RESEARCH ARTICLE

# Amelioration of Postpartum Fertility with Energy, Chelated Minerals and Vitamins Supplements in Dangi Cows

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## ABSTRACT

The study comprised 24 recently calved Dangi cows of the university farm with a body condition score of 3.0 ± 0.5, and parity 2-5, which were randomly divided into four groups. Group I (n=6) was the untreated control fed as per routine farm feeding schedule, while groups II, III and IV (n=6 each) were fed an additional 30 gm chelated minerals and vitamins supplement, 100 gm energy supplement (Bypass fat) and both 100 gm energy with 30 gm chelated minerals-vitamins, respectively, daily for 50 days postpartum. Supplementation of energy, chelated minerals, and vitamins had significant impact on serum biochemical parameters like glucose, urea, albumin, globulin and Mg levels, except Ca, P levels. All the cows in group IV showed postpartum ovarian activity earlier than in other groups. The first postpartum estrus (PPE) was observed in 6/6, 5/6, 4/6, and 3/6 cows in groups IV, III, II, and I, with an average day of 68.50±0.60, 72.00±0.49, 71.25±0.49 and 88.67±0.55, respectively, the differences were statistically non-significant. The first service conception rate was highest in group IV (66.66%) followed by group II (50%), group III (40%), and group I (33.33%). The number of days open were 91.00±00, 75.5±0.31, 85.00±0.63, and 70.00±0.60 in groups I, II, III, and IV, respectively, which did not differ significantly. Although the results were non-significant, it suggests that implementing chelated minerals, vitamins, and energy supplements could be a promising strategy to improve reproductive efficiency in postpartum cows.

**Key words:** Dangi cows, Nutritional supplement, Postpartum fertility, Serum biochemistry.

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## INTRODUCTION

The profitability of dairy cattle rearing depends largely on reproductive performance. Productivity is strongly influenced by reproductive efficiency, with the ultimate aim being to achieve a calf per year per cow (Rekwot *et al.,* 2000). Unfortunately, in India, calving intervals of 16 to 20 months are common, compared to the ideal interval of 12-13 months, largely due to prolonged postpartum anestrus. The health, production, and reproduction of animals can be greatly influenced by their nutritional status. Imbalances in essential nutrients such as energy, protein, and minerals can contribute to infertility and poor reproductive performance in cows. Hormonal balance and coordination of reproductive events rely heavily on adequate energy intake, especially during early lactation. Protein, macro, and micronutrients also play crucial roles in the production and reproduction of farm animals through direct and indirect effects. Organic or chelated mineral sources have higher bioavailability than inorganic sources. Subclinical deficiencies in trace minerals can cause reproductive failure and poor body condition scores. To determine the adequacy of essential minerals in an animal's diet, plasma mineral status analysis can be conducted (Yugal Raj *et al.,* 2013).

During the transition period from late gestation to early lactation, cows undergo significant metabolic and endocrine adjustments (DeFrain *et al.,* 2005). This can result in reduced feed intake and a negative energy balance, which has been shown to impede fertility (Drackley and Cardoso, 2014). First postpartum estrus serves as an indicator of complete uterine involution, which can lead to the resumption of normal ovarian cycles, estrus behaviour, and conception. However, transition disorders, potentially caused by negative energy balance and delayed synthesis of steroidogenic hormones, can negatively impact ovarian activity (Roche *et al.,* 2018). Dangi farmers traditionally feed their animals crop residues like paddy straw due to the geographic location of their breeding tracts. However, this feed has poor nutritional value and can lead to malnutrition and reproductive underperformance. Farmers do not have much incentive to invest in higher quality feed since these animals don't produce high amounts of milk. Nutritional deficiencies can also lead to

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infertility and failure of postpartum estrus. To ensure smooth transitions and early postpartum fertility, it is important to provide sufficient energy, minerals, and vitamins. This study aims to evaluate the effects of postpartum supplementation of energy, chelated minerals, and vitamins on reproductive performance in postpartum Dangi cows.

