

## Comparative Efficacy of Conventional and Sexed Semen on the Conception Rate of Achai Cattle and Sex Ratio of Calves at Zam Zam Dairy Farm, Bannu

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### Abstract

This study evaluated the comparative efficacy of conventional and sexed semen in Achai cattle at Zam Zam Dairy Farm, Bannu during 2023-24. Sixty healthy Achai cows (1st-3rd parity) were equally divided into two groups - one inseminated with conventional Jersey semen and the other with sexed Jersey semen. Estrus detection was performed thrice daily, followed by artificial insemination using standard protocols. Pregnancy diagnosis through rectal palpation at 60-90 days post-insemination revealed significantly higher ( $p < 0.05$ ) conception rates with conventional semen (56.67%) compared to sexed semen (23.33%). While sexed semen produced 100% female calves, the substantially lower conception rate highlights important practical limitations for its use in lactating indigenous cattle under field conditions.

**Keywords:** semen, parity, jersey, insemination, lactating, indigenous

## Introduction

The global dairy industry faces constant pressure to improve productivity and efficiency, with herd replacement strategies playing a crucial role in this endeavor [1]. Traditional breeding methods typically yield approximately equal numbers of male and female offspring, creating an economic burden as male dairy calves have limited value in milk production systems [2]. The development of sexed semen technology led to revolutionized dairy herd management by enabling producers to obtain over 90% female offspring through artificial insemination (AI) [3]. This technology, based on flow-cytometric separation of X and Y chromosome-bearing spermatozoa, demonstrated a significant advancement in genetic selection and herd improvement strategies [4].

Regardless of these advantages, several challenges hinder the wide-spread implementation of sexed semen technology in commercial dairy operations [5]. Several studies have exhibited reduced conception rates with sexed semen compared to conventional semen, particularly in lactating cows [6]. The sorting process subjects sperm cells to significant chemical and physical stresses that may compromise their fertilizing capacity and viability [7]. Furthermore, sexed semen straws contain considerably fewer sperm cells (approximately 02 millions) compared to conventional straws (15-20 millions), further reducing the probability of successful conception [8]. These limitations become particularly significant, while considering the application of this technology in indigenous cattle breeds under tropical conditions.

The Achai cattle breed, native to northern areas of Pakistan, demonstrated an important genetic resource due to its disease resistance, heat tolerance, and adaptation to local environmental conditions [9]. While cross-breeding with exotic Jersey semen has been practiced to improve milk production, there remains inadequate information regarding the efficacy of sexed semen in this indigenous breed [10]. This trial designed to compare conception rates between conventional and sexed Jersey semen in Achai cattle, while evaluating the resulting calf sex ratio, thereby providing valuable insights for smallholder dairy farmers considering this reproductive technology.

## Literature Review

Oikawa et al. [11] indicated that conception rates with sexed semen are lower than with conventional semen, posing challenges for dairy farming. Study on HF heifers in Japan were observed with conception rates of 56.9% with conventional semen versus 47.3% with sexed semen. Fertility reduction were more pronounced in warmer months (June–August), suggested seasonal impacts. Moreover, heifers inseminated with sexed semen were younger by about 21 days. These results shown the need for optimized A-I strategies, particularly in warmer periods, to improve sexed semen success.

Lu et al. [12] conducted a field study involving 4,521 crossbred (F1 or F2) buffaloes inseminated with X-sorted river buffalo sperm, where a pregnancy rate of 48.5% (2,194/4,521) was achieved, with a high sex accuracy rate of 87.6% (1,895/2,163) in the resulting calves. Particularly, semen from Murrah bulls yielded a significantly higher pregnancy rate (52.5%; 895/1,706) compared to Nili-Ravi bulls (46.1%; 1,299/2,815) ( $P < 0.01$ ), and technician skill also influenced success rates ( $P < 0.01$ ). In contrast, in Holsteins, parity (heifers vs. multiparous cows) did not affect pregnancy rates in buffaloes. Moreover, no significant fertility differences were observed among buffaloes of varying genetic backgrounds (swamp, F1, or F2 crossbreds) when inseminated with sexed semen. These conclusions highlight the adaptableness of sperm sexing technology in buffalo breeding under field conditions, supporting its potential application in small-scale farming systems with diverse genetic backgrounds. Additional research should focus on

refining insemination protocols to enhance conception rates and ensure wider adoption in village-based buffalo production systems.

