Primiparous	70	27	13.5	9.1	19.0	3.5*	
	Multiparous	130	74	37.0	30.3	44.1		
Cow age (years)	<5	87	36	18.0	12.9	24.0	3.1*	
	>5	113	65	32.5	26.1	39.5		
Affected quarters	1		51	50.5	40.4	60.6	25.5***	
	2		54	53.5	43.3	93.4		
	3		82	81.2	72.2	88.3		
	4		96	95.0	88.8	98.4		
Flooring type	Concrete	34	20	10.0	6.2	15.0	1.5 ^{NS}	
_	Earthen	166	91	45.5	38.5	82.7		
Body condition score	Poor (<3)	171	97	48.5	41.4	55.7	1.6 ^{NS}	
	Good (≥ 3)	29	14	7.0	3.9	11.5		

Table-4. Distribution and association of subclinical mastitis with different variables.

Note: NS Not significant (p>0.05), *p<0.05 and ***p<0.001. *CI = confidence interval.

In this study, the prevalence of SCMA increased with the advancing age. Therefore, the prevalence was significantly higher (p<0.05) in older cows (age>5 years) than younger cows (age<5 years). The findings of the study revealed a significant association (p<0.05) between the parity of cows and SCMA prevalence. The prevalence of SCMA was 13.5% (95% CI: 9.1-19.0) and 37.0% (95% CI: 30.3-44.1) in primiparous and multiparous cows, respectively. In contrast, body condition score and flooring types were not associated (p>0.05) with the occurrence of SCMA in dairy cows under study.

3.3. Effects of Subclinical Mastitis on Reproductive Performance

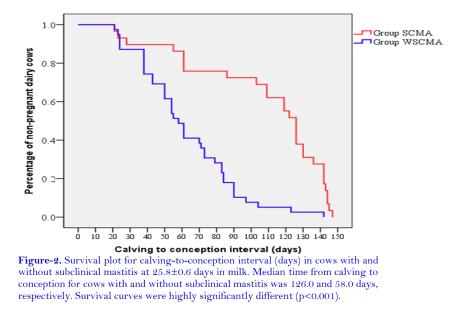
Table 5 shows the effects of SCMA on the reproductive performance in dairy cows. The mean days CFOI and CFSI were significantly longer (p<0.05) in cows with SCMA than cows without SCMA. Cows with SCMA required 108 (95% CI: 102–114) days to get pregnant and cows without SCMA needed 63 (95% CI: 58.1–65.9) days. Furthermore, in this study the overall mean value of NSC for cows became pregnant was 1.2. Cows with SCMA required (p<0.05) more breeding services to conceive (1.9 ± 0.1 services) than cows without SCMA (1.2 ± 0.1 services). On the other hand, there were no significant differences (p>0.05) in the NPC and PPA between cows with and without SCMA.

Reproductive performance parameters	Mastitis status groups						P-value
	SCMA* group		WSCMA* group				
	n	Mean±SE	Median	n	Mean±SE	Median	
Calving-to-first-oestrus interval (days)	63	87.6 ± 4.8	97.0	64	72.5 ± 4.3	65.5	0.022
Calving-to-first-service interval (days)	56	97.2 ± 5.6	102.5	63	78.9 ± 4.9	73.0	0.015
Calving-to-conception interval (days)	29	107.8 ± 7.4	126.0	39	62.5 ± 4.5	58.0	0.001
Cows that remained non-pregnant (%)	72	36.0		60	30.0		0.063
Postpartum anoestrus (%)	67	33.5		59	29.5		0.068
Total number of services per conception	29	1.9 ± 0.1	1.1	39	1.2 ± 0.1	1.0	0.025
(number)							

Table-5. The effects of subclinical mastitis on reproductive performance

Note: *SCMA = cows with subclinical mastitis, WSCMA = cows without subclinical mastitis, n = number of cows.

From time-to-event survival analysis (Figure 2), cows with SCMA had 68 days longer median time to conception than cows without SCMA.



