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Evaluation of Sex Fixer (Aulprofem®) in Indigenous Arsi Cattle Reared at Adami Tulu Agricultural Research Center, Oromia, Ethiopia

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Abstract: Evaluation of Aulprofem® (sex fixer) in Arsi cattle to produce female progenies was done on some selected cattle at Adami Tulu Agricultural Research Center. The results of the Aulprofem® indicated that there was indeed a change in the sex ratio of calves 39.5:60.5 (male/female) whereas among the non-administered group the corresponding ratio was 65.1:34.9. The result also indicated that the effect of sire and breeding season influenced the sex ratio of the calves born whereas parity had no effect on the same. Therefore, it can be concluded from the results that the usage of Aulprofem® can be used as one option in altering the sex ratio of dairy cattle whereby increasing number of female animals for dairy sector and increase more milk production initiated but under strict control of the livestock experts.

Keywords: Aulprofem®, Calf Sex Ratio, Sire Effect, Breeding Season, Oromia, Ethiopia

1. Introduction

Traditionally it has been reported that the secondary sex ratio among the mammals are 50:50 towards the birth of male and female offsprings. However, depending on the requirements of the livestock rearers researchers have over the years tried to alter this ratio favouring the birth of a particular sex. Several such studies have taken into account different hypothesis viz. time of insemination, provision of different types of feeds and minerals, alteration of vaginal and uterine pH, use of sexed semen [12] usages of certain chemicals. Observations of study by Khan et al. [23] indicated that alteration of secondary sex ratio in cattle is financially attractive. In beef cattle production it can result in the birth of the bull calves which have enhanced growth rate vis-a-vis those of the cow calves of the same age [21]. Contrary the dairy farmers generally seek the birth of cow calves to perpetuate their herd. Furthermore, the requirement for a fewer male to female sex ratio (i.e., lower sex ratio) in dairy cattle is augmented when the herd replacement rates are high which can also help in improving the selection intensity [5].

Except for the usages of sexed semen, most of the studies have provided conflicting results. Sex ratio of the calves favouring the birth of cow calves is important especially among the dairy farmers or those who are associated with the dairy industry [40]. Artificial insemination (AI) has been one of the widely accepted bio techniques what have revolutionised the optimum usages of good quality sires. However small and medium scale cattle owners in different areas of Ethiopia have been complaining about more numbers of bull calves being born following artificial insemination (AI) [7, 16] similar observations have also been reported from other parts of the globe [36, 40].

There has been several contradictions too which indicate that there is no significant deviation in the sex ratio.

The sex fixer/s have advantages over sexed semen as they are relatively cheap and the manufacturers claim that they

help enhance the rate of conception [3]. Reports of a study by Aulakh [4] have indicated that Aulprofem® administered orally to the heifers/cows in estrus at the time of mating and 30 minutes after mating during the natural service or 30 minutes before AI resulted in the birth of 100% cow calves.

Aulprofem® is a patented sex fixer composed of ingredients viz. ethanoic acid, sodium salt, edible grade pharmaceutical aides and accepted additives and has been patented [2]. Usages of this patented product minimizes the chances of Y-Sperms to bind with the corresponding receptors of the ovum and as a result the non H-Y antigens present on the bodies of X-Sperms get a chance to conjugate with the receptor antibody sites on ovum thereby resulting in a female zygote [2].

The ingredients used for the development of Aulprofem® assists in favouring the combinations of chromatin containing the X chromosome bearing spermatozoa to fertilize the ovum. This is achieved by reversible blockages of the receptor sites for the "Y" spermatozoa on the surface of the ovum [41]. The word "reversible" is used here because if there was a complete blockage no bull calves would have been born.

Therefore, the aim of this study was to check the efficacy of Aulprofem® vis-à-vis those of a control population of Arsi cows reared under on station management towards the sex of the calves born through artificial insemination. The study also took into account the effect of sires, season of birth and parity on the cows and the resultant sex of the calves born.

2. Materials and Methods

2.1. Studied Location

The study was conducted at Adami Tulu Agricultural Research Center situated at latitude of 7°9'N and 38°7'E longitude in the semi-arid middle rift valley region of Ethiopia. The Center is situated at 1500 meters above sea level and the soil type of the area is fine, sandy loam with sand, clay in the proportion of 34:48:18 respectively. The average annual rain fall is 760mm. The minimum and maximum temperature are 12.6 and 27°C, respectively [1].