Healy et al. [13] conducted a study conducted in a commercial Australian dairy herd in central western New South Wales between 2004 and 2009 analyzed retrospective data from 9,870 inseminations of 4,456 nulliparous Holstein heifers. The trial engaged mixed models to evaluate factors such as environmental conditions (temperature and humidity), insemination sire, AI technician, service number, and heifer age and weight at breeding. The conclusions exhibited that conception rates were significantly lower with sexed semen (empirical rate: 31.6%; model-adjusted: 21.3%) compared to conventional semen (empirical: 39.6%; model-adjusted: 32.1%). Key factors influencing conception included heifer age, semen type (sexed vs. unsexed), service number, environmental conditions, and A.I technician. Particularly, sexed semen produced a significantly higher proportion of female calves (86%) in comparison to conventional semen (48%), confirming its effectiveness in sex selection. Supplementary predictors of calf sex included insemination sire and gestation length. Twinning rates were comparatively higher (3.6%) for both semen types, with gestation length and heifer weight at breeding identified as significant influencing factors. Abortion rates were observed similar between sexed (6.1%) and unsexed (6.5%) conceptions, with heifer age at breeding playing a distinguished role. Still-birth rates were influenced by twinning, gestation length, calf sex, and A-I technician, with semen sorting and environmental conditions display marginal significance. Significantly, no developmental abnormalities were observed in off-spring, though sexed calves exhibited a slightly higher stillbirth rate, which warrants further investigation.

Hosseini-Zadeh et al. [14] employed stochastic bio-economic model to evaluate the impact of artificial insemination strategies on genetic progress and profitability in dairy herds. Research designates that using sexed semen significantly boosts genetic progression compared to conventional semen, particularly over a 15-year period. While theoretical genetic gains for milk production surpass realized gains, expanding herds demonstrate higher genetic means than fixed-sized herds. Economically, A-I with conventional semen yields higher net profits in expanding herds, whereas fixed herds often face decreasing profitability. Contrariwise, sexed semen generates high long-term profits, especially in growing herds. Sensitivity analyses demonstrated that fluctuations in milk revenue and feed costs substantially influence the net profitability of both A-I strategies. These results shown the economic and genetic advantages of tailored breeding programs in dairy production systems.

Otava, [15] evaluated sexed and conventional semen characteristics, such as sperm concentration, motility, and progressive movement, has been explored using Computer-Assisted Sperm Analysis (CASA). It had shown significant differences between the two semen types, with conventional semen exhibiting higher sperm counts ( $15.4 \times 10^6$  per straw), total motility ( $5.2 \times 10^6$ ), and progressive motility ( $3.3 \times 10^6$ ) compared to sexed semen, which exhibited markedly lower values ( $2.1 \times 10^6$ ,  $0.25 \times 10^6$ , and  $0.2 \times 10^6$ , respectively). Besides, artificial insemination trials involving heifers revealed distinguished differences in conception rates between the two semen types. In an experimental trial of 46 heifers, those inseminated with sexed semen (n=22) had a conception rate of 31.81%, whereas those inseminated with conventional semen (n=24) accomplished a significantly high rate of 75%. This concluded that sexed semen may result in condensed fertility compared to conventional semen. However, the use of sexed semen confirmed a higher proportion of female offspring, aligning with its intended purpose of sex selection.