The effects of SCMA on reproductive performance parameters in relation to cow breed are presented in Table 6. For indigenous cattle breeds, there was no difference (p>0.05) in reproductive performance parameters between

positive and negative cows. CCI of dairy crossbreds was affected with SCMA. It significantly (p<0.001) delayed conception by +54 days. Also, in dairy pure breeds, CFOI, CFSI, and CCI were increased (p<0.05) in dairy pure breeds with SCMA compared with those without SCMA. The condition significantly delayed the first estrus by +22 days, first breeding services by +27 days, and conception by +44 days. Furthermore, dairy pure breeds that exhibited SCMA had greater PPA (p<0.001) compared with cows that were uninfected with SCMA.

Reproductive performance		Indigenous c	attle breeds	Dairy cro	ssbreds	Dairy pure breeds		
		Positive Negativ		Positive	Negative	Positive	Negative	
measures			U		U		U	
CFOI	n	9	8	24	42	30	14	
	Mean	85.7±11.7	63.1±13.5	85.3±8.7	75.6 ± 5.2	90.1 ± 6.9	68.6 ± 9.7	
	P-value	0.224		0.3	10	0.04	48	
CFSI	n	9	8	21	41	26	14	
	Mean	99.4±13.7	81.5±14.3	93.8 ± 9.0	80.6 ± 6.2	99.1 ± 8.7	72.3 ± 9.9	
P-value		0.3	81	0.22	25	0.041		
CCI	n	1	5	10	23	18	11	
	Mean	61.0	60.6±10.6	117.3 ± 10.5	63.5 ± 5.9	105.1 ± 10.2	61.2 ± 9.5	
	P-value	0.988		0.001		0.001		
NPC	n	15	9	31	44	26	7	
NSC	n	1	5	10	23	18	11	
	Mean	2.0	1.0±0.0	1.3 ± 0.2	1.2 ± 0.1	1.3 ± 0.1	1.1±0.1	
	P-value	0.096		0.431		0.243		
PPA	n	7	6	17	25	14	4	
	%	23.3	20.0	15.7	23.1	22.6	6.5	
	P-value	0.2	15	0.03	31	0.001		

Table-6. Mean values for the effect of subclinical mastitis on the intervals from calving to first oestrus, first service and to conception, postpartum anoestrus, number of cows that remained non-pregnant and total number of services per conception in relation to cow breed.

Note: *n = number of cows, *CFOI = calving to first oestrus interval, *CFSI = calving to first service interval, *CCI = calving to conception interval, *NPC = number of cows that remained non-pregnant, *NSC = number of services per conception, *PPA = postpartum anoestrus.

4. DISCUSSION

This study is one of the first to estimate the effect of subclinical mastitis (SCMA) on reproductive performance of zero-grazed dairy cows on existing smallholder farm management conditions in Rwanda. Consequently, the main novelty of this study is that it was conducted in smallholder farms in Africa especially in Rwanda, thus highlighting a different scenario from that seen in modern high-yielding dairy cows. The study brings SCMA under the attention of animal health service providers and animal science researchers to understand the severity of the condition and to work for prevention and control of SCMA in dairy cows.

The findings of this study show that the prevalence of SCMA (50.5%) was high in dairy cows in the study area, indicating a need to control and prevent SCMA. The current findings corroborate with that reported in a recent studies conducted in Rwanda in the same type of farms (55.6%) (Pascal Nyabinwa, Kashongwe, Habimana, Hirwa, & Bebe, 2020) and in Tanzania (51.6%) (Mdegela et al., 2009). The observed prevalence is higher than 38.9% reported in Sudan (Hussein, 2012) 15.0% in Brazilian dairy cows (Ruegg & Pantoja, 2013) and 35.9% obtained in Tanzanian dairy cows (Komba & Kashoma, 2020). However, the present estimate of SCMA prevalence in this study was lower than the previously reported SCMA prevalence in Rwanda (70.2%) (Ndahetuye et al., 2019) Uganda (86.2%) (Abrahmsén, Persson, Kanyima, & Båge, 2014) Kenya (64.0%) (Mureithi & Njuguna, 2016) and Ethiopia (59.2%) (Abebe, Hatiya, Abera, Megersa, & Asmare, 2016). The different prevalence rates of SCMA in these studies might be explained by varied management conditions of the herds, SCMA prevalence, cow breeds, parity of cows, age of cows and milking hygiene.

