



Nano Zinc Supplementation in Sahiwal Cows

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Effect of Nano Zinc Supplementation on Hematological Parameters and Body Condition Score During Transition Period in Sahiwal Cows

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ABSTRACT

Twenty four Sahiwal cows during transition period were randomly divided into four groups (control, Zn₄₀, nanoZn₂₀ and nanoZn₄₀) with 6 animals in each, based on parity (2-5 parity) to evaluate the effect of nano zinc oxide on hematology and body condition score. Similar basal diet was followed in all the groups whereas in groups Zn₄₀, nanoZn₂₀ and nanoZn₄₀ additional 40 ppm Zn from ZnO, and 20 and 40 ppm Zn from nano ZnO was supplemented, respectively. Results indicated that prepartum and postpartum mean values of TEC, Hb and PCV were significantly (P<0.01) higher in nanoZn₂₀ group. Mean values of TLC was found to be higher in nanoZn₂₀ group. The lymphocyte and neutrophil per cent were higher and lower, respectively during prepartum compared to postpartum period. Significantly lower value of neutrophil was found on the day of calving in nanoZn₄₀. Monocyte, eosinophil and basophil levels were found similar in all groups throughout transition period, however, monocyte per cent was significantly higher at 21 days postpartum in nanoZn₂₀ group. There was no significant difference in mean BCS during prepartum and postpartum. There was maximum reduction in BCS from 21 days prepartum to 21 days postpartum in control group compared to that of Zn₄₀, nanoZn₄₀ and nanoZn₂₀ groups. Body condition score of Sahiwal cow was better in nanoZn₂₀ group. Sahiwal cows supplemented with 20 ppm nano Zn showed better hematological profile and BCS compared to unsupplemented and other 40 ppm nano Zn or inorganic Zn supplemented groups.

KEYWORDS: Body condition score, Nano zinc, Sahiwal cow, Transition period

Article received : 27 May 2022; Article Accepted: 27 June 2022

INTRODUCTION

In dairy cattle, transition period is crucial in terms of its effect on health and the subsequently on production performance. A significant shift in physiological and metabolic status occurs during this period. Hematological profile and body condition are important in assessing the physiological health of animals during this period. Body condition score is one of the important determinants of herd profitability. Decreased body condition score at post calving is likely an indicator of negative energy balance. Oxidative stress increases when there is significant loss in body condition score during peripartum period (Bernabucci et al., 2005). Poor body condition score is apparently associated with decreased profitability of herd as it may increase interval of post-partum oestrus and birth of weak calves etc. So, it needs time-dependent management

and dynamic approaches, especially implementation of effective feeding strategy. The target of nutritional management during transition period is to support the metabolic adaptations in energy and mineral metabolism that the animal undergoes, for milk yield and avoid metabolic disorders. Minerals, trace elements and vitamins play a vital role in nutrient metabolism that requires supplementation of trace minerals during the transition period (Santoshi et al., 2018).

Nano minerals have stronger absorbing ability, greater specific surface area, higher surface activity and high catalytic efficiency. The use of nanotechnology to alter the size, shape and crystalline structure to produce nano zinc oxide, has possibility against both organic and inorganic sources of zinc to improve livestock performance. The use of nano zinc was proven to have better

effects in comparison inorganic and organic zinc sources and was also much less toxic and further nano Zn has potential to cross the small intestine more effectively (Swain et al., 2021).

MATERIALS AND METHODS

Twenty-four pregnant pure bred Sahiwal cows were randomly selected on the basis of well-maintained farm records from Bull Mother Experimental Farm (BMEF), College of Veterinary Science and A.H. (COVS), Anjora, Durg. These animals were on a diet of green fodder, dry fodder (paddy straw) and concentrates as per feeding standard in the farm. The green fodder consisted of Hybrid Napier, Sudan grass and local grasses as per their availability along with daily grazing for 4 hours at morning. The concentrate mixture contained 20% crude protein, 18% crude fiber, 0.93% ether extract and 14% total ash. The nano zinc capsule was made and fed by mixing it with the concentrate. Feeding was done under our supervision to ensure the entrance of capsule. In control group no additional Zn was supplemented whereas in Zn₄₀ group 40 ppm Zn was supplemented from ZnO and in nanoZn₂₀ and nanoZn₄₀ groups 20 and 40 ppm Zn was supplemented from nano ZnO, respectively. The experimental supplementation was done during transition phase starting 30 days prepartum to 30 days postpartum.

