

Effect of Supplementing Certain Chelated Minerals and Vitamins to Overcome Infertility in Field Animals

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In Banaskantha district of Gujarat, 35 animals with normal reproductive organs, but suffering from anoestrus were selected. Nine anoestrus animals served as control, and remaining twenty-six were given daily 5 g supplement per animal, containing chelated Cu, Zn and Mn and fat-soluble vitamins A, D₃ and E. Minerals (Cu, Zn and Mn) and progesterone were analyzed in the blood serum before feeding of supplement and at bi-weekly intervals after feeding the supplement. Copper (0.92 ± 0.01 ppm) and zinc (1.02 ± 0.03 ppm) levels increased significantly ($p < 0.05$) in the blood serum of animals under experimental group, as compared to control Cu (0.50 ± 0.02 ppm) and Zn (0.60 ± 0.03 ppm). However, level of Mn in experimental (0.06 ± 0.01 ppm) and control (0.04 ± 0.01 ppm) groups was similar. Twenty-four animals in experimental group exhibited estrus, after on an average 27 days of feeding the supplement; however, only two animals exhibited estrus in control group during this period. In control group, progesterone level did not rise and remained low (0.25 - 3.81 ng/ml), but in experimental group the serum progesterone varied from 0.30 - 20 ng/ml. Animals that showed estrus were subsequently inseminated either through natural service or by artificial insemination. From the present study, the inference could be drawn that the supplementing of trace minerals, in the form of chelates, along with vitamins A, D₃ and E, can help in curing the problems of anoestrus/repeat breeding in dairy animals.

Keywords: Chelated minerals, vitamins, progesterone, blood serum, dairy animals, Banaskantha district

INTRODUCTION

Minerals and vitamins deficiencies may result in delayed onset of estrus, repeat breeding and/or infertility (Underwood and Suttle, 1999). Impaired reproduction performance results in an increased inter-calving period, causing great economic loss to the dairy farmers, which is often unrealized. Infertile dairy animal means a loss in milk production and profitability from dairying (Prasad and Gowda, 2005). Efforts should, therefore, be made to improve fertility in dairy animals by reducing inter-calving period and curing anoestrus animals through supplementation of minerals and vitamins (Garg *et al.* 2005). Minerals are supplied to the livestock through mineral mixture in the inorganic form. One of the major disadvantages of using such supplements is that the minerals from such sources are not fully absorbed due to antagonism and anti-nutritional factors present in the diet.

In view of the above, a bolus was formulated containing trace elements (Cu, Zn and Mn) required for reproductive functions, in the form

of organic chelates, along with vitamins A, D₃ and E.

MATERIALS AND METHODS

A primary survey was conducted in Banaskantha district of Gujarat, to identify anoestrus/repeat breeder animals. Forty-one animals comprising cow heifers (n=12), buffalo heifers (n=10), lactating cows (n=8), lactating buffaloes (n=10) and dry buffalo (n=1) suffering from problem of anoestrus were identified and subjected to gynecological examination. Out of 41, six animals were excluded from the study, as these animals were found to be suffering from anatomical disorders. Remaining 35 anoestrus animals having normal reproductive organs, but suffering from anoestrus of non-specific etiology were considered for the trial. Twenty-six animals were taken under the experimental group and supplemented daily with one bolus of 5 g per animal, containing chelated Cu, Zn, Mn and vitamins A, D₃ and E. The levels of supplemental Cu, Zn and Mn were according to level of deficiency in the total ration of animals, after considering total dietary mineral intake.

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Vitamins A, D₃ and E were supplemented to animals as per NRC (2001). Nine anoestrus animals having normal reproductive organs served as control.