## MATERIALS AND METHODS

The Dangi breed of draught cattle is indigenous to the Nashik and Ahmednagar districts of Maharashtra, as well as the hilly tracts of Western Ghats known as Dangs. A study was conducted between April and October 2021 on 24 recently calved Dangi cows with 2-5 parity at Dangi Cattle Breeding Farm in Igatpuri, District Nashik, with further laboratory research at Mumbai Veterinary College in Parel, Mumbai (India). Prior approval was obtained from the Institutional Animal Ethical Committee for the use of farm animals in the study. The cows were kept in well-ventilated and hygienic sheds, and were given access to open paddocks, green and dry fodder, concentrates, mineral mixture, and *ad-lib* water. Routine herd health programs were followed, including vaccination and deworming, to safeguard the animals from diseases.

The twenty-four experimental cows were divided into four groups at random. Group I (n=6) was assigned the routine standard farm feeding schedule and served as the control. Group II (n=6) was given a supplement of 30 gm of chelated minerals and vitamins (Bestmin Gold). Group III (n=6) was fed 100 gm of energy supplement (Galacshot / Bypass fat with herbal extracts, Provimin) and Group IV (n=6) received both 100 gm energy supplement and 30 gm chelated minerals and vitamins supplement daily/animal, in addition to their routine feeding schedule, for a period of 50 days following the calving. All the experimental cows were examined per-rectally on days 7, 14, 21 and 28 (±2 days) postpartum to study the status of the cervix, uterine horns and ovarian activity to monitor uterine involution and reproductive health status. Blood samples were collected on the day of calving, as well as on the 21st and 42nd day postpartum to analyze the serum levels of energy status (glucose and urea), protein (albumin and globulin), and minerals (calcium, phosphorus, and magnesium).

Cows that exhibited estrus were inseminated using the frozen semen of Dangi bulls with the standard protocol of artificial insemination. Various parameters like cervix status, uterine horns, uterine discharge, ovarian activity were observed during the experimental period. The study evaluated postpartum fertility by assessing various reproductive parameters, as calculated by Parkinson and Barrett (2009), over a period of 120 days following calving, *i.e.* First postpartum estrus in days = interval from calving to first post-partum exhibited estrus, First service conception rate = Percentage of cows conceived to first insemination, Submission rate = % of cows served over the total number of cows in the respective group, Reproductive efficiency = Submission rate x Overall conception rate /100, and Days open = period from calving to the subsequent effective / conceive service date. The data was analyzed statistically for analysis of variance (ANOVA) using Statistical Package for Social Sciences (SPSS, Version 20).

## RESULTS AND DISCUSSION

The postpartum period is a crucial phase in the reproductive cycle of cows as it significantly affects their future fertility. The findings on the effect of different supplements on various postpartum fertility parameters of Dangi cows are presented in Table 1.

Although there was no statistically significant difference between the groups in terms of the days required for the first estrus exhibition, Group IV had the lowest number of days required for the first postpartum estrus, followed by Group II and III, and Group I had the highest number of days required (Table 1). The energy balance is a crucial factor in re-establishing ovarian activity and insulin and IGF-I contribute to the impaired function of dominant follicles during the early postpartum period (Beam and Butler, 1999). Nanaware (2014) observed that postpartum first estrus was earlier in the cows with higher plane of the nutrition and energy supplement, compared to the control group. Theodore *et al.* (2016) revealed that peripartum nutritional supplementation had a positive impact on postpartum fertility, leading to a significantly shorter time to the first estrus and higher conception rates.

It is interesting to note that the first service conception rate (FSCR) was the highest in group IV (66.66%) compared to group II (50.00%), group III (40.00%) and the lowest in group I (33.33%). The combined chelated mineral, vitamins and energy supplements seemed to have improved FSCR in the

