Full length paper

Reproductive Efficiencies of Cows and Heifers Artificially Inseminated with Sex-Sorted and Conventional Semen in Bhutan

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ARTICLE HISTORY	A B S T R A C T			
Received: 25/11/18 Peer reviewed: 12/12/18 Received in revised form: 24/12/18 Accepted: 01/01/19	The study objective was to compare conception rates and sex of calf at birth from Artificial Insemination [AI] of cows and heifers, using sex-sorted [sexed] and conventional [unsexed] semen. Imported frozen semen from two sire lines for both sexed and conventional semen that had equivalent Post Thaw Motility of 40% was used. Predictors of conception rates and sex of calf at birth were semen type, animal type, insemination sire, number of AI per conception, AI Technician and breed. In total, 422 AI was done in 160 heifers and 130 cows of Jersey pure, Jersey cross and local cattle, of which 151 and 271 were sexed semen and conventional semen, respectively. The conception rates of 44% and 48% were recorded for sexed and unsexed semen respectively.			
Keywords	conventional semen, difference was not statically significant.			
Artificial insemination Conception rate Conventional semen Reproduction Sex ratio Sexed semen	Sexed sorted semen gave birth to 89.6% female calves as compared to 51.5% for conventional semen, indicating a strong influence of semen type on birth of female calves. Conception rate was significantly influenced by animal type; cow and heifer and not by other predictors. Conception rate was significantly lower in cows than in heifers. Hence, expensive sexed semen can be recommended for use in first-service heifers than in cows for optimizing conception rates.			

1. INTRODUCTION

In Bhutan, cattle breeding is carried out commonly through Artificial Insemination [AI]. The imported conventional semen and locally produced frozen semen are used for AI in accessible areas, and tested breeding bulls are used in inaccessible areas. This intervention has enabled government to organize dairy production system through formation of dairy farmers' groups and cooperatives. In the 11th Five Year Plan [2013-2018], the Department of Livestock [DoL] imported 2055 dairy animals from India [NDRC 2018] and supplied to interested farmers on cost sharing basis. Such intervention, although inevitable, poses high risks of introducing exotic diseases into the country despite having strict quarantine measures in place. Thus, to minimize such risks and also to address the problem of meeting dairy cattle demand within the country, NDRC imported and initiated field trial on AI with sex-sorted semen in Bhutan from 2014.

The use of sex sorted semen is efficient at commercial level to produce desired number of replacement heifers, particularly at herd level. Its application in virgin heifers has advantages in improving herd turnover rate, improving genetic progress, reducing price for superior replacement heifers and reducing costs of milk production [De Vries et al. 2008]. Generally, sexed semen has been proven to have lower conception rate, compared to conventional semen [Andersson et al. 2006; Cerchiaro et al. 2007; Norman et al. 2010; Norman et al. 2011; Healy et al. 2013; Karakaya et al. 2014; Sahereh et al. 2017]. Due to reduced conception rate, sex sorted semen is used usually in virgin heifers. It was ascertained that the use of sex sorted semen in second or later services could reduce conception rate, compared to first service heifers [De Jarnette et al. 2009]. However, these traits of sexed semen have not been tested in Bhutan. Therefore, a study was conducted with the primary objective to assess and compare the reproductive efficiency of sex-sorted and conventional frozen semen in cows and heifers under the farm and field conditions of Bhutan.

2. MATERIALS AND METHOD

2.1 Data collection

Data were collected from August 2014 to March 2018, from cows and heifers inseminated with 151 sex-sorted semen and 271 conventional semen (Figure 1). It covered animals from government farms viz. National Jersey Breeding Centre

Bhutan Journal of Animal Science 2019, 3 [1]: 37-41

(NJBC) at Samtse with Pure Jersey (PJ) and Cattle Rearing Centre (CRC) at Wangkha with Jersey Cross (JX) and farmers' field from three districts, namely Sarpang, Tsirang and Samdrupjongkhar with JX and local cattle (Table 1). The selection of government farms was based on availability of breeds of animals in the farms required for parallel inseminations, using both type of semen by equally fluent AI Technicians, and the districts to ensure application of same protocol, procedures and techniques by the same team. AI was performed under natural heat and after synchronization of estrus in the government farms and farmers' field. The numbers of inseminations by locations are presented in Figure 2.