Hematological parameters

In the morning before feeding and watering, blood samples were collected from jugular vein in EDTA vacutainer tubes. The sampling was done on -21, -14, -7, -5, -3, -1 days prepartum and on the day of calving (day 0) as well as on +1, +3, +5, +7, +14, +21 postpartum from individual animals. Before mixing the EDTA, blood smear was immediately prepared for DLC (differential leucocyte count) and smear was fixed in Methanol for 1 minute and was stained with Geimsa stain for 30 minutes followed by counting. Analysis of hematological parameters was done by automated haematology blood cell counter (URIT- 2900 Vet Plus). The parameters estimated were Total erythrocyte count (TEC), Haemoglobin (Hb),

Packed cell volume (PCV), Total leucocyte count (TLC) and Differential leucocyte count (DLC). The values were expressed in 10⁶/cu mm for TEC, gm/dl for Hb, percentage for PCV and DLC and 10³/cu mm blood for TLC.

Body condition score

Body condition score (BCS) of all the experimental animals were recorded on days -21, -14, -7, -5, -3, -1 prepartum, on the day of calving (day 0) as well as on +1, +3, +5, +7, +14, +21 days postpartum. A scoring system of 1 to 5 point scale was used with quarter increments for Sahiwal cows as suggested by Ferguson et al. (1994). For this purpose, 7 check points viz. Loin (short ribs), Pin bone, Hook bone, Thurl (rump), Tail head ligament, Sacral ligament were examined for their changes in appearance with respect to the changes in body check points of cattle.

Statistical analysis

To see the difference between different treatment groups one way analysis of variance was applied as per procedure given by Snedecor and Cochran (1994). If there was any significant difference existed in any group then DMRT was applied as per procedure given by Steel and Torrie (1984).

RESULTS AND DISCUSSION

Total erythrocyte count (TEC)

Increased level of TEC was observed from 21 days of prepartum period up to day of calving, whereas, after calving level of TEC decreased among all the groups. The prepartum and postpartum mean values of TEC in nanoZn₂₀ group was highly significant (Pd^{0.01}) than other groups. The overall mean TEC during prepartum period were higher than that of postpartum period among all the groups. These results were in agreement with the findings of Aswal et al. (2021), where they observed significantly (P<0.01) higher values of TEC on the day of calving compared to the prepartum and postpartum period in Sahiwal cows, which might be due to erythropoiesis and the slow destruction of erythrocytes. In contrary to present findings, Abdelrazek et al. (2018) reported TEC values were

decreased at parturition in HF cows. Mean values of TEC were significantly higher (Pd^{0.01}) in both nanoZn₂₀ and nanoZn₄₀ groups compared to Zn₄₀ and control (Table 1). In corroboration with present finding, Ulutas et al. (2020) who observed positive association of Zn with erythrocyte production

evident from increased level of RBC in Zn supplemented goats. Similarly, significant increase in the RBC with supplementation of different levels of Zinc in Murrah buffalo calves was reported by Kumar et al. (2018).

Table 1. Mean values (±SE) of Total erythrocyte count (x 10⁶/cu.mm) in Sahiwal cows