The basal diet of animals under trial consisted of 4.0-5.0 kg *bajra* straw supplemented with 10-12 kg green hybrid napier grass and 1.0 kg cattle feed/concentrate mixture. Feed and fodder samples were collected for proximate analysis and minerals. Blood samples were collected from all the animals before the start of the trial and at bi-weekly intervals during the trial period. Serum was separated and kept in deep freeze till analyzed. Feed and fodder samples were analyzed for proximate composition only (AOAC, 1999), whereas, feeds and blood serum samples were tested for trace minerals, using Inductively Coupled Plasma-Optical Emission Spectrometer (ICP-OES), Perkin Elmer, Optima-3300 RL. Progesterone level in serum samples was measured by RIA technique (Kamboj and Prakash,

1993). The data on serum mineral profile and progesterone level were subjected to analysis of variance for statistical significance as per Snedecor and Cochran (1967).

RESULTS AND DISCUSSION

Mineral Content in Feeds and Fodder

The feeds and fodder samples were analyzed for chemical composition, Cu, Zn, Mn and values are given in Table 1. Feedstuffs commonly fed to the dairy animals were found to be below the critical levels for Cu and Zn. Garg *et al.* (1999) reported similar results for Northern agro-climatic zone of Gujarat in which Banaskantha district falls, and indicated that these minerals need to be supplemented in the ration of animals.

Effect of Feeding Bolus on Mineral Levels in Blood Serum of Animals

The mean bi-weekly blood serum Cu, Zn and Mn levels in animals are presented in Table 2.

Table 1: Chemical Composition and Cu, Zn and Mn Content in Feedstuffs Offered to Animals During Trial Period

Feedstuff	DM (%)	CP (%)	EE (%)	CF (%)	Cu (ppm)	Zn (ppm)	Mn (ppm)
<i>Bajra</i> straw	90.0	6.88	0.70	29.20	6.11	27.61	48.79
Wheat straw	92.0	4.02	0.90	29.66	6.10	21.10	27.10
Local grasses	28.0	7.10	1.26	29.70	5.70	32.48	45.28
Hybrid napier	17.3	7.80	1.62	27.30	6.77	26.09	51.66
<i>Jowar</i> green	35.6	6.54	1.60	24.80	8.71	34.60	48.81
Cottonseed cake	92.0	25.10	6.37	24.35	9.03	39.19	17.01
Compound cattle feed	91.0	19.85	2.96	9.84	23.98	33.27	73.21
Critical level*	—	—	—	—	< 8.0	<30.0	<40.0

*McDowell *et al.* (1993)

Table 2: Average Cu, Zn, Mn and Progesterone Levels in Blood Serum of Animals

Week	Cu (ppm)		Zn (ppm)		Mn (ppm)		Progesterone (ng/ml)	
	Control	Experimental	Control	Experimental	Control	Experimental	Control	Experimental
0	0.46 ± 0.02	0.50 ± 0.02	0.59 ± 0.04	0.64 ± 0.03	0.04 ± 0.01	0.04 ± 0.00	2.75 (0.32-3.54)	4.32 (0.15-17.1)
2	0.50 ± 0.04	0.76 ± 0.02	0.60 ± 0.03	0.76 ± 0.03	0.04 ± 0.00	0.04 ± 0.00	2.13 (0.36-5.61)	2.05 (0.0-9.85)
4	0.53 ± 0.03	0.78 ± 0.02	0.61 ± 0.03	0.88 ± 0.02	0.04 ± 0.01	0.05 ± 0.00	0.43 (0.22-0.87)	2.71 (0.26-13.88)
6	0.52 ± 0.02	0.86 ± 0.01	0.59 ± 0.03	0.95 ± 0.02	0.04 ± 0.01	0.05 ± 0.00	1.54 (0.12-2.10)	3.04 (0.16-15.52)
8	0.50 ± 0.02	0.92* ± 0.01	0.60 ± 0.03	1.02* ± 0.03	0.04 ± 0.04	0.06 ± 0.01	1.36 (0.25-3.81)	5.27 (0.30-20.21)

n= 9 in control, n=26 in experimental. *p<0.05 Figures in the parentheses indicate range.

