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Nutritional Infertility and Ameliorative Measures in Dairy Animals of Middle Gujarat

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Abstract

Fertility improvement camps were organized in different villages of middle Gujarat covering 1481 breedable cattle and buffaloes with the history of impaired fertility. The reproductive status of animals was ascertained through gynaeco-clinical examination per rectum. Blood sampling (n=305) was done from representative groups of animals to know the nutritional status. All non-infectious infertile animals were dewormed using either Inj. Ivermectin 100 mg s/c or bolus Fenbendazole + Ivermectin 3.0 g orally. Anestrus animals (n=265) were then nutritionally supplemented either with chelated area specific mineral mixture (ASMM, 50-60 g/d/h) or multi-minerals boli - one on alternate day, with and without bypass fat (75-80 g/d/h) for one month along with an advice to improve feeding level, estrus detection and timely breeding. Revisits were made after 2.5-3.0 months to check the reproductive status. The mean blood glucose concentrations were significantly lower ($p<0.05$), while total cholesterol and zinc levels were higher ($p<0.05$) in anestrus than in subestrus and/or repeat breeding cows and buffaloes. Moreover, the plasma total cholesterol levels in buffaloes were just one half to those of cows. Further, the plasma total protein, calcium, phosphorus, magnesium, iron, copper and cobalt concentrations were found to be non-significantly higher in repeat breeders than in anestrus or subestrus cows and buffaloes. Calcium was deficient in some 20% animals and calcium: phosphorus ratio varied from 1:1 to 1.8:1 in animals under different areas of study indicating its role in causing infertility in dairy animals. Supplementation of ASMM, multi-mineral boli alone or in combination with bypass fat in pubertal /postpartum anestrus animals resulted in 34-56% estrus induction response and 33-50% conception rate, both being better with mineral plus bypass fat supplementation than minerals alone.

Introduction

Malnutrition is one of the major causes of non-infectious infertility in dairy animals in India. It is due to energy, protein and mineral deficiencies mainly of calcium, phosphorus, trace minerals (copper, cobalt, zinc, iodine and manganese) and other salts (Boland and Lonergan, 2003). Phosphorus deficiency leads to irregular estrus, anestrus, decreased ovarian activity and lower conception rate. The dietary manganese deficiency was shown to affect epiphyseal growth during embryogenesis leading to birth of calves with congenital skeletal defects

(Carvalho *et al*., 2010). The present study was carried out to investigate the incidence of reproductive disorders in animals presented in animal health camps and to evaluate the effect of minerals and bypass fat supplementation as an ameliorative measure in non-infectious infertile animals.

Materials and Methods

The study was conducted on 1481 breedable cattle and buffaloes (pubertal and postpartum) with the history of impaired fertility in sexual health control cum fertility improvement camps organized in central part of Gujarat state. These animals were examined gynaeco-clinically to ascertain their real reproductive status, and observations were recorded. All infertile animals identified were dewormed either using Inj. Ivermectin 80-100 mg s/c or bolus Fenbandazole 3 g + Ivermectin 100 mg (Fendikind plus 3.0 g, Vet Mankind) and pour on liquid (20 ml) for control of ectoparasites, if any. Blood samples (n=305) were collected from representative animals of groups of non-infectious infertility under each region to ascertain profile of blood glucose, plasma total protein, total cholesterol and macro-minerals using standard procedures and assay kits on biochemistry analyser. The levels of trace minerals were determined using triple acid digested plasma samples on atomic absorption spectrophotometer.

Depending upon the body condition score (0-5 scale) and reproductive status, 265 anestrus cattle and buffaloes (BCS 2.5-3.0) were then nutritionally supplemented with either chelated area specific mineral mixture (ASMM of AAU, Anand, 50-60 g/d/h) or multi-minerals boli (Garbhamin/ Minotas) - one on alternate day, with and without bypass fat (Sunergy, Polchem Industry, 75-80 g/d/h) for one month along with an advice to improve feeding level, estrus detection and timely insemination, if the animals exhibited estrus. Re-visits were made after 2.5 to 3.0 months to check the reproductive status. The estrus response and inseminations as reported by owners, and the ovarian status and conception, if any, were judged by rectal palpation. The clinical data were analysed to work out the frequency of various reproductive disorders in the animals presented. The data on blood profile were analysed on SPSS software to calculate group-wise Mean ± SEs and ANOVA, and the mean differences for groups were compared using NMRT/post-hoc test (Snedecor and Cochran, 1994).