**Table 1:** Effect of nutrient supplementation (treatment) over control on postpartum fertility parameters in Dangi cows

| Sr. No. | Postpartum fertility parameters | Group I | Group II | Group III | Group IV |
|---|---|---|---|---|---|
| 1. | Days required for first PPE | 88.67 ± 0.55 | 71.25 ± 0.49 | 72.00 ± 0.49 | 68.50 ± 0.60 |
| 2. | Submission rate (%) | 50.00 | 66.66 | 83.33 | 100.00 |
| 3. | First service conception rate (FSCR, %) | 33.33 | 50.00 | 40.00 | 66.66 |
| 4. | Reproductive efficiency (%) | 16.67 | 33.33 | 33.33 | 66.66 |
| 5. | Days open | 91.00 ± 0.00 | 75.5 ± 0.31 | 85.00 ± 0.63 | 70.00 ± 0.60 |

early postpartum period as compared to chelated minerals, vitamins or energy supplementation singly and the non-supplemented control group. These findings were consistent with previous research by Parkinson and Barrett (2009), who found mean FSCR values of 40 to 58%. Additionally, Ashmead *et al.* (2004) and Nanaware (2014) observed higher conception rates in animals that were supplemented with chelated minerals or fed high plane of nutrition. Furthermore, Group IV under study had higher reproductive efficiency than others. Nanaware (2014) also found better reproductive efficiency in treatment group (energy supplementation) in comparison to control group.

The study found that Group I had the highest number of days open (91.00±0.00), while Group IV had the lowest (70.00±0.60), Group II and III being intermediate. However, statistically, the differences were non-significant. These results aligned with Ashmead *et al.* (2004) in cows supplemented with chelated and inorganic minerals. However, Jadhav (2005) reported relatively higher number of days open for high-plane nutrition compared to the control group, while Nanaware (2014) found that energy supplementation resulted in fewer days open than the control group. In the present study, satisfactory estrus exhibition and conception rate were observed which may be due to combined effect of energy, various macro-micro minerals and vitamins, which have positive effects on steroidogenesis, follicular growth and symptoms of ovulatory estrus (Anonymous, 2018).

The mean serum biochemical parameters of Dangi cows observed on postpartum days 0, 21 and 42 in both control and treatment groups are presented in Table 2.

Average values of glucose, urea, albumin, globulin, calcium, phosphorus, and magnesium found in the study

**Table 2:** Effect of nutrient supplementation (treatment) over control on mean serum biochemical parameters in Dangi cows