Cerchiaro et al. [16] conducted a study evaluated the fertility and purity of commercially available sexed semen from four proven Holstein-Friesian bulls, used for artificial insemination (AI) in nulliparous heifers across 61 dairy farms in northern Italy. The analysis included data from 536 artificial insemination attempts with pregnancy diagnoses and 258 calving, employing logistic regression to assess various influencing factors. The overall pregnancy rate with sexed semen was observed at 51%, though significant differences were observed depending on the bull and the year of insemination. Particularly, semen from two bulls resulted in significantly lower conception rates compared to others, signifying inherent differences in the fertility of sexed semen among sires. Moreover, the purity of sexed sperm, measured by the proportion of female calves born, was consistently high at 87%, with no significant influence from factors such as heifer age, season, inbreeding level, or sperm dosage.

### **Materials and Methods**

The study was conducted at Zam Zam Dairy Farm located in Bannu district, which experiences a semi-arid climate with summer temperatures frequently exceeding 40°C. Sixty clinically healthy Achai cows in their first to third parity were selected based on normal reproductive history and body condition scores between 2.5-3.5 (on a 5-point scale). Animals were randomly allocated into two equal groups, with one group receiving conventional Jersey semen and the other receiving sexed Jersey semen (X-sorted sperm only) from high genetic merit bulls.

Estrus detection was performed three times daily (0600, 1200, and 1800 hours) by trained technicians using established behavioral and physiological indicators including standing heat, clear mucus discharge, and vulvar swelling. Cows exhibiting estrus signs were inseminated approximately 12 hours after initial detection following the AM/PM rule. Frozen semen straws (0.25 ml) were thawed in a water bath maintained at 37°C for 60 seconds immediately prior to insemination. The recto-vaginal technique was employed for all artificial inseminations, with semen deposited in the uterine body by an experienced technician. Pregnancy diagnosis was conducted through rectal palpation by a qualified veterinarian between 60-90 days post-insemination. Data on conception rates and calf sex (for pregnant animals) were recorded and statistically analyzed using chi-square tests to determine significant differences between treatment groups.

### **Results**

The reproductive performance analysis revealed significant differences between the two semen types. In the conventional semen group, 17 of 30 inseminated cows (56.67%) were confirmed pregnant, while the sexed semen group showed only 7 pregnancies out of 30 attempts (23.33%). This substantial difference in conception rates was statistically significant at  $p < 0.05$ . Regarding calf sex ratio, all seven pregnancies achieved with sexed semen resulted in female calves (100%), confirming the technology's effectiveness for sex selection. In contrast, the conventional semen group produced nine female and eight male calves, approximating the expected 50:50 sex ratio. Further analysis of non-pregnant animals demonstrated that cows in the sexed semen group required more repeat services (76.67%) compared to those in the conventional group (43.33%). Parity effects were also observed, with first-calf heifers showing better conception rates with sexed semen (30%) compared to multiparous cows (20%), though the small sample size limits definitive conclusions regarding parity-specific effects.

**Table 1. Conception rates of Achai cattle inseminated with conventional and sexed semen**

Semen Type	No. of Cows Inseminated	No. Pregnant	Conception Rate (%)	p-value
Conventional	30	17	56.67	<0.05
Sexed (X-sorted)	30	7	23.33	

Table 1. Conventional semen achieved significantly higher conception rates (56.67%) compared to sexed semen (23.33%,  $p<0.05$ ).