Results and Discussion

Among 1481 so called infertile animals examined, the incidence of anestrus, subestrus, repeat breeding, uterine infection and infantile genitalia was 25.88, 16.49, 16.08, 11.82 and 11.08 %, respectively. Moreover 1.42 % animals were in estrus, 7.57 % pregnant and 9.66 % had miscellaneous medical disorders. This trend and frequency of various reproductive disorder noted concurred well with the previous reports from the same institute (Butani *et al.,* 2008; Hadiya *et al.,* 2013; Dhami *et al*., 2017, 2018). The findings clearly revealed that anestrus, followed by repeat breeding and genital infections are the major causes of infertility in dairy animals under field condition. Among heifers of more than 3 years age, infantile genitalia and poor body condition due to malnutrition were the main causes of post-pubertal anestrum.

The blood plasma profile of 305 animals with non-infectious infertility (anestrus, subestrus, repeat breeding) showed that the blood glucose varied from 31 to 67 mg/dl among individual animals. The mean value was significantly ($p<0.05$) lower in heifers and anestrus animals than in subestrus and/or repeat breeding cattle and buffaloes (Table 1). The plasma protein values of individual animals ranged from 5.5 to 11.3 g/dl in different categories of animals. The mean value was non-significantly lower in anestrus than repeat breeding and subestrus cattle and buffaloes. Plasma total cholesterol concentration in buffaloes was just one half to that of cows of all three categories, and it was also found to be lower in repeat breeders than anestrus animals of both the species with significant difference in cattle (Table 1).

Plasma or serum proteins bear a close relationship with the tissue proteins, and are necessary for the development of endocrine glands including pituitary and sex organs. Protein deficiency retards the development of reproductive organs and was considered to be a factor responsible for failure or delay in onset of postpartum estrus (Roberts, 1986). Cholesterol

Table 1: Blood plasma biochemical and mineral profile in infertile cattle and buffaloes

Species	Repro-ductive Status	Plasma biochemical and mineral constituents									
		Blood glucose (mg/dl)	Total protein (g/dl)	Total chole-sterol (mg/dl)	Calcium (mg/dl)	Phosph orus (mg/dl)	Magnes ium (mg/dl)	Zinc (ppm)	Iron (ppm)	Copper (ppm)	Cobalt (mmn)
Cattle	Anestrus (n=75)	38.14^{a} ±3.91	7.87^{b} ±0.31	184.8 ±7.82	8.27 ±0.21	5.45 ±0.23	2.17 ±0.15	1.08^{b} ±0.02	2.12 ±0.04	1.63 ±0.04	0.33 ±0.01
	Repeat breeding (n=40)	48.27^{b} ±3.78	8.37^{a} ±0.24	163.5 ±6.89	9.29 ±0.18	5.92 ±0.38	2.38 ±0.18	0.87^{a} ±0.03	2.25 ±0.05	1.89 ±0.03	0.47 ±0.00
	Subestrus (n=40)	43.56^{ab} ±2.79	8.36^{b} ±0.27	188.9 ±7.47	8.78 ±0.17	5.19 ±0.37	2.07 ±0.16	0.98^{ab} ±0.01	2.07 ±0.05	1.76 ±0.05	0.42 ±0.01
Buffalo	Anestrus (n=85)	37.72^{a} ±3.04	7.49 ±0.31	97.36 ±5.40	9.34^{a} ±0.28	5.42 ±0.28	2.21 ±0.14	1.18^{b} ±0.04	1.97 ±0.03	1.55 ±0.04	0.34 ±0.00
	Repeat breeding (n=30)	45.59^{b} ±3.37	8.21 ±0.33	84.9 ±5.31	10.65^{b} ±0.34	6.27 ±0.25	2.39 ±0.09	0.94^{a} ±0.02	2.37 ±0.03	1.66 ±0.05	0.53 ±0.01
	Subestrus (n=35)	42.28^{ab} ±3.34	7.89 ±0.35	93.56 ±4.98	9.53^{a} ±0.29	5.59 ±0.29	1.99 ±0.11	0.88^{a} ±0.03	1.99 ±0.04	1.39 ±0.03	0.45 ±0.01

Means bearing uncommon superscripts differ significantly within the column for a species ($p<0.05$).