| Parameters | PP Days | Group I | Group II | Group III | Group IV | 'F' value |
|---|---|---|---|---|---|---|
| Glucose (g/dL) | 0 | 48.50 ± 2.41 | 47.33 ± 1.54 | 48.50 ± 2.86 | 49.67 ± 0.76 | $0.214^{NS}$ |
| | 21 | 44.50 ± 0.40 | 47.00 ± 0.35 | 47.17 ± 0.38 | 48.33 ± 0.37 | $0.613^{NS}$ |
| | 42 | $41.33^{a} \pm 0.32$ | $47.17^{b} \pm 0.37$ | $50.00^{bc} \pm 0.32$ | $53.50^{c} \pm 0.31$ | $9.811^{**}$ |
| | F value | $2.843^{NS}$ | $0.008^{NS}$ | $0.407^{NS}$ | $3.301^{NS}$ | |
| Urea (mg/dL) | 0 | $26.17^{ab}\pm 2.36$ | $20.89^{a\pm}$ 1.65 | $22.73^{ab} \pm 1.63$ | $28.45^{Ab}\pm 1.61$ | $3.395^{*}$ |
| | 21 | 26.08 ± 3.12 | 21.14 ± 1.68 | 19.52 ± 2.02 | $21.93^{B} \pm 1.50$ | $1.648^{NS}$ |
| | 42 | 28.79 ± 6.19 | 17.47 ± 1.10 | 20.50 ± 1.92 | $16.33^{B} \pm 2.27$ | $2.607^{NS}$ |
| | F value | $0.132^{NS}$ | $1.859^{NS}$ | $0.777^{NS}$ | 10.986** | |
| Albumin (g/dL) | 0 | 4.00 ± 0.52 | 3.13 ± 0.27 | 3.23 ± 0.34 | 2.63 ± 0.09 | $2.758^{NS}$ |
| | 21 | $4.81^{a} \pm 0.82$ | $3.30^{ab} \pm 0.35$ | $2.78^{b} \pm 0.13$ | $2.70^{b} \pm 0.05$ | $4.695^{*}$ |
| | 42 | 4.12 ± 0.57 | 3.12 ± 0.35 | 2.80 ± 0.36 | 2.70 ± 0.04 | $2.918^{NS}$ |
| | F value | $0.462^{NS}$ | $0.100^{NS}$ | $0.754^{NS}$ | $0.357^{NS}$ | |
| Globulin (g/dL) | 0 | $2.55^{a} \pm 0.32$ | $4.27^{b} \pm 0.23$ | $3.80^{b} \pm 0.30$ | $4.33^{b} \pm 0.19$ | 9.608** |
| | 21 | $2.59^{a} \pm 0.33$ | $3.96^{b} \pm 0.18$ | $4.47^{b} \pm 0.38$ | $4.15^{b} \pm 0.15$ | 8.792** |
| | 42 | $3.11^{a} \pm 0.55$ | $4.23^{ab} \pm 0.22$ | $4.08^{ab} \pm 0.16$ | $4.45^{b} \pm 0.19$ | 3.353* |
| | | $0.557^{NS}$ | $0.585^{NS}$ | $1.282^{NS}$ | $0.706^{NS}$ | |
| Calcium (mg/dL) | 0 | 7.99 ± 1.04 | 8.88 ± 0.31 | 8.57 ± 0.39 | 8.39 ± 0.29 | $0.392^{NS}$ |
| | 21 | 8.35 ± 0.72 | 8.28 ± 0.29 | 8.56 ± 0.46 | 8.62 ± 0.41 | $0.109^{NS}$ |
| | 42 | 9.54 ± 0.62 | 9.07 ± 0.72 | 8.37 ± 0.21 | 8.75 ± 0.18 | $1.019^{NS}$ |
| | F value | $1.000^{NS}$ | $0.735^{NS}$ | $0.098^{NS}$ | $0.341^{NS}$ | |
| Phosphorus (mg/dL) | 0 | 3.98 ± 0.99 | 4.82 ± 0.35 | 4.68 ± 0.31 | 4.53 ± 0.42 | $0.396^{NS}$ |
| | 21 | 4.56 ± 0.28 | 4.68 ± 0.50 | 4.63 ± 0.35 | 4.52 ± 0.29 | $0.040^{NS}$ |
| | 42 | 4.20 ± 0.27 | 4.03 ± 0.22 | 4.63 ± 0.67 | 6.72 ± 1.42 | $2.359^{NS}$ |
| | F value | $0.226^{NS}$ | $1.244^{NS}$ | $0.003^{NS}$ | $2.096^{NS}$ | |
| Magnesium (mg/dL) | 0 | $3.01^{a} \pm 0.15$ | $2.69^{ab} \pm 0.24$ | $2.11^{c} \pm 0.06$ | $2.20^{bc} \pm 0.09$ | 7.867** |
| | 21 | $3.15^{a} \pm 0.09$ | $2.51^{b} \pm 0.16$ | $2.32^{b} \pm 0.12$ | $2.22^{b} \pm 0.10$ | 11.882** |
| | 42 | $2.93^{a} \pm 0.19$ | $2.98^{a} \pm 0.26$ | $2.25^{b} \pm 0.10$ | $2.05^{b} \pm 0.08$ | 7.328** |
| | F value | $0.599^{NS}$ | $1.121^{NS}$ | $1.256^{NS}$ | $0.970^{NS}$ | |

*Significant at 5% level, **Significant at 1% level, NS Non-significant.
Means bearing uncommon small superscripts within the row and capital superscripts within the column differ significantly ($p < 0.05$).

were within the normal range of 45-75 g/dL, 12.84-57.8 mg/dL, 2.1-3.36 g/dL, 3.6-4.5 g/dL, 8.4-11 mg/dL, 4.3-7.8 mg/dL, and 1.55-3.0 mg/dL, respectively, as reported by Radostits *et al.* (2000).