This prevalence obtained in this study could be attributed to the lack of proper practice farming management and routinely testing their cows for SCMA at an earlier stage of the postpartum period. On the other hand, the predisposing factors contributing to SCMA were predominant in the study herds. These were cows kept in muddy condition, crossbreeding local with exotic breeds, low level of education, milker with dirty clothes milking with hand, absence of bedding material, earthen flooring in the cowshed, lactation period, high stocking density in a small cowshed and land size, a higher percentage of multiparous older cows, and a higher proportion of cows in poor body condition.

The current observation corroborates those of Hillerton, Bramley, Staker, and Mckinnonf (1995) in the United Kingdom and Mpatswenumugabo et al. (2017) in Rwanda. Additionally, the non-implementation of management interventions to prevent and control SCMA has been attributed to the low farmer's awareness about SCMA (FAO, 2014) and lack of quality or payment standards for milk in Rwanda (Ndahetuye et al., 2019). To address this, firstly animal health service providers should increase awareness and education of dairy farmers about mastitis prevention and control, secondly, quality or payment standards for bulk milk somatic cell count should be a priority to have cows with good udder health in smallholder dairy herds in Rwanda.

The finding of the current study was that the prevalence of SCMA increased with the advancing age which corroborates with the results of another study carried out in Bangladesh (Bari, Alam, Uddin, & Rahman, 2016). This may be due to the gradual suppression of the physiological condition of the body in older cows than in younger ones (Dego & Tareke, 2003). The higher risk of SCMA associated with increased par ity in this study may be attributed to the fact that multiparous cows in the studied dairy herds might have had cumulatively several exposures to mastitis pathogens from unhygienic hand milking and lack of mastitis management plan (Ndahetuye et al., 2019). Also, in their study, Dego and Tareke (2003) reported that multiparous cows have pendulous/large udder which increases the risk of mechanical trauma and bruises from lactating their calves and their glandular are sensitive to inflammation and the immunity of older cows is poorer than in younger ones.

The observation of higher SCMA prevalence in rear quarters is similar to earlier studies (FAO, 2014; Mpatswenumugabo et al., 2017). Although it is not easy to provide a clear explanation for this finding, this might be due to the lower position of rear quarters in relation to front ones, which make them more susceptible to contamination and/or physical trauma leading to SCMA.

Further, it is revealed in the present study that the higher prevalence of SCMA was among pure dairy breeds than among dairy crossbreeds and indigenous cattle breeds. Breed differences in susceptibility to SCMA has also been reported by other studies (Abrahmsén et al., 2014; Dego & Tareke, 2003). The findings of the current study could be associated with the lower degree of adaptation of improved breeds to tropical conditions of high temperature and humidity, inadequate feeding practices, and poor hygienic conditions (Iraguha, Hamudikuwanda, & Mushonga, 2015; Pal & Chakravarty, 2020) making them more susceptible to disease than indigenous cattle breeds. Another reason may also be since improved breeds require elaborated management practices, adequate feeding, and better health care to get better health status than indigenous cattle breeds (Pal & Chakravarty, 2020). These conditions are sometimes absent under smallholder zero-grazing dairy farming (Hernández-Castellano et al., 2019; Ndahetuye et al., 2019). To manage this, dairy farmers could help prevent SCMA by maintaining hygiene in the cows' environment and supplying adequate feeding in the peri- and post-parturient periods. This is because mastitis is one of the challenges faced by the dairy sector across the globe and leads to the subfertility of dairy cows (Birmani et al., 2019). This may increase the culling rate and decrease the profitability of the dairy herd (Geary et al., 2012; Ndahetuye, Olivier Basole, Claire d'Andre, & Bockline Omedo, 2021). Further study is required to assess the breed difference. Though the current study was based only on the qualitative test of CMT, there is a need to perform laboratory evaluations and establish the pathogens that may be involved in the present observation.

