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A TRIAL INTO SEX FIXING OF PROGENIES IN DAIRY ANIMALS AND CALCULATING THE DEVELOPMENTAL IMPACT FACTOR OF SUCH A RESEARCH IN INDIAN CONTEXT

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ABSTRACT

Sex fixing of mammalian progenies has been an adorable dream of reproductive physiologists and scientific history is replete with attempts to produce sexed offsprings. Most of them involved techniques producing sexed semen. However, present methodology is a novel in vivo patented technology which uses the administration of a liquid oral formulation which contains chemical ingredients like monosodiumethanoate and ethanoic acid which when given before insemination to females of dairy animals; yields female off-springs with considerable success rate. The following exercise is a sum of two such attempts undertaken in the state of Rajasthan in India and various outcomes are described therein.

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INTRODUCTION

Mammals are the most dominant family of animals on earth today. So, there are great social and economic benefits of the production of sexed offsprings in them. Many researches have worked on this since the first half of the last century. Firstly, there were attempts to alter the -pH of the female genital tract. Secondly, there were efforts to produce sexed semen (sexed semen is the one containing accentuated proportions of either X or Y bearing spermatozoa) and the third were the application of certain chemicals, hormones, sera etc to produce a shift in the sex ratio. The present research also known as Aulprofem technique (Kebede et al. 2013), is an in vivo attempt to produce female offsprings where sex ratios are altered probably by binding of receptor sites for Y sperm on the ovum by the interaction of certain moieties that are generated in the body system by the action of the active constituents in the oral medication which is given to the female animals prior to insemination.

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Department of Applied Pharmacology, Gregor Mendel Institute for Research in Genetics, No. 144/2, Netaji Park, Baloke Road, Haibowal Kalan, Ludhiana, India This leaves relatively more chances for binding of X sperm with the ovum as receptor sites for it are still unbound. Among various outcomes of this discovery may be a rapid increase in the numbers of females of high yielding milch varieties of cows and buffaloes. This will definitely be taking the profession of dairying to newer heights by increasing milk productions alongside benefiting dairy industry too. The developmental impact factor of such an advancement is simply tremendous especially in developing countries where due to the presence of small holdings of land, almost every farmer has to adopt dairying as the necessary co-profession.

MATERIALS AND METHODS

The present exercise was a group of two different trials undertaken by the Department of Animal Reproduction, Gynaecology & Obstetrics, College of Veterinary & Animal Sciences, Bikaner under the technical co-ordination with Gregor Mendel Institute for Research in Genetics, Ludhiana under the supervision of B.S. Aulakh, the founder and present director as well as discoverer and patent holder of this technology (Aulakh 2008).

Table 1. The results on cows and buffaloes	subjected to	manipulation of sex ratios
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Sr. No	No. of Inseminations	No. of pregnancies	Male Offsprings	Female Offsprings
Trial no. 1	40	22	02	25
Trial no. 2	42	30	05	25
Total	82	52	7	45

 Table 2. Showing the actual increase in number of female calves per a hundred of male calves produced as a result of the use of sex fixer drug

	Imaginary unitary sex	Manipulated sex ratio at	Percentage increase in	Manipulated sex ratio
Sr. No.	ratio at birth per 100	birth per 100 calves	sex ratio per 100 calves	as per number of male:
	calves born, before the	born, after the trial;	born that appears on	female calves born, per
	trial; males: females	males: females	percentage scale, after	every 100 male calves
			the trial	born; after the trial
1.	50:50	40:60	10%	100:150
2,	50:50	30:70	20%	100:233
3.	50:50	20:80	30%	100:400
4.	50:50	13.5:86.5	36.5%	100:770

Table 3. Showing the increased national benefit to farmers due to the use of sex fixer drug

Sr. No.	No. of animals taken	Immediate national benefit	Cost of the sex fixer drug	Further national benefit to	Annual national revenue to
	for application of sex	to the farmers as the value	valued at a unit price of	the farmers as the value	the farmers from the value
	fixer drug used in	of additional female calves	approx. rupees 500; in	of fully grown animals in	of milk produced in Indian
	present study	in Indian rupees in billions	Indian rupees in billions	Indian rupees in billions	rupees in billions
1.	82	0.000285	0.000041	0.00096	0.0024
2.	100,000	0.34756	0.05	1.1707	2.9268
3.	10,000,000	34.756	5	117.07	292.68
4.	100, 000, 000	347.56	50	1170.7	2926.8
4.	100, 000, 000	347.56	50	1170.7	2926.8