**Table 2. Calf sex ratio resulting from conventional and sexed semen insemination**

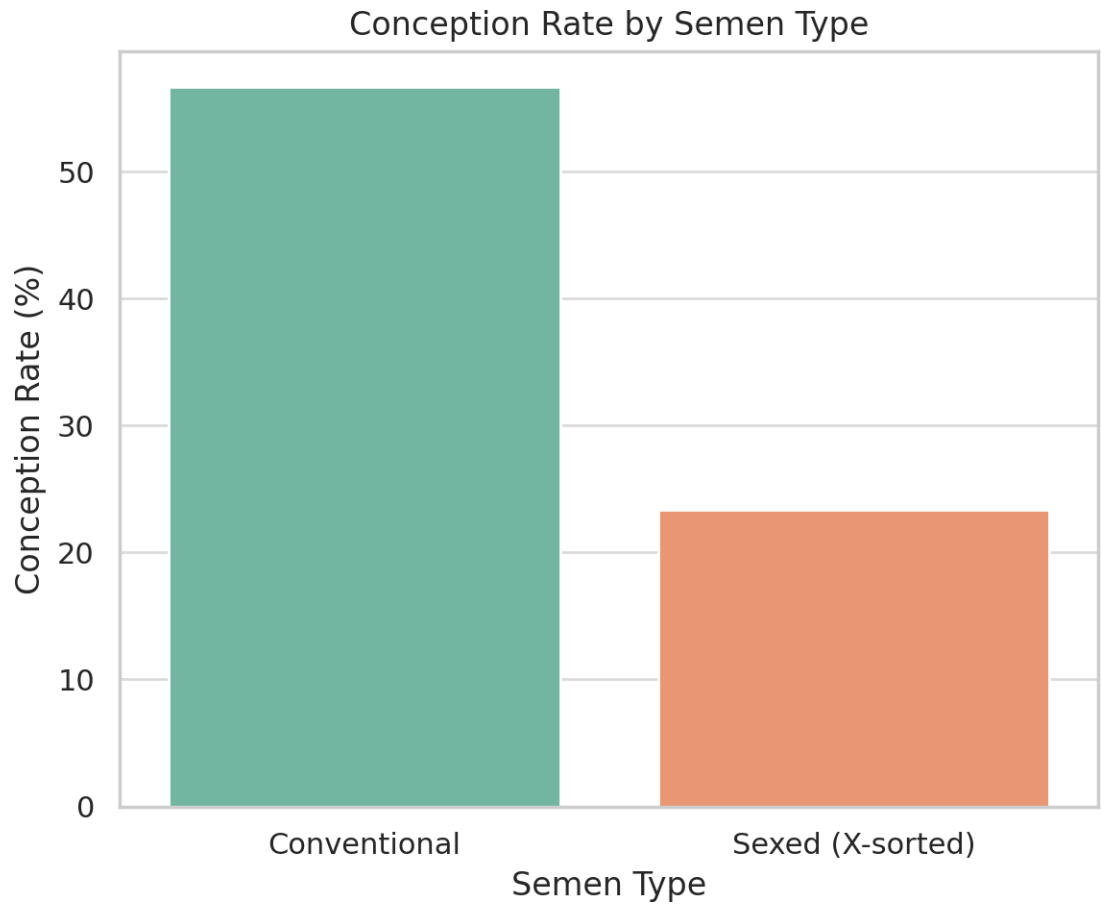
Semen Type	No. Pregnant	Female Calves	Male Calves	Female:Male Ratio
Conventional	17	9	8	53:47
Sexed (X-sorted)	7	7	0	100:0

Table 2. Sexed semen produced 100% female calves while conventional semen maintained the expected 50:50 sex ratio.

