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Impact of Lameness on Uterine Health, Reproductive Performances and Hormonal Profile of Zebu X Friesian Crossbred Dairy Cows In and Around Jimma Town Dairy Farms, Ethiopia

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Abstract

Longitudinal observation study was conducted from January 2020 to July 2021 in and around Jimma town dairy farms, Ethiopia. The goal of this study was to examine the first-service conception and pregnancy rates of cows classified as non-lame, mildly lame, moderately lame, lame, and severely lame. In addition, the link between lameness and subclinical endometritis was investigated. Finally, we wanted to look at progesterone and cortisol hormones to see if they had anything to do with lameness. Every month, cows were evaluated for lameness using a 5 points locomotor scoring system. The cytobrush approach was used to detect subclinical endometritis. The electrochemilu minescence immunoassay "ECLIA" was used to measure prog esterone and cortisol levels in cow blood serum. Cows that were clinically lame had a lower Conception Rate at First Services (CRFS) than cows that were never lame or mildly lame, although the difference was not statistically significant (P>0.05). When compared to cows that had never been lame and slightly lame cows, clinically lame cows had a lower Pregnancy Rate at First Services (PRFS). This difference was significant (P<0.05). Lameness can be influenced by a variety of circumstances, including parity and physical condition. Lameness was found to be strongly linked to subclinical endometritis (P=0.035). Lameness caused a decrease in progesterone levels and an increase in cortisol concentration. In conclusion significant associations were found in this study between different degrees of lameness and fertility. Reducing lameness should be part of the management plan used to improve fertility of dairy cows.

Keywords: Cortisol; Cytobrush; Dairy cows; Locomotion score; Progesterone; Reproductive performance

Introduction

The profitability of dairy farms is largely determined by reproductive performance [1]. Many factors influence reproductive losses, with lameness being one of the most important [2].

Any anomaly that causes the animal's stride or posture to shift is referred to as lameness [3].

Lameness after calving has been linked to delayed cyclicity [4], increased ovarian cyst incidence, and lower pregnancy to first AI [5] as well as longer intervals between calving and first AI and conception (Calving to Conception Interval, CCI). Huxley [6] found that caving to the first service interval was 7 days longer, calving to conception interval was 30 days longer, conception rate was 20% lower, and there were 1.2 more services per conception.

When compared to non-lame cows, lameness delays the onset of ovarian cyclicity by up to 18 days and the onset of oestrus by up to 24 days. Furthermore, higher blood cortisol levels linked to lameness interfere with the pituitary gland's secretion of Luteinizing Hormone (LH) [7]. Walker et al. discovered that a reduction in progesterone caused a decrease in sexual activity during oestrus in lame animals [8]. Due to a delay or blockage of the LH surge, lameness in dairy cows has also been linked to an increased incidence of ovarian cysts.

Ethiopia has a tremendous potential for dairy development because it has the highest livestock population in Africa, with cattle accounting for around 58 million head [9]. Despite these large numbers, milk production frequently falls short of the country's needs due to a variety of factors, the most prominent of which is the occurrence of lameness [10-12]. However, there is no literature available in the country that describes the effects of lameness on reproduction and its link to subclinical endometritis. As a result, the goals of this study were to investigate and quantify the influence of lameness on reproductive performance, as well as to assess the link between lameness and subclinical endometritis in cross breed cows and their hormonal profiles.

Materials and Methods

Description of study area

The study was conducted from November 2016 to April 2017 in and around Jimma town which is the town is located in the south western part of the Ethiopia in Oromia regional state. Jimma town is found at distance of about 352 km from Addis Ababa, the capital city of Ethiopia. Geographically, it is located at 7°13′ and 8°56′N latitude and 35°52′ and 37°E longitudes. The area has an altitude ranging between 880 and 3358 m above sea level. The annual rainfall is ranging between 1200 mm–2000 mm; and the annual temperature of the area ranges 7°C–30°C. Farmers in the area practices mixed crop-livestock agriculture. The zone is one of the major coffee growing areas in southwest part of Ethiopia. Furthermore, the zone is well known by livestock production which can be estimated at about

2,212,962 cattle, 866,561 sheep, 457,311 goats, 96,782 horses, 17,644 mules, 77,767 donkeys, 1,951,129 poultry and 546,722 beehives.

Study animals and reproductive management

The study was conducted on a total of 91 normally calved crossbred dairy cows (Holstein Friesian x Zebu crossbred). The data were collected between 2020 and 2021 which calved from January 2020 to July 2021. Based on a locomotion scoring system devised by Sprecher, et al. [13] study cows were classified into five categories of lameness: 1–non-lame (41), 2–mildly lame (8), 3-moderately lame (27), 4-lame (13) and 5-severely lame (2). Cows which scored ≥ 3 points were classified as Clinically Lame (CL).