Data collected for analysis were animal identity, insemination and calving date, natural/induced heat, animal type [cow/heifer], service number, semen type [sexed/conventional], AI technician, sex of calf and incidence of twining. AI was carried out by three experienced AI Technicians, using two sire lines each of sexed and unsexed imported semen straws. The approximate sperm dosage per straw in sexed and conventional semen were 2×10^6 and 20×10^6 , respectively, with 40% post thaw motility of spermatozoa in both semen type.

In two government farms, AI was performed under natural heat during standing oestrus [12-14 hours after onset of heat signs], whereas in farmers' field, AI was performed in synchronized animals that exhibited clear heat signs, upon per-rectal palpation, after 48-52 hours post removal of synchronizing device. The agent used for synchronization was 1.9 g Progesterone Releasing Internal Device [PRID] inserted in vagina for 7 days. All inseminations were performed in the uterine body. Conception rate was obtained by the reciprocal of the average number of services per conception. Service numbers greater than six were excluded from the data.



Figure 1: Number of insemination records and animal type.

Table 1: Number of animals b	y location included in the study	
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	Government farms				Farmers' field Sarpang/ Tsirang/ Sjongkhar		 Total
Location	NJBC-Samtse		CRC-Wangkha				
Animal category	Heifer	Cow	Heifer	Cow	Heifer	Cow	
Jersey pure	9	63					72
Jersey cross			89	8	50	52	199
Local					12	7	19



2.2 Statistical analysis

Analysis was performed using the Statistical Package for Social Science [SPSS] version 23. Multiple regression was used to determine the conception rate and influence of predictors on conception rate and calf sex. Analysis was performed by pooling data from all five locations together and assessed the reproductive efficiencies of cows and heifers inseminated with sex sorted and conventional semen. The possible sources of variation in conception rate were semen type, animal type, natural/induced heat, insemination sires and AI technician, and semen type for sex of calf.

3. RESULTS AND DISCUSSION

3.1 Conception rate and the number of services per conception

The overall conception rates [CRs] recorded for sex-sorted semen and conventional semen in this study were about 44% and 48%, respectively [Figure 3]. The fertility for sex-sorted semen in cows and heifers was lower. Although, statistically not significant, this study result was found consistent with the findings that the sex-sorted semen produced lower fertility results than conventional semen [Karakaya et. al 2014]. In this study, CRs with conventional semen is 11% higher than the CR reported by Tshering and Tamang [2018].

The overall CRs of 44.4% for sexed semen and 48% for conventional semen in this study are comparable with the findings of Sharma et al. [2018] in Indian *Bos indicus* cows, with 40% for sexed semen and 49% from conventional semen. When CRs were compared by animal type, CR in heifer was recorded slightly higher than those reported by other authors whose mean heifer conception rates were 39% for sexed semen and 56% for conventional semen [Cerchiaro et al. 2007; Chebel et al. 2010; Norman et al. 2010; Karakaya et al. 2014; Noonan et al. 2016]. However, CRs in heifer with sexed semen [47.7%] and conventional semen [59.1%] were much lower than the findings of De Jarnette et al. [2009] who reported CRs of 53% for sexed semen and 80% for conventional semen in Jersey heifers. Overall, in this study, CRs in cows and heifers from sexed semen were within 70 - 90% reported for conventional semen [Manzoor et al. 2017, SaFilo et al. 2014]. This study revealed that, CR was not influenced by semen type, natural/induced heat, insemination sires and AI technicians, but was influenced significantly [p≤0.05] by animal type [Table 2].This could be due to good health of heifers, leading to better fertility and subsequent higher conception rates than cows that may have lost body condition over successive calvings.





3.2 CR and sex-sorted semen

There was no correlation between CR and sex-sorted semen, but slight correlation of CR with AI Technician and animal type was observed only for conventional semen [Table 3], which was not coherent with the findings of Sahereh et. al. [2017] who reported strong correlation between CR and animal type for both semen type.

The number of services per conception [AI Index] for Sex-sorted semen in heifer and cow were 2.1 and 2.7 AIs, respectively, and for Conventional semen in heifer and cow were 1.8 and 2.4 AIs, respectively. The effect of number of services on decreasing conception rates was similar for sexed and conventional semen inseminations. Decreasing conception rates with increasing number of services was in agreement with the conception rates reported by few authors [De Jarnette et al. 2009; Norman et al. 2010]. Therefore, sexed semen straws should be reserved for the first and second services because a significant reduction in conception rate associated with increased number of services results in low cost-to-benefit ratios as ascertained by Sahereh et al. [2017].