Periods of observations Days relative to calving	TEC in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
-21	6.00 ± 0.29	6.44 ± 0.22	6.79 ± 0.24	6.69 ± 0.32	NS
-14	6.03 ± 0.27	6.45 ± 0.19	6.84 ± 0.34	6.78 ± 0.05	NS
-7	6.09 ± 0.31	6.47 ± 0.46	6.92 ± 0.42	6.82 ± 0.37	NS
-5	6.14 ± 0.33	6.48 ± 0.17	6.96 ± 0.31	6.83 ± 0.20	NS
-3	6.19 ± 0.27 ^b	6.51 ± 0.10 ^{ab}	6.98 ± 0.17 ^a	6.85 ± 0.02 ^a	*
-1	5.29 ± 0.11 ^b	6.60 ± 0.24 ^a	7.21 ± 0.19 ^a	6.92 ± 0.27 ^a	**
At Calving	6.32 ± 0.39 ^c	6.67 ± 0.07 ^{bc}	7.58 ± 0.14 ^a	7.09 ± 0.23 ^{ab}	**
+1	5.98 ± 0.30	6.17 ± 0.15	6.81 ± 0.21	6.75 ± 0.42	NS
+3	5.93 ± 0.26	6.15 ± 0.22	6.78 ± 0.37	6.65 ± 0.10	NS
+5	4.90 ± 0.14 ^c	6.20 ± 0.16 ^b	6.75 ± 0.20 ^a	6.74 ± 0.04 ^a	**
+7	5.62 ± 0.26 ^b	6.16 ± 0.21 ^{ab}	6.70 ± 0.31 ^a	6.70 ± 0.22 ^a	*
+14	5.12 ± 0.25 ^b	5.93 ± 0.23 ^{ab}	6.53 ± 0.25 ^a	6.62 ± 0.48 ^a	*
+21	4.70 ± 0.33 ^b	5.41 ± 0.27 ^b	6.54 ± 0.18 ^a	6.47 ± 0.13 ^a	**
Mean	5.71 ± 0.14 ^c	6.28 ± 0.09 ^b	6.87 ± 0.07 ^a	6.76 ± 0.04 ^a	**
Prepartum Mean	5.95 ± 0.13 ^c	6.49 ± 0.02 ^b	6.95 ± 0.05 ^a	6.81 ± 0.03 ^a	**
Postpartum Mean	5.37 ± 0.22 ^c	6.0 ± 0.12 ^b	6.68 ± 0.04 ^a	6.65 ± 0.04 ^a	**

^{a,b,c}- Mean with different superscript in a row differ significantly *(Pd^{0.05}) & ***(Pd^{0.01})

Hemoglobin profile (Hb)

Mean Hb during prepartum period were higher compared to postpartum period among all the groups

however; prepartum and postpartum mean values of Hb was significantly (Pd^{0.01}) higher in nanoZn₂₀ (Table 2).

Table 2. Mean values (±SE) of Hemoglobin concentration (g/dl) in Sahiwal cows

Periods of observations Days relative to calving	Mean Hb conc. (g/dl) in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
-21	9.34 ± 0.36 ^c	9.42 ± 0.43 ^c	11.7 ± 0.16 ^a	10.7 ± 0.28 ^b	**
-14	9.41 ± 0.52 ^b	9.67 ± 0.46 ^b	12.0 ± 0.27 ^a	10.9 ± 0.40 ^a	**
-7	9.49 ± 0.65 ^b	9.86 ± 0.50 ^b	11.8 ± 0.47 ^a	10.8 ± 0.29 ^{ab}	*
-5	9.56 ± 0.34 ^c	10.0 ± 0.58 ^{bc}	11.8 ± 0.47 ^a	11.0 ± 0.42 ^{ab}	**
-3	9.62 ± 0.73	10.2 ± .49	12.1 ± 0.76	11.1 ± 0.60	NS
-1	9.74 ± 0.97	10.3 ± .59	12.2 ± 0.47	11.3 ± 0.63	NS
At Calving	10.9 ± 0.48	10.9 ± 0.57	12.2 ± 0.68	11.4 ± 0.61	NS
+1	9.21 ± 0.20 ^c	9.94 ± 0.31 ^{bc}	11.6 ± 0.36 ^a	10.6 ± 0.42 ^b	**
+3	9.34 ± 0.34 ^c	9.90 ± 0.32 ^{bc}	11.5 ± 0.21 ^a	10.5 ± 0.38 ^b	**
+5	9.23 ± 0.29 ^b	9.86 ± 0.42 ^b	11.44 ± 0.49 ^a	10.39 ± 0.29 ^{ab}	**
+7	9.07 ± 0.32 ^b	9.75 ± 0.15 ^b	11.28 ± 0.32 ^a	10.64 ± 0.27 ^a	**
+14	9.39 ± 0.44	9.99 ± 0.58	11.26 ± 0.58	10.40 ± 0.48	NS
+21	10.00 ± 0.36	10.01 ± 0.68	11.52 ± 0.44	10.50 ± 0.61	NS
Mean	9.56 ± 0.13 ^d	10.00 ± 0.10 ^c	11.75 ± 0.09 ^a	10.82 ± 0.09 ^b	**
Prepartum Mean	9.52 ± 0.05 ^d	9.93 ± 0.14 ^c	11.96 ± 0.07 ^a	11.02 ± 0.08 ^b	**
Postpartum Mean	9.37 ± 0.13 ^d	9.90 ± 0.03 ^c	11.45 ± 0.06 ^a	10.52 ± 0.04 ^b	**

^{a,b,c,d}- Mean with different superscript in a row differ significantly *(Pd^{0.05}) & ***(Pd^{0.01})