It is in the form of a liquid oral dosage form which is administered to the female animals; cows and buffaloes, just before mating or AI (artificial insemination). It was procured from Kabir Remedies Pvt Ltd, a private company; as conveniently packed in unit doses of 225 ml each in PET bottles, labeled as Aulprofem and with instructions to be stored in moderate temperature conditions of 8°C to 25°C in a cool and dry place. The chemical constituents of this drug formula consisted of as having 0.6 gm of monosodiumethanoate dissolved in 10% v/v ethanoic acid q.s. to make required solution to 10 ml of such a preparation. No animals were harmed or harassed in any way during the trial. The active constituents of this drug formula are classified in all major pharmacopoeias of the world namely US, British, Australian, Indian etc as pharmaceutical aids which are naturally the safest group of pharma chemicals; so there arises no question of harassing or harming the animals under trial. The data was recorded as based in actual field conditions and various factors such as abortions, stillbirths, progress of pregnancy etc were regularly and routinely checked and sex of the off-springs were noted on actual deliveries. The inferences were drawn on simple percentage base and various outcomes and impact factors calculated in simple methodology.

RESULTS

The trial was conducted in two tiers; one included the application of drug formula on 40 animals and the other included 42 animals, cows and buffaloes both. Overall, the study covered 82 animals. There were 52 confirmed conceptions and luckily all animals successfully delivered viable offsprings as depicted in Table no. 1. In the commulative outcome, we have 7 male calves whereas the number of female ones is 45

DISCUSSION

On simple calculation, we have with us 86.5% female calves. This means that male calves are 13.5%.

This seems to convey that an imaginary unitary sex ratio at birth of 50:50; males to females is manipulated in such a way that it has moved up on the percentage scale about 36.5 points. This means that sex ratio is manipulated a mere 36.5% from the original imaginary sex ratio of 50% females to the now manipulated sex ratio of 86.5%. But actually this is not so because now the original male sex ratio at 50% has also moved down to a value of 13.5%. So, now we have 86.5 females at birth for every 13.5 males born. In the present calculation of 45 female births for 7 male births, this turns out to be 770 female calves at birth for every 100 male calves born. In another words, now the manipulated sex ratio is 770%. These results can be better understood with the help of Table no. 2 where it is well elaborated by taking the examples of sex ratio manipulation on female side from 60% to 80% on the simple percentage scale. One more fact should be taken into consideration that secondary sex ratios are seldom unitary. In most of the cases in dairy animals, they are male dominated i.e. there are more male births than female ones.

So, if a sex ratio in a sex fixer exercise is manipulated on the female side, this gives more value and performance weightage to such a technique. In the present exercise of sex fixing, we have 7 male calves whereas the number of female calves is 45. This means an excess of 38 female calves at the end of the sex fixing exercise. Had there been no such exercise being undertaken, we could have gotten 26 female as well as male calves both. Now we have 45 female calves. This means that we have 19 more female calves produced as the visible boon of the sex fixer drug. Calculating from an Indian context because this exercise was undertaken in India, we can safely conclude that the given technology of this sex fixer drug discovery has already gifted the farmers of Rajasthan province, an additional 19 female calves. Since such a valuable technology is usually tried in animals of superior and valuable exotic breeds, so it derives that we have now additional 19 valuable calves of better exotic breed stuff. If each of these calves is fixed a conservative price value of Indian rupees