2.2. Selection Criteria of Experimental Animals and Procedures Followed

The Arsi breed of cows reared in the center which was used as dam line were selected based on their age, parity and body condition. Progeny tested semen of different sires of Holstein Frisian breed was used as a sire line. The parity of the selected cattle ranged between 2 to 3 and the cattle were at different stages of lactation with active and functional corpus luteum (CL).

The cows were selected based on the following criteria:

1. Good body condition (Body condition score > 3 [11].
2. Free from any genital diseases.
3. Free from any history of abortion.
4. Animals which was cyclic with active CL.

An approved commercial sex fixer preparation Aulprofem® was used to skew the secondary sex ratio of these selected cattle with an aim to favour the birth of cow calves.

Oestrus synchronization: All cows had been palpated for the presence of active CL, and PGF2α analogue (inj Lutylase®) hormone was injected intra muscularly. Lutylase® was administered to each cow at 2ml/cow after confirmation of receptive corpus luteum through rectal palpation and animal's shows heat signs naturally (cows administered with Lutylase® was fitted with Kamar® heat mount detector and also monitored by herdsmen and night guards for signs of heat in order increase the efficiency of heat detection), were artificially inseminated.

Breeding and sex-fixer administration: Cows showing signs of estrus were administered with Aulprofem® @ 0.5ml/kg 30 minutes prior to AI, the dosage was calculated based on their body weight (as indicated by the manufacturers). A standard semen handling and insemination procedure as recommended by IAEA [18]. was used to inseminate the animals. Cows noted in heat in the morning were inseminated that afternoon and those identified in the afternoon were inseminated the next morning based on the "AM- PM guideline" [28].

Pregnancy diagnosis: The pregnancy diagnosis of the inseminated cattle was carried out using Twil Canada Bovipreg® kit at 18-22 days post the service and rectal palpated at 90 days after insemination by a skilled AI technicians to ascertain the results.

2.3. Design of Experiment

The study design was Randomized complete block design (RCBD). The selected cows were assigned to each treatment based on their parity, age and body weight. Progeny tested frozen semen of four Holstein Frisian sires of World Wide Sires® (WWS) and frozen semen one Holstein Friesian sire reared at National Artificial Insemination Center (NAIC), Kaliti, Addis Ababa was used as as a control. Ten cows were assigned to each treatment.

Treatment 1- Administered with Sex fixer + AI

Treatment 2- Without Sex fixer + AI

2.4. Statistical Analysis

Collected data were coded and analyzed using Statistical Package for Social Science Version 20.0 (SPSS). The Chi-square ($\chi 2$) test procedure of SPSS Version 24.0 was used to analyze the qualitative data. The variation between groups was considered significant when the $P < 0.05$. Experimental data were analyzed by using factorial analysis statistical methods and GLM procedure of SPSS v24.

3. Results and Discussion

The findings in Table 1 indicate that the effect of Aulprofem® influenced the sex ratio of the calves ($P<0.05$) favouring the birth of more number of cow calves the observation are in close accordance with those of Sundaramurthy et al. [37]. It has been reported that usage of

Aulprofem® selectively influences the binding moieties favouring those of the X chromosomes. It has been reported that enhancement of dietary and in vitro supplementation of omega-6 PUFAs can skew the sex ratio favouring the birth of bull calves [39]. Studies by Aulakh et al. [3, 4]. indicate that Aulprofem® mimics the Y sperm bearing lignads which provides selective advantages to the X sperm bearing chromosomes to fertilize the ovum. Conversely this study also indicates that the sex ratio favoured ($P<0.05$) the birth of the bull calves among the cows which were not drenched with Aulprofem® and were inseminated artificially, the findings are in close accordance with the observations of Berry et al. [8]. who reported that cows covered through artificial insemination give birth to more numbers of bull calves.

Table 1. *Overall sex ratio of calves born.*

Parameters	N	Cows provided with Aulprofem®		N	Cows without Aulprofem®		X^2
		Bull calves	Cow calves		Bull calves	Cow calves	
Overall	38	39.5	60.5	40	70	30	0.007
Effect of sire							
Garret	7	28.6	71.4	8	75	25	
Zantiwan	8	50	50	8	62.5	37.5	
Laramee	6	16.7	83.3	7	71.4	28.6	0.007
Kodak	9	55.6	44.4	8	62.5	37.5	
NAIC/10-230	8	37.5	62.5	9	77.8	22.2	
Overall	38	39.5	60.5	40	70	30	
Effects of season of calving							
Rainy	14	57.1	42.9	24	75	25	
Dry	24	29.2	70.8	16	62.5	37.5	0.007
Overall	38	39.5	60.5	40	70	30	
Effect of parity							
2nd	21	42.9	57.1	30	70	30	0.007
3rd	17	35.3	64.7	10	70	30	

Values across the row for a trait are significantly different.