Our results are in agreement with Aswal et al. (2021), where they observed increase in Hb concentration from 2 weeks before calving to the day of calving, which reached its highest value on the day of calving. Significantly higher ($P < 0.01$) level of hemoglobin was noticed in nanoZn₂₀ group, particularly on 5, 14 and 21 days prepartum and 1, 3, 5 and 7 days postpartum related to other groups. Mean values of Hb were significantly ($P < 0.01$) higher in nanoZn₂₀ group across the treatments and control group which might be due to the role of zinc as cofactor for enzymes involved in Hb synthesis. The present findings are in accordance with EI Hendy et al. (2001) who reported decrease in hemoglobin on Zn deficient diet in growing rats. However, Mandal and Dass (2010) reported that concentration of hemoglobin was similar and within the normal range for organic and inorganic Zn supplemented groups. Nevertheless Hb is within the

reference range of 8.4-13.0 g/dl for cattle (Wood and Rocha, 2010).

Packed cell volume (PCV)

Higher significant level of PCV was noticed in nanoZn₂₀ group particularly at 5 days ($P < 0.01$) and 3 days ($P < 0.05$) of prepartum period. The level of PCV reached its maximum value on the day of calving among all the groups, followed by a steady reduction after the day of calving to 3rd week after calving. The prepartum and postpartum mean values of PCV in nanoZn₂₀ group was highly significant ($P < 0.01$) than other groups. The overall mean PCV during prepartum period were higher compared to postpartum period among all the groups (Table 3). Our results are in agreement with Aswal et al. (2021), where they observed that PCV level increased significantly ($P < 0.01$) up to the day of calving, which reached its maximum on the day of calving followed by a steady reduction.

Table 3. Mean values (\pm SE) of Packed Cell Volume (%) in Sahiwal cows

Periods of observations	Mean PCV in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
Days relative to calving					
-21	35.1 \pm 0.86	36.3 \pm 1.01	38.0 \pm 1.05	36.1 \pm 1.01	NS
-14	35.2 \pm 0.74	36.6 \pm 0.85	38.1 \pm 1.31	36.4 \pm 1.31	NS
-7	34.6 \pm 1.08	36.2 \pm 0.59	37.6 \pm 0.76	36.7 \pm 1.36	NS
-5	34.2 \pm 0.44 ^c	35.4 \pm 0.67 ^b ^c	37.8 \pm 0.53 ^a	37.1 \pm 0.75 ^{ab}	**
-3	34.8 \pm 0.77 ^b	35.6 \pm 1.00 ^b	38.2 \pm 0.59 ^a	36.7 \pm 0.67 ^{ab}	*
-1	35.2 \pm 0.93	36.4 \pm 1.31	38.3 \pm 0.49	37.2 \pm 0.82	NS
At Calving	35.6 \pm 1.00	36.5 \pm 0.74	38.4 \pm 0.59	37.4 \pm 1.4	NS
+1	35.0 \pm 0.70	35.4 \pm 0.75	37.7 \pm 0.63	36.5 \pm 1.36	NS
+3	34.6 \pm 0.63	34.9 \pm 0.68	37.4 \pm 0.70	36.1 \pm 1.07	NS
+5	34.8 \pm 0.60	35.0 \pm 0.59	38.0 \pm 1.05	37.2 \pm 1.24	NS
+7	34.1 \pm 0.75	34.8 \pm 0.75	36.4 \pm 1.31	34.5 \pm 1.56	NS
+14	32.6 \pm 0.74	35.2 \pm 1.21	35.6 \pm 13.6	33.9 \pm 1.46	NS
+21	32.0 \pm 0.63	33.1 \pm 1.21	34.1 \pm 1.10	34.6 \pm 1.31	NS
Mean	34.4 \pm 0.28 ^c	35.5 \pm 0.26 ^b	37.4 \pm 0.35 ^a	36.2 \pm 0.31 ^b	**
Prepartum Mean	34.8 \pm 0.16 ^d	36.1 \pm 0.20 ^c	38.0 \pm 0.10 ^a	36.7 \pm 0.17 ^b	**
Postpartum Mean	33.9 \pm 0.25 ^c	34.7 \pm 0.81 ^{bc}	36.5 \pm 1.48 ^a	35.5 \pm 1.30 ^{ab}	**

^{a,b,c,d} Mean with different superscript in a row differ significantly *($P < 0.05$) and **($P < 0.01$)

Mean value of PCV was significantly higher in nanoZn₂₀ group. Besides, variation in PCV values in relation to the prepartum and postpartum period. These findings are in accordance with the findings of Kumar et al. (2018), reported that the PCV level were highest in group supplemented with zinc sulphate. Sethy et al. (2018) observed that the mean

PCV values (%) was found comparable in among all groups, however, values were within normal range. Different blood parameters were not affected by different sources of zinc was reported by Kumar et al. (2021). The present finding is within the reference range of 24-46 % for cattle reported by Jain (1989).

