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Research Article

Effects of Semen Sexing Agent (Heiferplus^{tm}) on Sperm Characteristics and Fertility in Cows

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Abstract

The present study was designed to study the effects of Heiferplus, a sperm sexing agent on sperm characteristics, conception rate, sex ratio, abortion rate and maternal mortality in Goudali and F_1 Goudali-Simmental (Simgoud) cattle breeds. The first part of the study consisted of evaluating *in vitro*, the effects of Heiferplus on sperm characteristics. For the second part, 48 cows (300 - 350 kg) of each breed were divided into 2 groups (24 cows in parity 1 and 24 in parity 2). Each group was again subdivided into 12 cows each which were inseminated with semen containing Heiferplus and the other 12 without. Pregnancy diagnosis was done 28 days following artificial insemination and 60 days after, the sex of foetus was determined using trans-rectal ultra sonography. The motility of sperm with Heiferplus was 4.4 ± 0.55 and that of sperm without was 4.6 ± 0.55. The viability of sexed sperm was 84.92 ± 5.17and that of unsexed sperm was 75.46 ± 9.88. Sperm morphology showed 90.4 ± 1.67 normal sperm for semen with Heiferplus and 90.0 ± 1.58 for semen without Heiferplus. Conception rate in both breeds was improved by an average of 4.13% in Heiferplus cows as compared to the control group. Female calf percentage in Heiferplus cows was 72.09%, significantly higher than that of control. Breed, parity and body condition score had no significant effect on calf sex. Abortion rate (13.95% in Heiferplus cows and 14.63% in control) was not significantly affected by Heiferplus. Maternal mortality rate was 4.65% for Heiferplus cows and 4.88% for control, thus, no significant difference. It was concluded in this study that Heiferplus increases female sex ratio but has no significant effect on semen characteristics, conception rate, abortion rate and maternal mortality.

Keywords: *Semen; Sexing Agent; Sex Determination; Sex Ratio; Gudali; Simgud Cattle*

Introduction

The demand for predetermination of sex in livestock offspring is high. This is of critical importance to provide the most efficient production of the world's food supply. In agriculture over the past generation, the application of sex pre-selection to production systems becomes increasingly necessary [1]. In beef cattle, the increased growth rate and efficient production of lean meat by males have financial advantages [2]. In contrast, dairy farmers generally seek female dairy breed calves to perpetuate their herd while male beef crossbred animals are desired for reasons above, that is, increased growth rate and efficient production of lean meat [3]. In natural reproduction, the sex ratio is approximately 50:50 [4]. In the dairy cattle industry, only female animals (and very few breeding males) are productive and most male calves are slaughtered with little financial return to the farmers. Because of high market prices for replacement heifers, high milk prices, reproductive inefficiency of dairy herds and the shortage of heifers for replacements, sexed semen has been used to get more heifers [5]. The only accurate and potentially cost effective approach for achieving sex pre-selection at the present time involves separating the X-chromosome from the Y-chromosome bearing sperm followed by its use for artificial insemination (AI), or for *in vitro* fertilization (IVF) with subsequent embryo transfer (EF) [6].

Heiferplus is a commercial sperm sexing agent that works by enhancing the motility of the X-chromosome bearing sperm (female) and slowing the motility of the Y-chromosome bearing sperm (male). When inseminated, the sperm sort in the reproductive tract of the dam. The result is more ova fertilized by the X-chromosome bearing sperm (female), producing more heifer calves [7].

Considering the utilization of most assisted reproductive practices (IVF-ET), sexed sperm for production of livestock of preselected sex is dependent on efficiency, economics, effectiveness and ease of use.

To the best of our knowledge, little is known on the influence of the sexing agent on the fertility, particularly on tropical breeds. Thus, the aim of this study was to investigate the effects of Heiferplus, a commercial sexing agent on the fertility of 2 cattle breeds (Goudali and its crossbreed with Simmental).