The high prevalence of SCMA in dairy cows at 25.8 ± 0.6 days in milk resulted in detrimental effects on subsequent reproductive performance. The effect on all the reproductive performance measures examined was consistent: SCMA positive cows performed poorer than healthy cows.

CFOI and CFSI in SCMA group were significantly increased (+15 and +18 days, respectively) compared to WSCMA group. Also, CCI and NSC were significantly increased in SCMA group (+45 days and +0.7 services) compared to WSCMA group. This demonstrates that for maintenance and sustainability of a dairy herd to be

economically viable, optimizing the reproduction efficiency is crucial (Rodgers et al., 2012). These findings give the animal health service providers a further opportunity to improve the udder health, welfare and performance of Rwandan smallholder dairy herds. The observed findings are in accordance with that previously reported in Turkey (Gunay & Gunay, 2008). In this study, the NSC and CCI were found to be 1.9 ± 0.1 services and 107.8 ± 7.4 days, respectively, lower than 3.1 ± 0.3 services and 143.5 ± 11.4 days for cows with SCMA reported by Schrick et al. (2001) in Knoxville. This shows that increased NSC was reflected in the prolonged CCI indicating a shorter inter calving interval in cows without SCMA leading to a decrease in herd profitability and sustainability. This is because smallholder farmers being unable to ensure proper feeding practices, they have to wait for long periods for dairy cows as a proxy of inadequate feeding practices may result in more cows having a negative energy and protein balance which increases the exposure of cows to udder infections and this results in poor reproductive performance of dairy cows. Therefore, it is desirable to farmers to ensure good hygiene and proper feeding practices in the transition period to prevent SCMA and consequently improve the reproductive performance of their dairy cows.

A plausible mechanism for an increase in the NSC, NPC and PPA in cows with SCMA is a disturbance in hypothalamic-pituitary-ovarian-uterian axis (Sharma et al., 2017) through a depression of gonadotropin-releasing hormone which delays follicle growth, ovulation, and functioning of corpus luteum (Heravi, Danesh, & Gilbert, 2012). In their study, Sharma et al. (2017) concluded that poor reproductive performance in SCMA positive cows could be linked to the fertilization failure, impaired oocyte development, altered hormonal profile, and unfavorable uterine milieu for embryonic development. Furthermore, Ribeiro et al. (2016) reported that SCMA interferes with feed intake, an expenditure of energy and nutrients which results in lowering body condition and delaying the resumption of the postpartum ovarian cycle. This delays ovulation which results in increased CFSI and CCI (Birmani et al., 2019). In Rwanda, the per capita milk consumption is 63.0 litres per person per year (Rwanda Livestock Master Pan, 2017). This indicates that an increase of 3.5 fold would be necessary to achieve per capita milk consumption threshold of 220 litres recommended by the Food and Agriculture Organization of the United Nations (FAO, 2013). In this context, it is the utmost need of the time to set proper management interventions for prevention and control of SCMA to improve the reproductive efficiency and the production of high-quality milk of smallholder dairy cows so that milk consumption per capita requirements can be met.

5. CONCLUSION

The prevalence of SCMA was high and SCMA positive cows had lesser reproductive performance than SCMA negative cows. The dairy industry, in turn, incurs huge economic losses through reduced the profitability and sustainability of dairy herds. Therefore, to prevent these losses, prevention and control program, and early diagnosis of SCMA in dairy cows should be integrated into any reproductive performance-enhancing approach particularly during the early postpartum period.

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