Total leucocyte count (TLC)

There were higher values of TLC on the day of calving among all the groups due to secretion of higher level of cortisol, which in turn stimulate the bone marrow which increases the leukocyte count in blood and then decreased after the day of calving to 1st week of postpartum. Mean TLC during prepartum period was higher than that of postpartum period among all the groups. Our results are in

agreement with Aswal et al. (2021), where they observed the concentration of TLC was increased significantly from 2nd weeks before calving until the day of calving and then decreased in the 1st week of calving due to result of an increase in the neutrophil count in dairy cows during transition period. Mean values of TLC were highest in nanoZn₂₀ group than other groups (Table 4).

Table 4. Mean values (\pm SE) of Total leucocyte count ($\times 10^3/\text{cu.mm}$) in Sahiwal cows

Periods of observations	TLC in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
Days relative to calving					
-21	6.87 \pm 0.66	7.15 \pm 0.72	7.72 \pm 0.62	7.57 \pm 0.56	NS
-14	7.08 \pm 0.81	7.36 \pm 0.76	8.17 \pm 0.69	7.43 \pm 0.65	NS
-7	8.16 \pm 0.69	8.22 \pm 0.59	8.78 \pm 0.77	8.37 \pm 0.69	NS
-5	8.34 \pm 0.48	8.41 \pm 0.36	8.92 \pm 0.300	8.52 \pm 0.34	NS
-3	8.55 \pm 0.59	8.55 \pm 0.38	9.11 \pm 0.54	9.21 \pm 0.33	NS
-1	8.90 \pm 0.56	9.16 \pm 0.54	9.71 \pm 0.64	9.68 \pm 0.69	NS
At Calving	8.95 \pm 0.66	9.32 \pm 0.64	9.76 \pm 0.74	9.55 \pm 0.69	NS
+1	8.51 \pm 0.78	9.52 \pm 0.51	10.4 \pm 0.39	8.99 \pm 0.63	NS
+3	8.46 \pm 0.55	8.91 \pm 0.35	9.09 \pm 0.53	8.99 \pm 0.61	NS
+5	8.04 \pm 0.69	8.50 \pm 1.96	8.49 \pm 0.55	8.27 \pm 0.60	NS
+7	6.96 \pm 0.57	7.13 \pm 0.78	7.76 \pm 0.69	7.51 \pm 0.75	NS
+14	7.08 \pm 0.43	7.40 \pm 0.68	7.98 \pm 0.55	7.72 \pm 0.48	NS
+21	7.22 \pm 0.54	8.08 \pm 0.79	8.40 \pm 0.74	8.56 \pm 0.89	NS
Mean	7.93 \pm 0.21	8.29 \pm 0.22	8.79 \pm 0.22	8.49 \pm 0.21	NS
Prepartum Mean	7.98 \pm 0.33	8.14 \pm 0.30	8.73 \pm 0.28	8.46 \pm 0.36	NS
Postpartum Mean	7.71 \pm 0.28	8.25 \pm 0.37	8.69 \pm 0.39	8.34 \pm 0.25	NS

Present finding are in accordance with Kumar et al. (2018) where, they observed that the values of TLC were highest in group supplemented either with zinc sulphate or zinc methionine. Poonia et al. (2018) reported a similar trend of TLC that there were gradually higher value of leukocyte count in all supplemented and control cows from 30 days prepartum period to the days of calving and remained highest on the day of calving but gradually lower number of leukocytes after calving was observed. Nevertheless conc. of TLC is within the reference range of 5.1-13.3 $\times 10^3/\mu\text{l}$ for cattle (Wood and Rocha, 2010).