15,000; this means that farmers of Rajasthan who benefited from this trial have already pocketed a sum of nearly rupees 2, 85, 000. So, even if such a technology is priced at around a unit price of rupees 500 each meaning an input cost of nearly rupees 41, 000 because 84 units were used in the entire exercise; this has generated an additional national profit of almost rupees 2, 44, 000 in the first step only. One more aspect of this research is that these additional 19 calves that have emerged from a beforehand figure of nowhere are now a valuable national asset. If about 20% of them i.e. 4 of them die of calf mortality and more 20% of the remaining ones i.e. about 3 of them succumb to death before reaching maturity thus leaving us with a figure of 12 fully grown and mature, additional superior breed cows and buffaloes ready to deliver. If each one of them is priced at a mere rupees 80, 000; this means that Indian farmers have further pocketed rupees, 9, 60, 000. Further if each one of these of arguably exotic breed cows and buffaloes yields a conservative milk yield of 4000 liters per lactation cycle. This means that at a price of rupees 50 per liter of milk this becomes an output per animal of nearly rupees 2,00, 000 per a lactation cycle. Again this means an additional national revenue for such 12 animals at nearly rupees 2, 400, 000. This is a huge figure of nearly 2.4 million rupees. Suppose this cycle of lactations goes on year after year for many years, this means an annual contribution of equal amount per year if the lactation cycle is of about one year each. Even if it is a little more or less, it does not make much a difference as rupees 2.4 millions is a very, very huge amount as compared to the input cost just explained above of the sex fixer drug standing at about rupees 41, 000. The corresponding figures of the cost of drug if such a trial sample is extended to animals; hundred thousand, ten millions and hundred millions in numbers, stands at values of 50 millions, five billions and fifty billions in Indian rupees but the outcome increase in national annual income of farmers comes out to be at exorbitant figures of 2.926 billions, 292.6 billions and 2926 billions as depicted in table no. 3. Further, such benefits as the immediate benefit as the price of the female calves born and the value of fully grown animals for same numbers of animals taken for trial are also given in the same table. It may also be added that India has an animal wealth of nearly 300 million cows and buffaloes of which nearly 160 millions are of breedable category. The value of Indian rupees at present is at about 65 rupees per a US dollar. To correlate this calculation with respect to economies of other nations, this exchange rate value of Indian rupee should be kept in mind.

Sex fixing in mammals has a long history. Many scientists have worked on this (Beernik & Ericsson 1982, Lindahl 1956). The major drawbacks of these techniques remained the lack of laboratory tests to evaluate the degree of sperm separation (Hafez 1985) and the inability to know the precise mechanism of binding and fusion of mammalian sperm with ova. Overall, the manipulation of mammalian sex ratio has still remained a mirage on the horizon (Hunter 1982). The fusion of a sperm with ovum is the most magnificent event of the world but our knowledge on this important aspect of life is still limited. Recently there have been attempts to study the effects of certain compounds like tetraspanins like CD9 (Hemler, 2003), CD81 (Cormier at al., 2004), glycosylphosphatidylinositol anchored proteins like CD55 (Coonrad et al., 1999), integrins (He et al., 2003), disintegrins (Primakoff et al., 2000), fertilins, cyritestin (Cho et al., 1998) etc on the process of gamete fusion. These researches have clearly established the presence of receptor sites on oocyte and their respective ligands on the surface of sperms. But the bigger questions are far from being answered. The 'tetraspanin-web' is still a big mystery (Boucheix et al., 2001). The role of other candidate molecules the likes of which are mentioned above; needs a lot more investigating. Recently there have been attempts to differentially bind X and Y bearing spermatozoa using H-Y antigen antibody interactions using non protein substrata such as agarose beads and U.S. patents granted for them (Bryant 1984, Zavos & Dawson, 1991). These developments clearly demonstrate the existence of differential binding sites for X and Y sperms and this difference can be exploited in developing a viable technology for sex fixing in mammals, either in vivo or in vitro. The present technology is an in vivo technique and it can be hypothesized that the material of present research produces certain YSBLM (Y sperm binding ligand mimics) moieties in the living system and also in the genital system of the female animals undergoing this treatment in such a way that these YSBLM moieties differentially bind with candidate receptor sites on oocyte involving ZP and membrane binding and penetration to inhibit their binding ability and/or their fusion with candidate ligands on the Y bearing sperm. The importance of this technology becomes even more marked with the view that it will not only provide an insight into the differential behaving and working of X and Y bearing sperms in the process of fertilization but also provide valuable data and knowledge on the role played by the candidate molecules in understanding fully well the exact mechanism of gamete fusion.

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