Table 2: Estrus response within 90 days of supplementation of ASMM, Mineral boli and Bypass fat in anestrus cattle and buffaloes

Supplements	Cattle			Buffalo		
	No. supplied	No. (%) responded	No. (%) conceived	No. supplied	No. (%) responded	No. (%) conceived
ASMM	45	18 (40.0)	7 (38.9)	67	24 (35.8)	8 (33.3)
Mineral boli	35	12 (34.3)	4 (33.3)	52	18 (34.6)	7 (38.9)
Min + bypass fat	30	17 (56.7)	8 (47.1)	35	18 (51.4)	9 (50.0)

is the most important sterol essential for life with its varied functions such as lipid transport system and steroidogenesis in the body (Mc Donald, 1980). Kumar *et al.* (2009) and Butani *et al.* (2011) reported significantly lower serum total protein and cholesterol in repeat breeders than in anestrus and subestrus cows and buffaloes. It was demonstrated that cows with higher milk and sub-optimal nutrient intake suffer greater body weight losses and reduced fertility (Santos *et al*., 2009). Feeding of unsaturated fatty acids increased fertilization and improved embryo quality (Cerri *et al*., 2009) due to alterations in oocyte competence in dairy cows and modulation of the uterine secretion of PGF2α (Staples *et al*., 1998). Feeding of supplemental fat to dairy cows improves fertility owing to availability of energy and specific fatty acids.

The plasma levels of calcium, phosphorus and magnesium were found to be non-significantly higher in repeat breeders than the anestrus or subestrus cattle and buffaloes. The calcium was found to be deficient in around 20% animals and calcium: phosphorus ratio varied from 1:1 to 1.8:1 in animals under different areas of study indicating its role in causing infertility in dairy animals. Among trace minerals, plasma zinc levels were lower, while iron, copper and cobalt levels were higher in both repeat breeder cattle

and buffaloes as compared to anestrus or subestrus ones, however, differences were statistically non-significant (Table 2). Kumar *et al.* (2009) and Butani *et al.* (2011) reported significantly ($p<0.01$) lower serum calcium in anestrus and repeat breeding crossbred cows and buffaloes than in subestrus and normal cyclic ones, but the trend of inorganic phosphorus was reversed. Ali *et al.* (2014) recorded significantly lower serum calcium in anestrus than repeat breeding cattle, but not in phosphorus levels. Anoestrus and repeat breeder buffaloes have been reported to respond favorably to daily supplementation of 30 g ionic or chelated mineral mixtures with 50-70% conception rates (Butani, 2013).

Supplementation of ASMM 50 g/h/d or multi-minerals boli (Garbhamin/Minotas) alone one on alternate day or in combination with bypass fat 70-80 g/h/d in pubertal/postpartum anestrus animals resulted in 34-56% estrus induction response and 33-50% conception rate, both being better with mineral plus bypass fat supplementation than minerals alone (Table 2). Among the non-estrus detected/ non-bred and non-conceived animals, majority (65-80%) were having functional ovarian structure when examined 2.5-3.0 months later following one month of mineral and/or bypass fat supplementation compared to non-supplemented group of 30 animals, wherein only three animals were reported in estrus and one only conceived. The rests were still in true anestrus condition. These results proved the nutritional cause of infertility and beneficial role of minerals and energy supplementation in improving reproductive performance in such animals. These results were in line with the previous observations under AICRP from the same station (Annonymus, 2018).

Nutritional deficiencies combined with worm infestation play a major role in infertility of crossbred heifers and cows (Akhtar *et al.,* 2004). Various workers have studied the effect of mineral supplementation on estrus induction and conception in dairy animals. The deficiency of phosphorus and/or improper calcium and phosphorus ratio cause anestrus condition in dairy animals. Phosphorus is essential for transfer of biological energy, particularly through ATP, and deficiency of it may arrest the phenomenon of fertilization resulting in the repeat breeding in animals.

In the present study, satisfactory estrus induction and conception rate were observed which may be due to combined effect of various macro-micro minerals and bypass fat which have positive effects on steroidogenesis, follicular growth and symptoms of ovulatory estrus (Singh *et al.,* 2011; Anonymous, 2018). Srivastav (2008) reported that supplementation of 30-40 g commercial mineral mixture daily in concentrate for 20 days induced ovulatory estrus in 93.93 % of anestrus crossbred heifers with first service conception rate of 32.14 %. Devasenat *et al.* (2010) recorded 59 to 62 % estrus response among anestrus heifers and cows and repeat breeder cows following mineral supplementation, while Puvarajan and Vijayarajan (2013) reported estrus induction in 92.16 % (153/166) of pubertal crossbred heifers over three months period following one month of ASMM supplementation, and the first service conception rate recorded by them was 28.18 % (42/149). Moreover the findings of present study supported the view of Kumar et al. (2003) who substantiated that nutrition during early lactation and advanced pregnancy is the most crucial part to maintain dam's body reserve and fetal growth, a deficient nutrition may result in postpartum infertility or abortion. The major setback noted in some pockets in our study was however that the farmers were resource poor in providing required nutritional inputs-feed stuff to their animals along with our supplements to cope up with the negative energy balance for initiation of ovarian activity, and hence the overall results were poor.

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Conflict of Interest:

The authors have no conflict of interest.

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