Differential leucocyte count (DLC)**Lymphocyte profile**

There were lower values of lymphocyte were found on the day of calving among all the groups due to the mammary gland, which extensively influx the neutrophils into colostrums and milk occurs around calving (Guidry et al., 1976) and then increased after the day of calving to 3rd week of postpartum, there was a gradual rise in mean lymphocyte levels among the entire groups. Mean lymphocyte during prepartum period was higher than that of postpartum period. The mean values of lymphocyte were highest in control group (Table 5) in present finding but the variations were found to be statistically insignificant.

Table 5. Mean values (\pm SE) of lymphocyte (%) in Sahiwal cows

Periods of observations	Mean lymphocyte in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
Days relative to calving					
-21	70.8 \pm 0.70	70.2 \pm 1.48	71.4 \pm 0.69	70.8 \pm 2.10	NS
-14	70.4 \pm 1.44	71.3 \pm 2.21	70.3 \pm 0.88	71.3 \pm 0.93	NS
-7	65.5 \pm 1.55	66.8 \pm 1.29	66.6 \pm 2.17	65.2 \pm 1.69	NS
-5	63.7 \pm 1.34	63.0 \pm 1.85	61.7 \pm 0.89	62.6 \pm 1.43	NS
-3	62.4 \pm 2.29	60.3 \pm 2.11	61.3 \pm 1.56	60.2 \pm 1.28	NS
-1	59.6 \pm 2.25	57.8 \pm 1.99	58.0 \pm 2.20	57.9 \pm 1.49	NS
At calving	56.7 \pm 2.69	56.1 \pm 1.58	56.7 \pm 2.36	57.2 \pm 2.30	NS
+1	58.3 \pm 2.78	59.3 \pm 3.30	57.4 \pm 2.34	58.5 \pm 2.50	NS
+3	61.7 \pm 2.98	62.8 \pm 3.57	59.5 \pm 2.35	59.8 \pm 2.31	NS
+5	61.7 \pm 2.55	61.4 \pm 2.83	60.7 \pm 1.23	60.9 \pm 2.01	NS
+7	61.7 \pm 1.84	61.8 \pm 1.87	61.0 \pm 1.29	61.1 \pm 1.44	NS
+14	66.4 \pm 2.42	66.0 \pm 2.33	64.5 \pm 0.97	66.5 \pm 1.03	NS
+21	67.3 \pm 2.93	65.6 \pm 2.69	67.4 \pm 1.60	69.7 \pm 1.05	NS
Mean	63.5 \pm 1.20	63.2 \pm 1.26	62.8 \pm 1.33	63.2 \pm 1.37	NS
Prepartum Mean	65.4 \pm 1.82	64.9 \pm 2.20	64.9 \pm 2.19	64.7 \pm 2.24	NS
Postpartum Mean	62.8 \pm 1.38	62.8 \pm 1.05	61.8 \pm 1.47	62.7 \pm 1.77	NS

Neutrophil profile

Mean neutrophil values were increased consequently from 21 day prepartum to day of calving and then after the day of calving to 3rd week of postpartum, there was a gradual fall in mean neutrophil levels among the entire groups. Mean

neutrophil level during prepartum period was lower than that of postpartum period among all the groups. There were significantly ($P < 0.05$) lower values of neutrophil were found on the day of calving in nanoZn₄₀ group compared to control, Zn₄₀ and nanoZn₂₀ (Table 6).