Materials and Methods

Animals

Healthy 48 cows from Goudali breed and 48 cows of Simgoud breed, weighing 300 - 350 kg and from 3 to 6 years old were used in this study. They were lodged under field conditions and barrier was constructed with wires. They grazed on pastures dominated by *Pennisetum clandestinum* and *Trypsacum laxum*.

Sexing semen using sexing agent

Semen from Simmental breed preserved in liquid nitrogen at -195°C was warmed at 35 - 37°C and mixed with Heiferpplus at 0.50 ml of semen per vial Heiferplus warmed at 35 - 37°C to prevent cold shock to semen. The semen straw after being mixed with the sexing agent was incubated in water bath for 20 minutes at 35 - 37°C. The straw was removed and dried before loading the insemination gun and ready to inseminate.

Ethical consideration

Experimental protocols used in this study were approved by the Ethical committee of the Department of Animal Science of the University of Dschang (ECDAS-UDs 23/02/2015/UDs/FASA/DSAES) and was in conformity with the internationally accepted standard ethical guide lines for laboratory animal use and care as described in the European Community guidelines; EEC Directive86/609/EEC, of the 24thNovember1986 [8].

Experimental design

Forty-eight (48) cows of Goudali were divided into 2 groups of 24cows each which were first calving (parity 1) and 24 cows were second calving (parity 2). Each of the groups was subdivided into 12 cows each to be inseminated with Heiferplus and the other 12 without. The subgroups were divided into body condition scores 6,7 and 8 (i.e M^+, F^- and F) of 4 cows each. Each subgroup of parity was inseminated with semen from Simmental breed mixed with Heiferpplus. The other 12 cows for each parity groups were inseminated with pure semen to serve as control group. The same procedure was done for Simgoud.

Heat detection

Cows were observed for heat signs daily and each time, heat signs were confirmed using an electrical heat detector. The electrical heat detector was inserted into the vagina and observed for resistance in which a value greater than 250 confirms that the cow is truly on heat.

Artificial insemination

The cows were inseminated 24 hours after standing heat. The left hand of the inseminator was gloved with rubber gloves to shoulder-length and glove lubricant was applied. The loaded insemination gun was carried in the mouth and the cow was made to be aware of his presence. The cow's anus was cleaned and lubricated to relax. The vulva was also cleaned and the gloved hand was gently eased through the anus into the rectum. With the use of the other hand, the gun was inserted gently between the lips of the vulva into the vagina with the

help of the hand in the rectum. The semen was placed in the body of the uterus and the gun was gently withdrawn followed by the hand from the rectum.

Pregnancy diagnosis and determination of calf sex

Pregnancy diagnosis was performed by ultrasonic visualization of fetal heart beat with a 5MHz probe (Aloka, Wallingford, CT), 28 days following artificial insemination (AI). The embryo initially appeared as a short line (Day 20 - 22), later became C- shaped (Day 22 - 30) and finally, by day 30 - 32 of gestation assumed an L- shape.

Sex of fetus was determined 60 days after AI with the use of trans-rectal ultra sonography. This was done using easy scan ultrasound machine. Fetal sex was determined by the absence and/presence of the fetal genitalia as described by Romano and Masgee [9].

Evaluation of sperm characteristics

The volume of semen in each straw was 0.5 ml containing 20 million sperm cells. For individual motility, 50 µl of conventional semen was diluted with 500 µl of Ringer solution (physiological liquid); earlier placed in water bath at 37°C. A sample of diluted semen was placed on a pre-warmed slide and covered with a cover-slip. Individual motility was subjectively assessed using a phase contrast microscope at 40X magnification. A 6-point Scale as proposed by Baril., *et al.* [10] was used. (0 = No sperm motion, 1 = Very slow motion, trembling, or oscillation of the tail, 2 = Slow motion, trembling, uncoordinated motion, with a few rapid motions, 3 = Circular motions without trembling, 4 = Rapid motion, some with linear motion and some without linear motion, 5 = Rapid and linear motion of sperms). The same procedure was carried out for sexed semen.