The study animals were Holstein x Zebu crossbred cows selected from 11 intensively managed farms in the study area. The intensively managed cattle were kept in doors and received concentrate feeds in addition to hay and crop residues (such as corn stalks, wheat/barley straw and other leftovers from grain threshing). Animals on the farm were regularly vaccinated against common infectious diseases like Lump skin disease, Anthrax, Blackleg, and Foot and mouth disease. Regular preventive treatments were administered against prevalent endo-parasites and ecto-parasites.

All cows for observational study were kept under the same environment, feeding regime and husbandry system for the whole study period. In order to avoid confounding factors, the presence of reproductive health problems, some metabolic diseases and mastitis were recorded and excluded from the study, since these diseases can influence reproductive performance.

The first service in each herd did occur after the voluntary waiting period, which was 50 days after calving in all farms. Oestrus signs were monitored by visual observation. Pregnancy was confirmed by rectal palpation 60-65 days after Artificial Insemination (AI).

Longitudinal study

This is longitudinal observation study where lame cows and non-lame cows compared for different outcome variables. The study animals were postpartum crossbred dairy cows (Zebu x Holstein Friesian) were followed up for 330 days after calving to record their reproductive indices.

Screening of lameness

Lameness was assessed based on a scale of 1 to 5 locomotion score, devised by Sprecher, et al. considering as lame dairy cattle those ones obtaining scores ≥ 2 [13]. For the locomotion assessment of the dairy cattle kept in tie stalls, these were loosened and were taken out of the barns. Similarly in some of the farms where the dairy cattle were being let to exercise and/cleaning purpose were assessed after the morning milking.

Cytobrush technique to determine subclinical endometritis

Subclinical endometritis was determined using cytobrush technique connecting a modified human smear cytobrush, to the plunger of an insemination gun and protected by the covered with plastic AI sheath, introducing it into the uterus as for doing artificial insemination. The cytobrush was introduced into the vagina and by per rectal manoeuvring it traversed the cervix and reached the uterus. Inside the uterus, the cytobrush is pushed out of the catheter, gently rotated on to the uterine wall a full 360 degrees clock wise to obtain cellular material from the endometrium. Immediately after removal from reproductive tract, the cytobrush was smeared on clean glass slide and stained with Geimsa stain. The slide was observed under microscope on 400X and oil immersion (for endometrial cells+PMN cells). The subclinical endometritis was declared on the basis of ≥ 4% Polymorphnuclear cells (PMN) [14].

Progesterone and cortisol analysis

Approximately 10 ml of blood sample was collected from each selected cattle and the serum was separated. The sera were stored at -20°C until analyses. Analysis of progesterone and cortisol concentrations in blood serum of cows were performed using electrochemiluminescence immunoassay "ECLIA" at Jimma specialized hospital, clinical chemistry laboratory.

Data collection and statistical analysis

For the purpose of this study body condition score (5 scores: 1-5 scores) of all sampled cows was measured as described by Edmonson et al. and categorized as poor (≤ 3) or good BCS (>3)[15]. Parity classified as primiparous and multiparous. All the dairy cattle in selected farms were repeatedly observed for the presence of abnormal gait. All data were collected and recorded in a Microsoft Excel 2010 spreadsheet and statistical analysis was conducted using SPSS statistical software version 20.0. Descriptive statistics were used to compute percentages, proportions and frequency distributions of the data. In order to determine the degree of association between lameness and subclinical endometritis logistic regression was used. Conception Rate at First Service (CRFS) and pregnancy Rate at First Service (PRFS), progesterone and cortisol concentrations of the lame cows and non-lame cows were compared by ANOVA. Tukey's test was used to express the difference among the groups. Statistical analysis was conducted using SPSS version 20.0. Statistical significance was set at $P < 0.05$ to determine whether there are significant differences between the parameters measured among the groups.

Results

Impact of lameness on reproductive performance

The conception rate at first service (CRFS) in non-lame cows (n=41) was 23 (56.1%) while in mildly lame cows (n=8) was 4 (50%). The conception rate at first service (CRFS) in moderately lame cows (n=27) was 9 (33.3%), in lame cows (n=13) was 3 (23.1%) and in severely lame cows (n=2) was 1 (50%) but this difference was not significant (P=0.189) (Table 1).