Drenching the cows with Aulprofem® is expected to change the uterine environment like the energy substrates in the uterus and also the pH, which may favour the binding of the Y spermatozoa to the lignards. Findings of a study on Hamsters indicates that the numbers of male offspring's were fewer when the uterine pH was less acidic while in the later stages of fertile cycle the vaginal and uterine pH was highly acidic and that hence more numbers of male offspring's were born [30].

The variation in uterine pH can be ascribed to the diets of the cattle while the vaginal pH can also be ascribed to the hormonal status of the cows [30]. It has been reported that alteration in the hormonal status can also lead to changes in secondary sex ratio at conception [19, 20].

The findings from this study are also in close accordance with the findings of Berry et al. [8] who reported that more numbers of bull calves are born through AI, however studies by Ballinger et al. [6], Rorie [33] and Effa Kefena Delesa et al. [14] indicated that there were no credible evidence to the fact that AI favours the birth of more numbers of bull calves.

The results further indicate that there were variations across the sires in the birth of cow and bull calves [29]. However, in this case the sex ratio favoured the birth of the cow calves when the dams were drenched with Aulprofem®. Contrary to the same there were higher numbers of bull calves born from undrenched cows. There were differences across the sex ratio of the calves born from different bulls, which may be ascribed to the differences in X and Y bearing spermatozoa, as suggested by Chandler et al. [10] and Rorie et al. [34]. Therefore, it can also be concluded that the contribution of the bulls towards the secondary sex ratio too cannot be over ruled, the observations are also in close accordance with the findings of Pauciullo et al. [27].

Differences in sex ratio was also observed across the season of calving, in both the case cases (cows drenched with Aulprofem® and those which were not drenched) gave birth to more numbers of bull calves in the rainy season. During the rainy season the cows have access to high quality forages which are high in protein content [13].

Metabolism of higher amount of protein consumed (than what is metabolised for maintenance requirement, milk yield and other body functions) may lead to high levels of plasma urea nitrogen can lead to alteration in the uterine pH favouring the development of acidic pH and hence more numbers of bull calves [9, 15].

Under nutrition and over nutrition during gestation can lead to low body weight of the offspring and thereby leading to embryonic/foetal losses [26]. Under nutrition is quite common during the dry season when there are lack of green fodder and the fodder available is deficient even in the basic nutrients [13]. Therefore inadequate quantity and poor quality of feed alongside environmental stress leads to release in stress hormones and consecutively embryonic mortality [25, 35] especially the male embryos [36, 22, 31]. Therefore, the numbers of female calves born in the dry season are higher irrespective of whether the cow was drenched with Aulprofem®.

It has also been reported in a study by Trivers et al. [38] that cows with good body condition and expected to conceive bull calves. Similar observations have also been reported by Roche et al. [32] and Berry and Cromie [8]. However, cows which calved during the wet season and lactating, have a negative energy

balance (due to poor quality of feed in the dry season) are expected to conceive with more numbers of cow calves [24].

The differences in secondary sex ratio as observed in the study also follow the results of Roche et al. [32] who reported that high temperature and humidity at the time of conception favours the conception of bull calves. Thus the cows which conceived during the dry season (October to February) gave birth to bull calves during the subsequent rainy season.

The differences due to parity in secondary sex ratio was recorded only among the cows drenched with Aulprofem®. The proportion of cow calves increased with parity which his in close accordance with those of Goshu et al. [17], however, contrary to the findings of Berry et al. [8] who recorded that the proportion of the bull calves increased with the parity of the dam. The differences as observed may be due to the effect of Aulprofem® as no variation in sex ratio was observed across parity among the undrenched cows where the proportions of the bull calves were higher across both the parities studied.

4. Conclusion

The findings from the study concluded that the secondary sex ratio favouring the birth of the cow calves were observed among the cows drenched with Aulprofem®, vis a vis control population. Therefore, it can be concluded that usage of Aulprofem® can benefit dairy cattle rearing farmers in the developing countries

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