When the data were evaluated critically to study the effect of duration (treatment period effect) of supplementation on animals from various treatment groups, it was observed that there has been a significant increase in serum glucose at postpartum 42nd day in energy supplement groups, with maximum positive effect seen in group IV suggesting the impact of nutritional supplement on the energy status of animals. Similar results were also reported by Burke *et al.* (2010) and Nanaware (2014). However, Dhami *et al.* (2019) found that the blood glucose levels ranged from 31 to 67 mg/dL among individual animals. The mean urea value observed in group IV on day 42 was significantly decreased. Similar findings were reported in the postpartum period by Pal and Acharya (2013) and Nanaware (2014)

The average albumin values were significantly lower in groups III and IV on day 21, whereas for all groups values fell within normal range. These findings were consistent with findings of Rego (2007) and Nanaware (2014). Additionally, statistical analysis revealed that there were no significant differences in albumin levels between or within the four groups. The mean globulin values were significantly higher for treatment groups compared to control. These findings concurred with the reports of Rego (2007) and Nanaware (2014). However, there were no significant differences in albumin levels between the four groups. It is worth noting that Nanaware (2014) did observe an increase in globulin concentration three weeks after parturition.

Throughout the study, it was found that the average calcium and phosphorus levels for all four groups remained in a normal range. On statistical analysis, no significant differences either between or within the groups were revealed. The observations of the postpartum period's changes in calcium concentrations align with the conclusions of various experts, including Yokus *et al.* (2010) and Nanaware (2014). As a result, we can assertively state that there is no notable variation in calcium and phosphorus levels among the groups. However, Benzaquen *et al.* (2015) did find relatively higher values than those observed in this study.

There was significant difference in magnesium levels between the four groups, whereas non-significant within the group. Melendez *et al.* (2004) noted a trend of fluctuation in serum magnesium levels, while Hadiya *et al.* (2010) and Piccione *et al.* (2012) reported an increase in magnesium levels from the day of calving, with a rise in lactation days. However, the average serum magnesium concentration did not align with the observations of Yokus *et al.* (2010), who recorded relatively higher magnesium levels. According to Dhami *et al.* (2019), repeat breeder cattle had slightly higher levels of calcium, phosphorus, and magnesium in their plasma than anestrus or subestrus cattle. Phosphorus deficiency or an improper calcium and phosphorus ratio may cause anestrus conditions in dairy animals.

## Conclusion

From present study, it can be concluded that supplementing with combined chelated minerals, vitamins, and energy can significantly improve postpartum reproductive efficiency in Dangi cows by accelerating uterine involution, resumption of ovarian cyclist, early postpartum estrus, reduced days open, and improved first service conception rate. The combined supplementation is more effective compared to chelated minerals, vitamin supplements, or energy supplements alone, and control groups. Based on the findings of the study, the use of combined chelated minerals, vitamins, and energy supplementation can be recommended for improving postpartum reproductive efficiency in cows.

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## References

Anonymous (2018). Annual Report of AICRP on nutritional and physiological interventions for enhancing reproductive performance in animals, AAU, Anand Centre. ICAR-NIANP, Bangalore, India.

Ashmead, H.D., Ashmead, S.D., & Samford, R.A. (2004). Effect of metal amino acid chelated on milk production, reproduction and body conditions in Holstein first calf heifers. *International Journal of Applied Research in Veterinary Medicine, 2*(4), 252-260.

Beam, S.W., & Butler, W.R. (1999). Effect of energy balance on follicular development and first ovulation in postpartum dairy cows. *Journal* of *reproduction* and *fertility*. *Supplement*, *54,* 411-421.

Benzaquen, N., Galvao, K.N., Coleman, A.E., Santos, J.E.P., Goff, J.P., & Risco, C.A. (2015). Effect of oral mineral and energy supplementation on blood mineral concentrations, energetic and inflammatory profile and milk yield in dairy cows affected with dystocia. *The Veterinary Journal, 204*(2), 186-191.

Burke, C.R., Meier, S., McDougall, S., Compton, C., Mitchell, M., & Roche, J.R. (2010). Relationships between endometritis and metabolic state during the transition period in pasture-grazed dairy cows. *Journal* of *Dairy Science, 93*(11), 5363-5373.

DeFrain, J.M., Hippen, A.R., Kalscheurand, K.F., & Patton, R.S. (2005). Effects of feeding propionate and calcium salts of long-chain fatty acids on transition dairy cow performance. *Journal* of *Dairy Science, 88,* 983-993.