All the animals suffering from problems of anoestrus and repeat breeding were found to contain Cu and Zn in blood serum below the critical limits of 0.65 and 0.8 ppm, respectively. Chauhan *et al.* (1992) reported low Cu and Zn values in animals suffering from anoestrus and repeat breeding. On supplementation of bolus, the effect was more pronounced for Cu and Zn levels in blood serum, whereas, in case of manganese (Mn) appreciable change was not noticed. This could probably be due to high turn over of Mn in blood. There was a significant increase in the concentration of Cu (0.92 ± 0.01 ppm) and Zn (1.02 ± 0.03 ppm) in blood serum of experimental group, crucial mineral elements for reproduction (Campbell *et al.* 1999). Gowda *et al.* (2004) reported significant ($p < 0.05$) increase in blood plasma Cu and Zn in mineral supplemented group. In the present study, a clear and positive relationship was found between Cu and Zn levels in blood serum and postpartum fertility. Copper deficiency can affect insulin release and glucose tolerance (Choudhary *et al.* 1981; Hassel *et al.* 1983), which in turn affects progesterone hormone production adversely. Reduced insulin concentration leads to delay in estrus cyclicity and impaired oocyte quality and corpus luteum function (Robinson *et al.* 2006).

Optimum level of Zn is essential to maintain the activity of FSH and LH, thereby, facilitating normal reproductive performance. Moreover, prostaglandins bind Zn and facilitate its transport. A reduction in Zn level probably interfere with prostaglandins receptor mediated phase and consequently luteolytic process (Spears, 1996). The overall mean Mn levels in experimental and control group were identical (Table 2), which could be due to high turn over of Mn in blood serum. Present findings with respect to trend in serum Mn level are in agreement with the observations of Shah (1999).

Effect of Feeding Bolus on Concentration of Progesterone in Blood Serum

Average initial progesterone level in blood serum of anoestrus and repeat breeder animals ranged from 0.15 to 17.10 ng/ml. Subsequently, progesterone analysis in blood serum revealed that supplementation of bolus brought about normalization of progesterone secretion pattern. Level of progesterone was higher (0.30-20.21 ng/ml) in experimental group. However, such trend

was absent in animals, which did not exhibit estrus and their progesterone level in blood serum remained at low level (0.25-3.81 ng/ml). These levels are in agreement with the findings of Shah *et al.* (2006). Recent studies have shown that diets, which induce ovarian function that leads to normal progesterone production by the corpus luteum, improves fertility in dairy animals (Robinson *et al.* 2006).

The assay of progesterone level in the serum samples is a reliable test for early pregnancy detection. In present study, animals that did not report a sharp decline in progesterone level post insemination were found to be pregnant later on. The average serum progesterone level in pregnant animals was maintained at over 6 ng/ml.

Effect of Feeding Bolus on Curing the Problem of Anoestrus in Field Animals

Out of 26 animals in experimental group, 24 animals exhibited estrus at an average feeding rate of 4 weeks. Two animals did not exhibit estrus. The problem of anoestrus in those two animals could be due to factors other than micronutrient deficiency. Only two animals in control group exhibited estrus during this period. It is important to mention that despite poor feeding and management practices, animals responded to bolus in an average time of 4 weeks, which shows that the animals suffer from anoestrus primarily due to minerals and vitamins deficiency. Manspeaker *et al.* (1987) reported that Cu, Zn and Mn (chelated forms) supplementation to heifers reduced the percentage of infections, embryonic mortality and improved postpartum involution and tone of the pregnant horn. All the animals in the present study that exhibited estrus were bred either by natural service or by artificial insemination. Out of 24 animals that exhibited estrus, 9 animals conceived. It is quite evident from the present study that supplementing specific trace minerals in the form of chelates, along with fat-soluble vitamins A, D₃ and E, can help curing the problems of anoestrus/repeat breeding in dairy animals under field conditions.

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