Sperm viability was determined by hypo-osmotic swelling test (HOST). The procedure was that described by Lodhi., *et al*. [11]. and adapted by Jeyendran., *et al*. [12] for bull semen. The hypo-osmotic solution was prepared by dissolving 0.59g sodium citrate and 1g fructose in 11.11 ml of distilled water. The solution was stored at 4°C till used. A 1.0 ml of hypo-osmotic solution was mixed with 0.1 ml of undiluted conventional semen and incubated at 37°C for 1 hour. A drop of the diluted unsexed semen was placed on a clean sterilized dry glass slide and covered with a cover slip. A total of 200 spermatozoa were counted in different fields at 40X under phase contrast microscope and percentage of spermatozoa positive to HOS test (having coiled tails) was determined. A drop of diluted sexed semen was placed on another clean sterilized dry glass slide and viability of sexed semen was observed in the same manner.

Morphological aberrations were determined from a total count of 100 spermatozoa in smears colored with eosine. The slides were kept to dry and observed at 100X under a light microscope. Sperm abnormalities were classified into 2 groups: major abnormalities at the level of the head of spermatozoa and minor abnormalities at the tail [10]. Conception rate was obtained by [(number of pregnant cow/ number of females inseminated) X 100].

Sex-ratio in % was determined as number of female calves/total number of calves X 100.Abortion rate was obtained by number of abortions/total number of pregnant cows X 100. Maternal mortality rate was obtained by number of dead pregnant cows/total number of pregnant cows X 100.

Statistical analysis

Analysis of variance with multiple factors (ANOVA) was performed for comparison with post hoc Duncan test at 5%. Chi square was used to compare the percentages. A value of ($P < 0.05$) was considered statistically significant. Statistical analysis was performed with the aid of SPSS version 21 for Windows software programme.

Results

The effects of Heiferplus on sperm mobility, viability and morphology are summarized in table 1. The individual mobility and tail abnormalities of sperms were more elevated in semen without Heiferplus as compared to semen mixed with the sexing agent. The inverse

was obtained with the percentage of normal sperm. Statistical analysis didn't show any significant difference between treatments. The percentage of head abnormalities of sperms was not affected by Heiferplus. Although semen mixed with Heiferplus had a higher sperm viability, no significant difference was recorded when compared with semen without Heiferplus.

Treatments	Sperm characteristics				
	Morphology			Viability	I. mobility
	N. sperm (%)	A. sperm (%)			
		A. Heads (%)	A. Tails (%)		
Semen with H^+	90.80 ± 1.67[a]	2.80 ± 0.55[a]	6.40 ± 1.30[a]	84.92 ± 5.1[a]	4.4 ± 0.5[a]
Semen without H^+	90.00 ± 1.58[a]	2.80 ± 0.55[a]	7.20 ± 1.52[a]	75.46 ± 9.8[a]	4.6 ± 0.5[a]

Table 1: *Effects of Heiferplus on sperm characteristics.*

a, on the same column, the values with the same letter do not differ significantly (P>0.05).

I: individual, A: abnormal, N: normal, H^+: Heiferplus.

The effects of Heiferplus and cattle breed on conception rate are reported in table 2.

Breed and Treatment	Conception rate (%)
Goudali	
GHP (n = 24)	91.67[a]
G (n = 24)	87.50[a]
Simgoud	
SHP (n = 24)	87.50[a]
S (n = 24)	83.33[a]
All the breeds	
GHP + SH (n = 24)	89.58[a]
G + S (n = 24)	85.45[a]

Table 2: *Effects of Heiferplus and cattle breed on conception rate.*

n = number of cows inseminated.

a, on the same column, the values with the same letter does not differ significantly (P > 0.05).

GHP=goudali cows inseminated with Heiferplus; G=Goudali cows inseminated without heifer plus;

SHP= Simgoud cows inseminated with Heiferplus; S= Simgoud cows inseminated without Heiferplus.