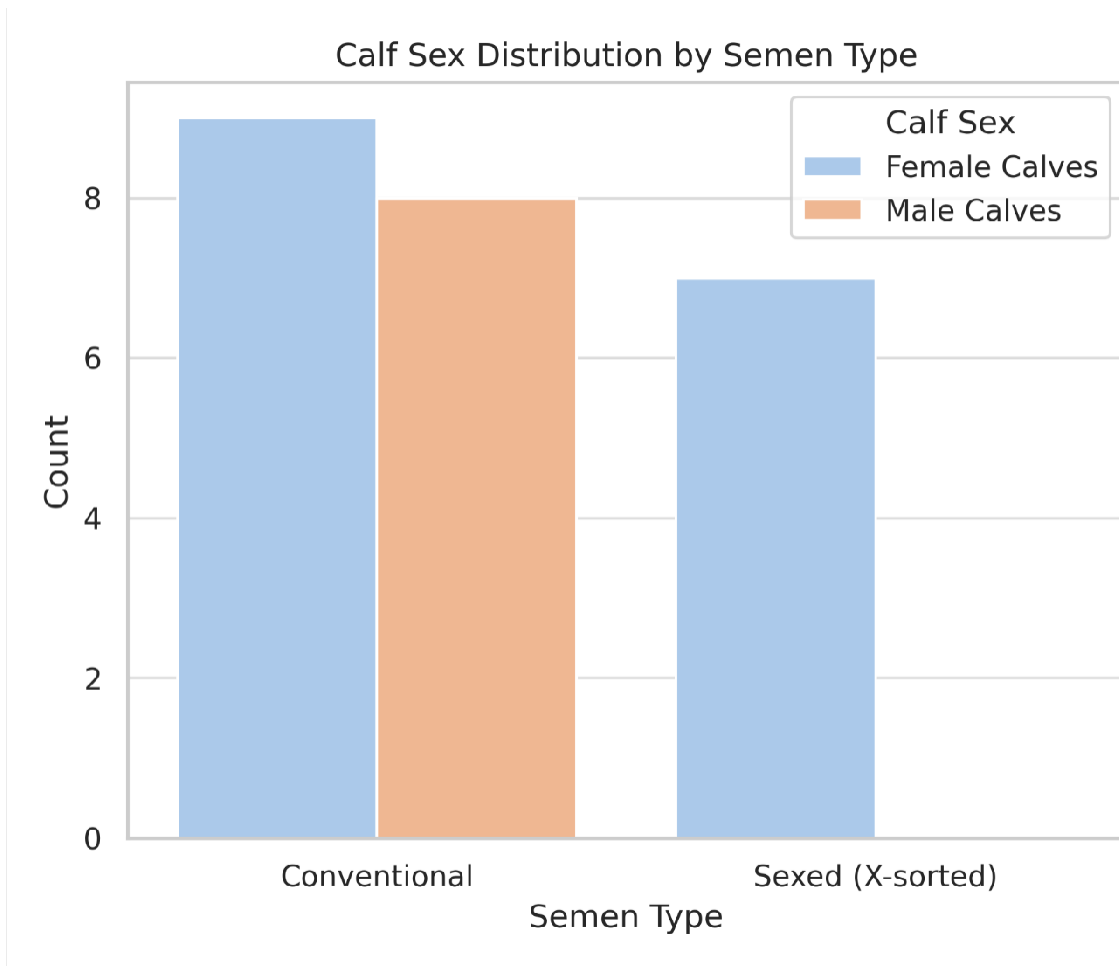
**Table 3. Repeat breeding rates and parity effects on conception with sexed semen**

Parameter	Conventional Semen (%)	Sexed Semen (%)
Repeat breeding rate	43.33	76.67
Conception rate - Heifers	-	30.00
Conception rate - Cows	-	20.00