Locomotion score	No examined	CRFS n (%)	x^2	P-value
1 (Nonlame)	41	23 (56.1)	5.08	0.189

2 (Mildly lame)	8	4 (50)		
3 (Moderately lame)	27	9 (33.3)		
4 (Lame)	13	3 (23.1)		
5(Severely lame)	2	1(50)		
X^2=*Chi square*, P=Probability				

Table 1: The relationship between Conception Rate at First Service (CRFS) and different degrees of lameness in crossbred cows.

The Pregnancy Rate at First Service (PRFS) in non-lame cows (n=41) was 19 (46.3%) while in mildly lame cows (n=8) was 2 (25%). The severely lame cows (n=2) was 0(0%), this difference was significant (P=0.026) (Table 2).

Locomotion score	No examined	PRFS n (%)	X^2	P-value
1 (Nonlame)	41	19 (46.3)	11	0.026
2 (Mildly lame)	8	2 (25)		
3 (Moderately lame)	27	5 (18.5)		
4 (Lame)	13	1 (7.7)		
5(Severely lame)	2	0 (0)		
X^2=*Chi square*, P=Probability				

Table 2: The relationship between Pregnancy Rate at First Service (PRFS) and different degrees of lameness in crossbred cows.

Risk factors

Of the good body conditioned cows, 13 of 37 (35.1%) were positive and of the poor body condition cows, 37 of 54 (68.5%) were positive for lameness (P=0.002). From 37 examined primiparous cows, 15 (40.5%) were positive, and among 54 examined multiparous cows, 35 (64.8) of them were positive (P=0.022) for lameness (Table 3).

Factor	Group	No examined	Prevalence (%)	OR	95% CI	P-value
Body condition score	Good (>3)	37	13 (35.1)			0.002
	Poor (≤ 3)	54	37 (68.5)	0.25	0.103-0.6	0.022
Parity	Primiparous	37	15 (40.5)			
	Multiparous	54	35 (64.8)	2.7	1.14-6.4	
CI=confidence interval; OR=Odds ratio; P=Probability						

Table 3: Factors associated with the occurrence of lameness.

The lesions that were found causing lameness were excessive hoof growth with vertical and horizontal fissure (76.32%), sole ulceration(13.16%) and foot rot (10.52%) (Table 4).

S. No	Type	Frequencies	Percentage	Quarters
1	Excessive hoof growth with vertical and horizontal fissure	29	76.32	Both feet
2	Foot rot	4	10.52	Rear feet
3	Sole ulceration	5	13.16	Rear feet
4	Total	38	100	

Table 4: Proportions of foot and leg problems in animals with locomotion score ≥ 3.

Effect of lameness on hormone profiles

The Serum progesterone (P4) concentration at 35 days post first Artificial Insemination (AI) in nonlame cows (n=41) was 8.74 ± 0.95 ng/ml while in mildly lame cows (n=8) was 6.2 ± 1.35 ng/ml. The Serum progesterone (P4) concentration in moderately lame cows (n=27) was 5.7 ± 2.1 ng/ml, in lame cows (n=13) was 4.7 ± 1.7 ng/ml and in severely lame cows (n=2) was 3.75 ± 4.1 ng/ml (Table 5).

Locomotion score	Description	No examined	Plasma progesterone (ng/ml)
1	Normal (Not lame)	41	8.74 ± 0.95
2	Mildly lame	8	6.2 ± 1.35
3	Moderately lame	27	5.7 ± 2.1
4	Lame	13	4.7 ± 1.7
5	Severely lame	2	3.75 ± 4.1
SD=Standard Deviation			

Table 5: Plasma progesterone concentration (Mean ± SD) indifferent degrees of lameness in crossbred cows.

The Serum cortisol concentration in nonlame cows (n=41) was 53.52 ± 14.98 ng/ml while in mildly lame cows (n=8) was 48.44 ± 5.22 ng/ml. The Serum cortisol concentration in moderately lame cows (n=27) was 63.2 ± 17.2 ng/ml, in lame cows (n=13) was 79.6 ± 20.4 ng/ml and in severely lame cows (n=2) was 105.9 ± 3.1 ng/ml (Table 6).

Locomotion score	Description	No examined	Plasma cortisol (ng/ml)
1	Normal (Not lame)	41	53.52 ± 14.98
2	Mildly lame	8	48.44 ± 5.22
3	Moderately lame	27	63.2 ± 17.2
4	Lame	13	79.6 ± 20.4
5	Severely lame	2	105.9 ± 3.1
SD=Standard Deviation			

Table 6: Plasma cortisol concentration (Mean ± SD) indifferent degrees of lameness in crossbred cows.