Independently of cattle breed, the conception rate was higher in cattle inseminated with semen containing the product compared to those inseminated with semen without the product; however, no significant difference was noticed. Whatever the cattle breed, the conception rate was higher in cattle inseminated with sexed semen compared to the values of this parameter recorded in animals inseminated conventional semen. Statistical analysis didn't reveal any significant difference.

The effects of Heiferplus and cattle breed on calf sex are presented in table 3. It appears from this table that: Independently and whatever the cattle breed, cattle inseminated with semen mixed with the sexing agent registered a significantly higher percentage of female calves ($P < 0.05$) compared to male calves. On the other hand, although a higher value of female calves compared to males was recorded in animals inseminated with semen not containing the sexing agent, no significant difference was recorded ($P > 0.05$).

Breed and treatment	Male calves (%)	Female calves (%)
Goudali		
GH (n = 24)	27.27 $^{b\beta}$	72.73 $^{a\alpha}$
G (n = 24)	47.62 $^{a\alpha}$	52.38 $^{a\beta}$
Simgoud		
SHP (n = 24)	28.57 $^{b\beta}$	71.43 $^{a\alpha}$
S (n = 24)	45.00 $^{a\alpha}$	55.00 $^{a\beta}$
All the breeds		
GHP + SHP (n = 48)	27.92 $^{b\beta}$	72.09 $^{a\alpha}$
G + S (n = 48)	46.34 $^{a\alpha}$	53.66 $^{a\beta}$

Table 3: *Effects of Heiferplus and cattle breed on calf sex.*

n= number of cows inseminated.

a, b on the same column, the values with the same letter does not differ significantly (P>0.05).

α, ß on the same row, the values with the same superscript does not differ significantly (P>0.05).

GHP =goudali cows inseminated with heiferplus; G=Goudali cows inseminated without heifer plus;

SHP = Simgoud cows inseminated with heiferplus; S= Simgoud cows inseminated without heifer plus.

The effects of Heiferplus, cattle breed and parity on calf sex are summarized on table 4. When parity was considered, independently of cattle breed, in cows inseminated with semen containing the sexing agent, the percentage of female calves was higher at second calving as compared to those at first calving. The inverse was recorded in animals inseminated with conventional semen. Statistical analysis didn't show any significant difference (P > 0.05).

Breed and Parity	Cows inseminated with Heiferplus		Cows inseminated without Heiferplus	
	Male calves (%)	Female calves (%)	Male calves (%)	Female calves (%)
Goudali n = 48				
1	27.27^{b*}	72.73^{a*}	45.45$^{\alpha*}$	54.55$^{\alpha*}$
2	27.27 ^{b*}	72.73^{a*}	50.00$^{\alpha*}$	50.00$^{\alpha*}$
1 + 2	27.27 ^{b}	72.73 ^{a}	47.62$^{\alpha}$	52.38$^{\alpha}$
Simgoud n = 48				
1	30.00 ^{b*}	70.00^{a*}	36.36$^{\alpha*}$	63.64$^{\alpha*}$
2	27.27 ^{b*}	72.73^{a*}	55.56$^{\alpha*}$	44.44$^{\alpha*}$
1 + 2	28.57 ^{b}	71.43 ^{a}	45.00$^{\alpha}$	55.00$^{\alpha}$
Goudali + Simgoud n = 96				
1	28.57 ^{b*}	71.43^{a*}	40.91$^{\alpha*}$	59.09$^{\alpha*}$
2	27.27^{b*}	72.73^{a*}	52.63$^{\alpha*}$	47.37$^{\alpha*}$
1+2	27.91^{b}	72.09 ^{a}	46.34$^{\alpha}$	53.67$^{\alpha}$

Table 4: *Effects of Heiferplus, cattle breed and parity on calf sex.*

n= number of cows inseminated.