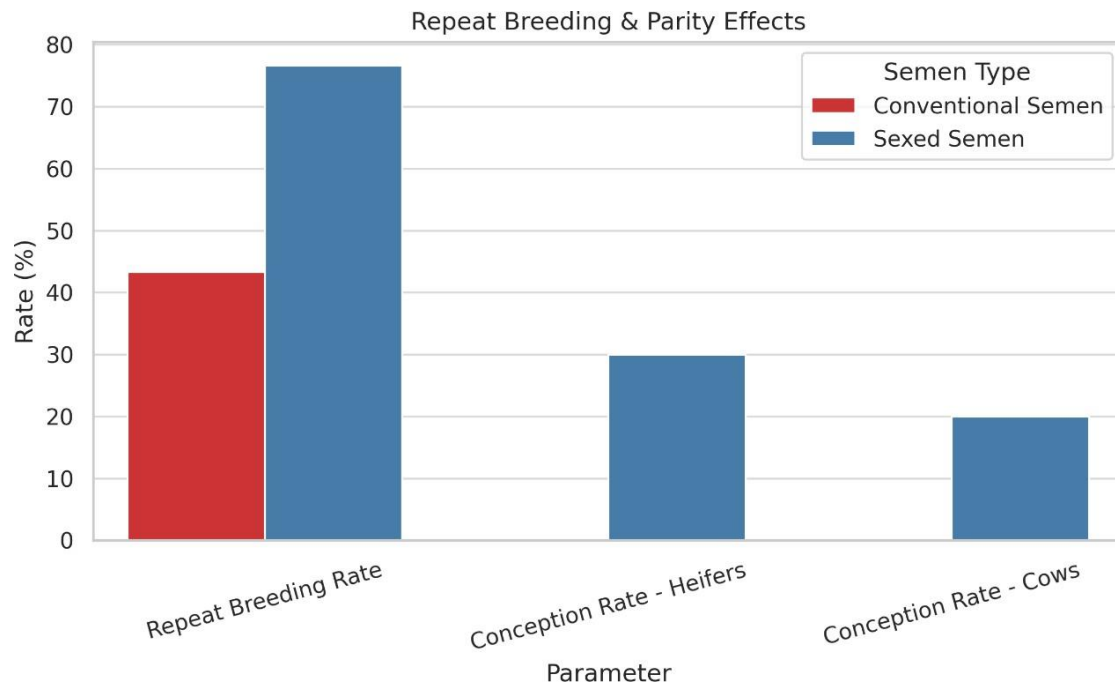
Table 3. Sexed semen required more repeat services (76.67%) and showed better conception in heifers (30%) than cows (20%).



*Figure 1 Conception rate by Semen Type*



*Figure 2 Calf Sex Distribution by Semen Type*



**Figure 3 Repeat Breeding & Parity Effects**

## Discussion

The current findings align with previous research documenting reduced conception rates associated with sexed semen in lactating dairy cattle [11]. The 23.33% conception rate achieved with sexed semen in Achai cattle closely matches reports from other breeds under similar management conditions [12], suggesting that the fertility challenges of sexed semen application may not be breed-specific. The markedly higher conception rate with conventional semen (56.67%) underscores the significant fertility trade-off inherent in current sexed semen technology [13].

Several biological and technical factors likely contribute to the observed differences in conception rates [14]. The flow cytometric sorting process subjects sperm to considerable physical stresses that may impair cellular function and membrane integrity [15]. The reduced sperm numbers per straw (approximately one-tenth of conventional doses) substantially decreases the probability of successful fertilization [16]. Additionally, emerging evidence suggests that optimal insemination timing may differ for sexed semen due to altered sperm longevity in the female reproductive tract [17]. These factors may be particularly consequential for lactating cows, which already experience metabolic challenges that can compromise fertility [18].

The perfect female: male ratio achieved with sexed semen confirms the technology's remarkable precision for sex selection [19]. This capability holds particular value for Achai cattle genetic improvement programs, potentially enabling rapid expansion of productive female populations while maintaining desirable adaptive traits through selective breeding with Jersey genetics [20]. However, the economic viability of this approach requires careful consideration, as the substantially higher cost of sexed semen combined with lower conception rates may outweigh the benefits of female calf production in many commercial scenarios [21].



## Conclusion

This study demonstrates that while sexed Jersey semen can effectively produce female calves in Achai cattle, its use results in significantly lower conception rates compared to conventional semen under field conditions. The 23.33% conception rate achieved with sexed semen versus 56.67% with conventional semen suggests that careful economic analysis is warranted before widespread adoption in smallholder dairy systems. The technology appears most promising for virgin heifers and genetically superior cows within structured breeding programs, where the value of female offspring may justify the additional costs and reduced fertility. Future research should focus on optimizing insemination protocols and developing breed-specific adaptations to enhance the effectiveness of sexed semen technology in indigenous cattle populations.

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