The association between lameness and subclinical endometritis Uterine samples revealed an overall prevalence of subclinical endometritis infections of 30.8% (28/91). Of 91 cross breed dairy cows the prevalence of subclinical endometritis was 40% (20/50) and 19.5% (8/41) in lame cows and nonlame cows, respectively (Table 7).

Lameness	Subclinical endometritis		OR	95% CI	P-value
	Negative (%)	Positive (%)			
Negative (%)	33(80.5)	8(19.5)	2.75	1.1-7.2	0.035
Positive (%)	30(60)	20(40)			
Total	63(69.23)	28(30.8)			
CI=confidence Interval, OR=Odds Ratio, P=Probability					

Table 7: The association between lameness and subclinical endometritis.

Discussion

The results obtained in the present study showed that clinically lame cows had lower Conception Rates at First Service (CRFS) (31%) than the non-lame cows (56.1%) (P=0.035). Our finding was similar to Chapinal, et al. [16], who reported 36% for lame cows in the north eastern United States. Ferguson and Skidmore [17] recorded a higher CRFS of 44% for lame cows in the United States, whereas Morton [18] reported a CRFS of 47% in Australia. Serhat, et al. also recorded conception rate was lower in lame cows compared with non-lame cows [19]. Huxley reported 20% lesser conception rate due to lameness. Melendez, et al. also found that lameness was associated with poorer conception rates at first service. These all variability might be due to pain, hormonal insufficiency and related to negative energy balance.

The current study revealed lame cows had a lower pregnancy rate at first service (14.3%) comparing with non-lame cows (46.3%) (P=0.021). In accordance to our finding Jan and Jędrzej [20] reported the pregnancy rate to first service was significantly lower in lame cows as compared with non-lame cows. Similarly, Chapinal, et al. Also reported 20% in the north eastern United States. Our finding concurs with the report of de Vries and Risco (12%), however lower than the 32% reported by Ferguson and Skidmore. This difference could be due to the management factors affect reproductive performance [21].

This study revealed that lameness prevalence was highly associated with prevalence of subclinical endometritis and lame cows were found to have significantly higher subclinical endometritis than control cows. Results reported by Garbarino, et al. declared that lameness has a detrimental effect on ovarian activity in Holstein cows in the postpartum period. Lameness in cows is associated with a higher incidence of ovarian cysts and the occurrence of ovarian cysts was associated with uterine infection [22]. Cows with endometritis also had ovarian cysts significantly more often.

The serum progesterone (P4) profile after first insemination of cows that exhibited lameness was 4.7 ± 1.7 ng/ml whereas in lameness free cows were 8.47 ± 0.95 ng/ml. Our finding agrees with previous reports of Walker, et al. that lameness reduces progesterone concentrations [23].

In the current study cows stressed by lameness had significantly higher levels of cortisol (79.6 ± 20.4 ng/ml) than control (non-lame) cows (53.52 ± 14.98 ng/ml). In agreement to our study Almeida et al. reported cortisol is used as an indicator of stress and pain and elevated serum cortisol has been shown in cows suffering from inflammatory foot lesions [24]. Endo, et al. also recorded cows with lameness were to show significantly elevated plasma cortisol compared to a control group [25]. Plasma cortisol increase is the result of pain and stress of lameness that disrupted normal reproductive hormone releases and affected the intensity of oestrus behaviour and oocyte production [26].

Lameness prevalence in this study was significantly associated with BCS and parity which agrees with findings from other authors [27,28], however contradicts with Sadiq, et al. and Mulatu. Differences might be due to system of sample size, breeds, and corresponding milk yielding capacity.

In 76.32% of the dairy cattle with LS ≥ 3 foot and leg problems were due to excessive hoof growths associated with vertical and horizontal fissures that were in agreement with the reports of Sulayeman and Fromsa, Abunna, et al. [11] and Mulatu [12]. Similarly, Sadiq, et al. reported that for cows with claw lesions, prevalence of lameness was associated with overgrown claw in dairy farms in Selangor, Malaysia [29]. According to the guideline made by Shearer [30], increased hoof growth (particularly of the outside claw of rear feet) leads to overgrowth and eventually overloading of the affected claws. The end result is a greater likelihood of developing claw disease.

Conclusion

This study confirmed consequence of lameness on reproductive performance by reducing Conception Rate at First Service (CRFS) and Pregnancy Rate at First Service (PRFS) in dairy cows. The outcome of this study revealed that lameness was highly associated with subclinical endometritis. The result of this research demonstrates lameness causes change of the concentration of progesterone and cortisol. Significant associations were found in this study between different degrees of lameness and fertility. The results from this study indicate that reducing lameness should be part of the overall management plan used to improve fertility.

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Conflict of interest

The authors declare that they have no competing interests.

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