1, 2= parity 1, parity 2.

a, b on the same row, the values with the same letter does not differ significantly (P > 0.05).

α: on the same row, the values with the same superscript does not differ significantly (P > 0.05).

**, on the same column, the values with the same superscript does not differ significantly (P > 0.05).*

When breed was taken into account, independently of parity, the percentage of female calves was higher ($P < 0.05$) in females inseminated with semen containing Heiferplus as compared to that of females inseminated without the sexing agent.

Whatever the parity and breed, the percentage of female calves was higher in female inseminated with sexed semen compared to that of females inseminated with semen free of the sexing agent. In addition, whatever the breed, parity doesn't have any significant effect ($P > 0.05$) on calf sex when cows were inseminated with sexed or conventional semen.

Table 5 presents the effects of Heiferplus, cattle breed and body condition score on calf sex.

Breed and BCS	Cows inseminated with H^+		Cows inseminated without H^+	
	Male calves (%)	Female calves (%)	Male calves (%)	Female calves (%)
Goudali (n = 48)				
M^+(n = 16)	25.00^{b*}	75.00^{a*}	$50.00^{\alpha*}$	$50.00^{\alpha*}$
F^-(n = 16)	28.57^{b*}	71.43^{a*}	$42.86^{\alpha*}$	$57.14^{\alpha*}$
F(n = 16)	28.57^{bs*}	71.43^{a*}	$50.00^{\alpha*}$	$50.00^{\alpha*}$
All the BCS M^++F^-+F	27.38^{b}	72.62^{a}	47.62^{α}	52.38^{α}
Simgoud (n = 48)				
M^+(n = 16)	28.57^{b*}	71.43^{a*}	$62.50^{\alpha*}$	$37.50^{\alpha*}$
F^-(n = 16)	37.50^{b*}	62.50^{a*}	$42.86^{\alpha*}$	$57.14^{\alpha*}$
F(n = 16)	16.67^{b*}	83.33^{a*}	$60.00^{\alpha*}$	$40.00^{\alpha*}$
All the BCS M^++F^-+F	27.58^{b}	72.42^{a}	55.12^{α}	44.88^{α}
Goudali+Simgoud (n = 96)				
M^+(n = 16)	26.67^{b*}	73.33^{a*}	$56.25^{\alpha*}$	$43.75^{\alpha*}$
F^-(n = 16)	33.33^{b*}	66.67^{a*}	$42.86^{\alpha*}$	$57.14^{\alpha*}$
F(n = 16)	23.08^{b*}	76.92^{a*}	$54.55^{\alpha*}$	$45.45^{\alpha*}$
All the BCS M^++F^-+F	27.69^{b}	72.31^{a}	51.22^{α}	48.78^{α}

Table 5: *Effects of Heiferplus, cattle breed and body condition score on calf sex.*

n: number of cattle inseminated; M+: more medium; F-: less full; F:normally full; H+:Heiferplus.

a, b on the same row, the values with the same letter does not differ significantly ($P > 0.05$).

α, on the same row, the values with the same superscript does not differ significantly ($P > 0.05$).

**, on the same column, the values with the same superscript does not differ significantly ($P > 0.05$).*

BCS= Body Condition Score.

Independently of cattle breed, whatever the body condition score, the percentage of female calves was significantly ($P < 0.05$) higher in cattle inseminated with semen containing Heiferplus as compared to those inseminated without Heiferplus. The same results were obtained when breed was taken into consideration.

The effects of Heiferplus and cattle breed on abortion rate are presented in table 6. Independently of breed, cattle inseminated with semen not containing Heiferplus showed a non-significantly higher ($P > 0.05$) abortion rate than compared to those inseminated with semen containing the sexing agent.

When breed was taken into consideration, breed recorded a higher abortion rate than breed in animals inseminated with sexed semen, though this difference wasn't statistically significant ($P > 0.05$). On the contrary, a higher abortion rate was noticed in cattle inseminated without the sexing agent. This difference was still not significant.