3.2 Sex ratio

Female calf sex ratios obtained for sexed and conventional semen in cows and heifers in all study locations are illustrated in Figure 4. Regression analysis confirmed that the calf sex was significantly dependent on the type of semen used [p<0.000, adj. R.Sq=0.137], which is in concurrence with other findings [Sahereh et. al. 2017; Sharma et al. 2018]. Average female calf ratio obtained from the use of sexed semen was 89.6%. Similar ratio between 85 and 91% has been reported [De Jarnette et al. 2009; Norman et al. 2010; Healy et al. 2013].

The female sex ratio for conventional semen obtained in this study was 51.5%, which is in agreement with the findings of Norman et al. [2010] with 48.5% single males born to heifer dams, in contrary to most trials, which reported 50 to 52% males [Tubman et al. 2004; De Jarnette et al. 2009]. No twin birth was recorded in the study.



Figure 4: Female calf sex ratio [%] in cows and heifer dams by semen type.

Table 2: Overall	conception rate	with different	predictors.
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Predictors	Variables	Conception rate [%]	n	p-value	adj. R. Sq.
Semen type	Sexed semen	44.4	151		
	Conventional semen	48.0	271	0.479	001
Heat type	Natural	47.5	301		
Tiedd type	Induced	44.6	121	0.593	002
Insemination sire	130128	47.5	59		
	302761	42.4	92		
	303587	51.7	29	0.851	002
	115479838	47.5	242		
AITech	Leki_NJBC	42.2	180		
Ai Ittii	Bijay_CRC	55.4	121	0 517	- 001
	Abi_Ext.	44.6	121	0.517	001
Animal type	Heifer	52.8	199	0.019	0.011
J 1	Cow	41.3	223	0.018	0.011

Table 3: Conception rate of Sexed and Conventional semen with different predictors.

Predictors Heat type	Variables	ariables Sexed seme		Conventional semen =	n [%]
	Natural Induced	28/59 39/92	[47.5] [42.4]	115/242 [15/29 [47.4] 51.7]
	p-value	0.544 [adj.R.So	q.=004]	0.670 [adj.R.Sq.=-	.003]
AI Tech	Leki_NJBC	9/19	[47.4]	67/161	41.6]
	Abi_Ext.	19/40 39/92	[47.5] [42.4]	48/81 [15/29 [59.3] 51.7]
	p-value	0.851 [adj.R.So	q.=005]	0.043 [adj.R.Sq.=0.011]	
Breed	Jersey pure	12/25	[48.0]	71/165	43.0]
	Local	5/16	[43.5]	1/3 [49.4] 33.3]
	p-value	0.345 [adj.R.So	q.=001]	0.068 [adj.R.Sq.=0	.009]
Animal type	Heifer Cow p-value	53/111 14/40 0.166 [adj.R.Sc	[47.7] [35.0] [.=0.006]	52/88 [78/183 [0.011 [adj.R.Sq.=0	59.1] 42.6] .020]

4. CONCLUSIONS

The study concluded that CR, using conventional semen, is higher than the sexed semen. However, CR was not statistically significant for both semen types. Reproductive waste management via synchronization of animals not coming to heat at breed-able time and their insemination in proper heat were effective because no significant difference in CR was observed between insemination in natural heat and induced heat.

Overall CR was significantly lower in cows than in heifer dams. Overall calf sex ratio was higher for sexed semen with more female calves than the sex ratio for conventional semen. Thus, female birth is greatly influenced by the semen type used. Hence, expensive sexed semen could be best reserved for use in first-service heifers to optimize conception rates and the return on investment, as it would enable farmers to increase the number of available replacement heifers, while doing away with the burden of having unwanted male calves born in the herd.

Acknowledgements

The authors greatly acknowledge the cooperation and support of farm management; NJBC, Samtse and CRC, Wangkha as well as Dzongkhag Livestock Sectors; Sarpang, Tsirang and S/jongkharin on record keeping, data furnishing and logistic while conducting this study. The endeavor of the Department of Livestock in disseminating such technical information via publication in Bhutan Journal of Animal Science is highly appreciated.

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