Why Indian dairy farmers must adopt Sex-Sorted Semen Breeding

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The sex pre-selection of the offspring using flow cytometry was proven possible in rabbits in 1989. Since this breakthrough, the technology has been slowly adapted to produce gender-selected offspring in several species including cattle, horses, pigs, goats and sheep. The benefits of sex-sorted semen are considerable. The exceptional qualities of SexedULTRA sex-sorted semen have resulted in field conception arates equal to or better than those of conventional semen under similar conditions, and a female gender ratio of calves born of over 93 per cent.

It has been calculated that by 2050 global milk production will increase to 1,075 × 106 tonnes (liquid milk equivalent; 62 per cent increase relative to 2005) (FAO, 2017). This increase will require marked improvements in production efficiency, as arable land resources are not increasing. To meet the world's increasing demand for milk production, it is necessary to substantially increase the number of elite females, which can best be achieved by shifting the sex ratio in a straw of semen. In addition, producing only female calves has proven to help farmers save resources that would have been shared with unwanted males.

Studies in cattle have demonstrated that the use of sex-sorted semen facilitates faster, more profitable herd expansion in seasonal, pasture-based dairy herds. The use of sex-sorted semen also offers benefits in terms of improved biosecurity since, if all replacements can be generated on-farm, the need to buy animals from external sources is eliminated. Similarly, it has been demonstrated that, by increasing the proportion of heifer calves born at the expense of heavier male calves, the incidence of dystocia is reduced in 1st-calving heifers by approximately 20 per cent, reducing losses in production, fertility, cow and calf mortality, increased culling and veterinary and management costs. It has also been suggested that gestation of a female calf could result in increased milk production, especially if the daughter pregnancy occurs at the first parity. Another important reason for incorporating sex-sorted semen in any dairy system in India is to reduce animal welfare issues due to abandoning of unwanted males, since many states have now banned slaughtering of cattle.

In the past ten years, STgenetics has made a significant investment in Research and Development, which has resulted in an overall improvement of the methods and technology

used for sperm sex- sorting. The current method, with the trade name SexedULTRA, was developed as a system to remove the main sperm stressors and retain cells in a benign environment throughout the sex-sorting process. This method accommodates changes in pH, temperature, and tonicity, and includes modified extenders that take into account the low dose required for sex-sorted semen. The method also removes dead, dying and damaged sperm cells, which helps increase the fertility of the sex-sorted semen, even at a low sperm numbers.

The improvements to the methodology of sperm sorting were initially reflected in laboratory evaluations, where semen processed using the SexedULTRA method presented improved in vitro sperm characteristics such as motility, acrosome integrity and freezable embryos produced compared with the previous method. Further publications revealed that SexedULTRA sperm visual and CASA sperm motility, viability, acrosome integrity and DNA fragmentation are now equal or better than those of conventional semen.

Large scale field trials in collaboration with Select Sires Inc. in the United States demonstrated that the in vitro improvements observed in semen processed following the SexedULTRA method were also reflected in a significant increase in conception rates compared to the previous method (10.8 per cent relative improvement). Further field trials in collaboration with German Genetics International GmbH confirmed the fertility improvement, where semen processed following the SexedULTRA method and inseminated at 4.0 million per dose present comparable fertility to that of conventional semen (102 per cent relative heifer fertility compared to conventional). These initial field trials confirmed that the deleterious effects of the previous methods of sperm sorting had been greatly alleviated, and that fertility rates are now equivalent to those of conventional semen.

In a meta-analysis on the long-term usage of sex-sorted semen, conception rates were estimated at STgenetics partnering herds for conventional and sex-sorted semen in Holstein and Jersey, and in heifers and cows, between the years 2012 and 2017. According to this study, conception rates for sex-sorted semen presented a progressive improvement starting in 2013, after the introduction of SexedULTRA. A similar study was performed by the United States Department of Agriculture, which confirmed the improvement in overall fertility of sex-sorted semen soon after the introduction of the SexedULTRA method in 2013. In the same period, sex-sorted semen utilization increased from 9.4 per cent to 30.7 per cent in heifers and from 0.2 per cent to 1 per cent in cows.

In addition to the sperm biology and fertility developments, the introduction of application specific sperm sorters has improved sorting performance and efficiency. The original MoFlo flow cytometers were expensive, bulky, had low throughput, and required high technical expertise to operate. The modern Genesis Digital multi-headed sperm sorters have advanced electronics, integrated fluidics and automated features that provide sorted subpopulations of X- or Y-sperm at rates of 8,000 sperm/second with over 90 per cent gender purity.

Another recent technology development is the

Figure 1

Report date	Calves	Female
August 2, 2019	221,500	91%
September 10, 2018	223,000	91%
August 3, 2017	247,000	91%
August 11, 2016	217,000	91%
May 4, 2015	155,000	90%
TOTAL	**1,063,500**	**91%**

Figure 1. Female calf percentages in the field after insemination with sex-sorted semen in France between the years 2015 and 2019.