Breed and treatment	Number of pregnant cows	Abortion rate (%)	Mortality rate (%)
GHP (n = 24)	22	13.64[a]	9.09[a]
G (n = 24)	21	19.05[a]	4.76[a]
SHP (n = 24)	21	14.29[a]	0.00[a]
S (n = 24)	20	10.00[a]	5.00[a]
GHP + SHP (n = 48)	43	13.95[a]	4.65[a]
G + S (n = 48)	41	14.63[a]	4.88[a]

Table 6: *Effects of Heifeplusand cattle breed on abortion and maternal mortality rates.*

n= number of cows inseminated, Numbers in brackets under abortion rate indicate number of abortions while those under mortality rate indicate number maternal deaths.

a, on the same column, the values with the same letter does not differ significantly (P > 0.05).

GHP= goudali cows inseminated with heiferplus; G= Goudali cows inseminated without heiferplus; SHP= Simgoud cows inseminated with heiferplus; S= Simgoud cows inseminated without heiferplus.

SHP= Simgoud cows inseminated with heifer plus; S= Simgoud cows inseminated without heifer plus.

Independently of breed, cattle inseminated with semen without showed a non-significantly higher (P > 0.05) maternal mortality rate than those inseminated with semen mixed with the sexing agent. When breed was taken into account, cattle inseminated with semen containing registered a higher mortality rate as compared to cattle which had no mortality, though the difference was not statistically significant (P > 0.05). No significant difference was equally recorded in cattle inseminated with conventional semen.

Discussion

Studies on sperm motility, viability and morphology showed that motility of sperm cells in sperm without Heiferplus was better than that of semen with sexing agent but this difference was not statistically significant (P > 0.05). Viability of sperm didn't register any significant difference between semen with the sexing agent and unsexed semen. This doesn't agree with Minervini., *et al*. [13], who stated that treatment of buffalo semen with Heiferplus showed a significant (P < 0.05) reduction in sperm viability. There was no significant difference between morphology of sperm with and without Heiferplus. According to DeJarnette., *et al*. [14], the average fertility of sexed semen is about 75% of the fertility of conventional semen in virgin heifers (a decrease of 20 to 30%). The results of this study do not agree with this since semen quality (morphology) which has a significant predictive power in terms of fertility [15,16]; was not affected by the sexing agent.

The average increase in conception rate (4.13%) recalls the 5 - 10% increase in conception rate when the semen contains Heiferplus [7]. The 91.67% conception rate in Goudali, recalls the 90% conception rate compared to sperm without the sexing agent recorded by Deutscher., *et al*. [17]. The lower conception rate (87.50%) in Simgoud can be because of a slight fault in AI. Semen deposition in the uterine horn is undesirable because the non-ovulating uterine horn might be selected or injury may occur. (Pregnancies occur more frequently in the right uterine horn of cows). Deposition of semen in the vagina results in dilution, contamination and lowered conception rate [18]. Of course, when a bull inseminates a cow naturally, approximately 5 billion spermatozoa are deposited in the vagina. However, when semen is deposited artificially into the cervix considerably fewer sperms are required to achieve conception [18]. Transport of the sperm is achieved by wave-like contractions of the uterus, caused by the hormones prostaglandin and oxytocin. Rough handling of the cow can lead to release of adrenalin, which stops these contractions occurring and may result in a lower conception rate [19]. However, the results obtained in the present study disagree with Andersson., *et al*. [20], who showed a decrease of 25% conception rate in dairy lactating cows inseminated with sexed sperm as compared to conventional sperm.