Dhami, A.J., Patel, J.A., Hadiya, K.K., Parmar, S.C., & Chaudhari, D.V. (2019) Nutritional infertility and ameliorative measures in dairy animals of middle Gujarat. *The Indian Journal of Veterinary Science and Biotechnology, 14*(3), 5-9.

Drackley, J.K., & Cardoso F.C. (2014) Pre partum and postpartum nutritional management to optimize fertility in high-yielding dairy cows in confined TMR systems, *Animal, 8*(s1), 5-14.

Hadiya, K.K., Derashri, H.J., Devalia, B.R., & Jani, R.G. (2010). Effect of supplementation of minerals and enzymes on service period and postpartum plasma minerals profile in crossbred cows. *Veterinary World, 3*(4), 173-176.

Jadhav, P.D. (2005). Study of postpartum resumption of ovarian activity and uterine involution, monitored ultrasonography and hormonal assays in Gir and crossbred cows. *PhD Thesis.* Maharashtra Animal & Fishery Sciences University, Nagpur, India

Melendez, P., McHale, J., Bartolome, J., Archbald, L.F., & Donovan, G.A. (2004). Uterine involution and fertility of Holstein cows subsequent to early postpartum PGF2α treatment for acute puerperal metritis. *Journal* of *Dairy Science, 87,* 3238-3246.

Nanaware, R.D. (2014). Amelioration of post-partum fertility with increased dry matter intake and energy supplementation in Gir and its crosses. *M.V.Sc. Thesis.* Maharashtra Animal & Fishery Sciences University, Nagpur, India

Pal, K., & Acharya, H.R. (2013). Subclinical metabolic disorders in post-partum cross bred HF cattle in central part of Nepal. *International Journal of Pharma Medicine and Biological Sciences, 2*(2), 57-62.

Parkinson, T.J., & Barrett, D. (2009). Veterinary control of hard fertility Ch. 24, In: D. E. Noakes, T. J. Parkinson and G.C.W. England, eds. *Veterinary Reproduction and Obstetrics*. 9th edn. Saunders, p. 517-558.

Piccione, G., Messina, V., Marafioti, S., Casella, S., Giannetto, C., & Fazio, F. (2012). Changes of some haemato chemical parameters in dairy cows during late gestation, postpartum, lactation and dry periods. *Veterinary Medicine and Zootechnics, 58,* 59-64.

Radostits, O.M., Gay, C.C. Blood, D.C., & Hinchcliff, K.W. (2000). *Veterinary Mdicine*. 9th ed. W.B. Saunders Co., London.

Rego, N.L. (2007). Evaluation of reproductive performance by ultrasonography and serum progesterone estimation in crossbred cows supplemented with chealated trace minerals. *M.V.Sc. Thesis*. Maharashtra Animal & Fishery Sciences University, Nagpur, India

Rekwot, P.I., Ogwu, D., & Oyedipe, E. O. (2000). Influence of bull bio stimulation, season and parity on resumption of ovarian activity of Zebu (*Bos indicus*) cattle following parturition. *Animal Reproduction Science, 63*(1-2), 1-11.

Roche, J.R., Burke, C.R., Crookenden, M.A., Heiser, A., Loor, J.L., Meier, S., & Turner, S.A. (2018). Fertility and the transition dairy cow. *Reproduction, Fertility and Development, 30*(1), 85-100.

Theodore, V.K., Panchal, M.T., Dhami, A.J., Patel, J.A., & Ramani, V.P. (2016). Influence of peripartum nutritional supplementation on plasma macro-micro minerals profile and postpartum fertility in crossbred cows. *International Journal of Environmental Science and Technology*, *5*(6), 4052-4060.

Yokus, B., Cakir, D., Icen, H., Dukar, H., & Bademkiran, S. (2010). Prepartum and postpartum serum mineral and steroid hormone concentration in cows with dystocia. *Veteriner Fakultesi Dergisi, 21,* 185-90.

Yugal Raj, B., Sulochana, S., Nabaraj, S., & Tara Nath, G. (2013). Effects of nutrition on reproduction - A review. *Advances in Applied Science Research, 4*(1), 421-429.