Figure 2

ST INDIA: QC & DELIVERY REPORT SUMMARY - COMPILED DATA FROM 3 LABS

SN	BREED	# of Batch	# of straw produced	Avg # of Straws Produced/Batch	Avg Conc (Million sperm/ straw)	Avg 0hr Progressive Motility (%)	Avg 3hr Progressive Motility (%)	Avg Acrosome Integrity %(3Hr)	Avg purity (%)	Avg bacterial Count (CFU)
1	GIR	305	84,270	411.1	2.2	60.1	51.5	74.6	93.5	0
2	HF	594	263,886	540.1	2.2	65.7	58.1	78.0	93.8	0
3	HF CROSSBRED	143	62,994	440.1	2.2	59.9	53.2	75.6	93.8	0
4	JERSEY	198	101,560	536.2	2.2	61.9	54.9	75.1	93.5	0
5	JERSEY CROSSBRED	91	45,939	504.9	2.2	62.9	55.0	76.0	93.7	0
6	RED SINDHI	272	135,456	498.0	2.2	60.9	54.0	75.2	93.2	0
7	SAHIWAL	460	232,954	506.3	2.2	62.0	54.0	74.9	93.8	0
8	MURRH A BUFFALO	666	322,169	380.0	2.2	59.8	52.5	74.8	94.0	0
9	THARPARKAR	46	16,241	353.1	2.2	61.6	54.3	76.2	93.0	0
10	JAFFRABADI BUFFALO	23	6,175	268.5	2.2	54.3	50.9	75.7	93.8	0
11	NATHI	5	1,444	288.8	2.2	55.2	49.0	76.2	92.0	0
12	KHILLAR	5	2,194	438.8	2.2	67.0	58.5	74.6	93.8	0
13	GAILAO	2	182	91.0	2.3	79.5	75.0	82.5	92.0	0
14	MEHSANA BUFFALO	1	366	366.0	2.2	52.0	52.0	73.0	93.0	0
15	PANDHARPURI BUFFALO	2	523	261.5	2.2	56.0	52.5	75.5	93.5	0
16	DEONI	2	980	490.0	2.2	73.5	61.0	74.5	93.0	0
17	BADRI	12	1,705	169.6	2.3	64.0	58.1	75.4	94.5	0
18	HARIANA	1	629	629.0	2.2	63.0	56.0	68.0	95.8	0
	Total/Avg	2,894	1,298,389	450	2.2	62	56	76	93.5	0

Figure 2. Semen quality and delivery report compiled from BAIF, ULDB and MPSLDC sex-sorting laboratories.

use of Genesis I sperm analyzers to assess real time sperm gender purities. The implementation of these technological advancements provides high confidence in the quality assessment of the sex-sorted semen, which reflects in field results collected in the years of 2016-2018 confirming that an in vitro analyzed X-chromosome bearing sperm purity of 91.1±0.18 per cent resulted in an in vivo female calf percentage of 90.3±0.18 per cent. Similar to these results, the Institut de L'Elevage Idele, an organization appointed by the French ministry of agriculture as technical center for agriculture, reported that a 90 per cent purity product resulted in a 91.0 per cent female calf ratio in the field from 1,063,500 calvings between 2015 and 2019 (Figure 1).

STgenetics established its subsidiary in India, Inguran Sorting Technologies LLP (ST India), in 2018 and registered several patents. ST India has established three state-of-the-art sex-sorting laboratories; the first laboratory located in Pune Maharashtra in association with BAIF, the second one at Rishikesh Uttarakhand in association with ULDB, and the third laboratory at Bhopal in association with Madhya Pradesh Livestock and Poultry Development Corporation. In these laboratories, ST India has already produced about 1.3 million doses of sex-sorted semen from eighteen different breeds of cattle and buffaloes (Figure 2).

The average in vitro female purity of SexedULTRA sex-sorted semen produced in India is 93.5 per cent, with other quality control parameters also exceeding international and Indian standards. The average post-thaw progressive motility at 0 and 3 h is 60 per cent and 53 per cent, compared to the standard requirement of 50 per cent and 30 per cent, respectively. The average acrosome integrity is 75 per cent at 3 h, whereas the standard requirement is established at 50 per cent. These exceptional qualities of SexedULTRA sex-sorted semen have resulted in field conception rates equal to or better than those of conventional semen under similar conditions, and a female gender ratio of calves born of over 93 per cent.

The advantages of sex-sorted semen over conventional are numerous and varied. A high fertility and gender purity sex-sorted semen product has the potential to accelerate herd expansion, minimise waste production, improve animal welfare, and increase profitability compared with non-sorted conventional semen. While producing over 90 per cent females, the number of unproductive male calves will also be reduced to less than 10 per cent. The reduction of male calves will have a significant positive impact on the environment and socio-cultural factors. It will also leave more feed available for productive animals enhancing productivity and economic growth.