The study showed a percentage of 72.73% female calves for Goudali breed inseminated with semen mixed with the sexing agent as compared to those inseminated without the sexing agent. That of Simgoud was 71.43% female calves for females inseminated with semen containing the sexing agent and 55% for those inseminated without the sexing agent. This showed that the increase of 18.43% in the expected number of female calves independently of cattle breed when inseminated with sexed sperm is lower compared to 20% recorded by Brusveen., *et al*. [21]. The 20.35% increase in Goudali is similar to the 20% that was found by Brusveen., *et al*. [21]. The lower percentage in Simgoud can be explained by the fact that, a lower number of females were pregnant in Simgoud than in Goudali cattle. There was no significant difference between male and female calf sex ratio in cows inseminated with conventional sperm in both breeds. This is in disagreement with the findings of Singh., *et al*. [22] and Kaygisiz and Vanli, [23], who stated that breed has an influence on sex ratio. The difference between cows with sexed and unsexed sperm can be explained by the action of the sexing agent which increases the percentage of female calves.

The percentage of female calves was greater in cows at first calving inseminated with semen mixed with Heiferplus as compared to those at second calving though no significant difference was recorded. The reverse was observed in females inseminated with the unsexed semen. These results from the two breeds were in agreement with findings of Roche., *et al*. [24]; Kaygisiz., *et al*. [25] and Lari [26], who also reported a non-significant association between calf sex and parity. This corresponds to the results found in this study. However, results of the present study were not consistent with Singh., *et al*. [22], who stated that sex was influenced by parity. The significant difference between female calves in cows with sexed and unsexed semen can only be explained by the action of the sexing agent which increases female calf percentage.

In females inseminated with sexed semen the highest percentage of female calves as compared to male calves was registered in body condition score F and in those inseminated with semen without the Heiferplus, the highest was recorded in body condition score F^{-}. These results disagree with the predictions that females in better body condition would produce more male than female progeny observed in red deer [27-31] reindeer [32], Barbary sheep [33], domestic pigs [34] and a number of other species, although there are exceptions [35,36]. It was remarked that body condition scores had positive effect on secondary sex ratio [24]. Thus, the higher female calve sex ratio in cows with semen containing Heiferplus as compared to those without the sexing agent is due to the presence of the product and not the Body Condition Score effect.

Abortion rate was non significantly higher ($p > 0.05$) in females inseminated with semen without Heiferplus as compared to those inseminated with semen containing the sexing agent. This means that though the composition of Heiferplus is not revealed by the producer, we can be sure that it does not contain substances in quantities that can cause abortion in cows. The results obtained in this study agree with those found by Tubman., *et al*. [37], who stated that sexed sperm does not increase abortion rate when compared to controls. Abortion rates can be influenced by environment. Bulls and other factors associated with artificial insemination may also influence abortion rates [37]. There are many factors that cause embryonic death in cows. These are endocrine, genetic, climate, stress, age, insemination time, semen quality, infectious agents, nutrition, chromosomal anomalies. Especially, abnormal progesterone and estrogen profiles cause embryonic deaths. Moreover, in high producing cows steroid metabolism is faster because of liver blood circulation increase. This causes lower levels of progesterone in luteal period of estrus cycle [38,39].

In females inseminated with semen not containing the sexing agent, the maternal mortality rate was higher, though this difference was not significant. The maternal mortality can be due to any other factor and not Heiferplus. Generally, no literature has yet been found in order to compare the maternal mortality rate with.

Conclusion

From the study of the effects of Heiferplus, a commercial sexing agent on the fertility of Goudali and Simgoud, we can bring out the following conclusions:

1. Fertility is not affected by Heiferplus as the motility, viability and morphology of sperm was not influenced by this sexing agent.
2. Heiferplus did not have any effect on conception rate.
3. Heiferplus increases female sex ratio in Goudali and Simgoud cattle breeds.
4. Body condition score of Goudali and Simgoud have no influence on calf sex.
5. Heiferplus reacts the same way in Goudali and Simgoud of both parities considered in the study.
6. Conception rate is not affected by Heiferplus whatever the cattle breed. Heiferplus doesn't have any effect on abortion rate and maternal mortality.

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Conflict of Interest

None of the authors have any conflict of interest to declare.

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