Table 6. Mean values (\pm SE) of neutrophil (%) in Sahiwal cows

Periods of observations	Mean neutrophil in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
Days relative to calving					
-21	23.0 \pm 0.38	22.9 \pm 0.65	23.1 \pm 0.71	23.5 \pm 0.79	NS
-14	24.8 \pm 1.00	24.7 \pm 0.45	24.3 \pm 1.03	24.0 \pm 0.99	NS
-7	30.3 \pm 1.31	28.8 \pm 1.62	29.3 \pm 1.04	29.1 \pm 0.49	NS
-5	32.7 \pm 0.77	33.4 \pm 1.29	30.9 \pm 0.52	31.3 \pm 1.24	NS
-3	33.6 \pm 0.52	33.1 \pm 0.51	31.6 \pm 1.17	33.6 \pm 1.76	NS
-1	34.6 \pm 0.48	34.7 \pm 0.70	33.3 \pm 1.54	34.3 \pm 1.37	NS
At calving	36.2 \pm 0.33 ^a	35.4 \pm 0.76 ^a	35.8 \pm 1.28 ^a	32.9 \pm 0.38 ^b	*
+1	34.9 \pm 0.77	35.8 \pm 0.89	34.6 \pm 2.03	33.9 \pm 1.59	NS
+3	34.4 \pm 1.09	33.4 \pm 1.47	30.9 \pm 0.52	30.8 \pm 1.21	NS
+5	33.5 \pm 0.85	33.4 \pm 1.27	31.9 \pm 1.26	31.7 \pm 1.59	NS
+7	33.7 \pm 0.52	32.2 \pm 0.46	31.5 \pm 1.07	31.3 \pm 2.1	NS
+14	27.7 \pm 0.64	29.9 \pm 0.71	26.6 \pm 1.20	27.9 \pm 1.06	NS
+21	24.7 \pm 1.29	25.6 \pm 0.59	24.9 \pm 2.18	27.0 \pm 2.52	NS
Mean	31.1 \pm 1.25	31.0 \pm 1.18	29.9 \pm 1.10	30.1 \pm 0.99	NS
Prepartum Mean	29.8 \pm 1.97	29.6 \pm 2.02	28.8 \pm 1.68	29.3 \pm 1.90	NS
Postpartum Mean	31.5 \pm 1.73	31.7 \pm 1.45	30.1 \pm 1.46	30.4 \pm 1.04	NS

^{a,b}- Mean with different superscript in a row differ significantly *($P < 0.05$)

Monocyte profile

Postpartum mean values of monocyte were higher than that of prepartum mean among all the

groups (Table 7). The significant (Pd^{**}0.05) higher level of monocyte was noticed at 21 days of postpartum period in nanoZn₂₀ group compared to nanoZn₄₀, Zn40 and control group.

Table 7. Mean values (±SE) of monocyte (%) in Sahiwal cows

Periods of observations Days relative to calving	Mean monocyte in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
-21	1.85 ± 0.34	1.97 ± 0.29	2.15 ± 0.34	2.18 ± 0.34	NS
-14	1.71 ± 0.31	1.86 ± 0.24	2.01 ± 0.30	2.12 ± 0.30	NS
-7	1.34 ± 0.30	1.74 ± 0.25	1.78 ± 0.26	1.53 ± 0.38	NS
-5	1.77 ± 0.29	1.88 ± 0.33	1.95 ± 0.33	1.64 ± 0.40	NS
-3	1.87 ± 0.39	2.26 ± 0.30	2.11 ± 0.30	2.64 ± 0.31	NS
-1	2.58 ± 0.26	2.52 ± 0.26	2.86 ± 0.38	2.92 ± 0.30	NS
At calving	2.72 ± 0.55	2.77 ± 0.33	3.00 ± 0.35	3.12 ± 0.29	NS
+1	2.44 ± 0.34	2.55 ± 0.30	2.68 ± 0.31	2.72 ± 0.24	NS
+3	2.31 ± 0.30	2.20 ± 0.38	2.60 ± 0.30	2.52 ± 0.24	NS
+5	2.30 ± 0.26	1.83 ± 0.34	2.56 ± 0.29	2.37 ± 0.26	NS
+7	2.25 ± 0.33	1.62 ± 0.31	2.51 ± 0.29	2.25 ± 0.34	NS
+14	1.95 ± 0.30	1.99 ± 0.30	2.37 ± 0.19	2.24 ± 0.30	NS
+21	2.00 ± 0.38 ^c	2.97 ± 0.29 ^{ab}	3.33 ± 0.26 ^a	2.08 ± 0.26 ^{bc}	*
Mean	2.08 ± 0.10	2.16 ± 0.11	2.45 ± 0.12	2.33 ± 0.12	NS
Prepartum Mean	1.87 ± 0.18	2.03 ± 0.11	2.14 ± 0.15	2.17 ± 0.22	NS
Postpartum Mean	2.20 ± 0.07	2.19 ± 0.20	2.67 ± 0.13	2.36 ± 0.09	NS

^{a,b,c}- Mean with different superscript in a row differ significantly *(P<0.05)

Eosinophil profile

Significant (Pd^{**}0.01) higher level of postpartum mean eosinophil was noticed in Zn₄₀ group. Postpartum mean eosinophil values were higher than

that of prepartum mean among all the groups exception with nanoZn₄₀ group. There were significant (P<0.05) variation (Table 8) in mean values of eosinophil which was highest in Zn₄₀ group.

Table 8. Mean values (±SE) of eosinophil (%) in Sahiwal cows

Periods of observations Days relative to calving	Mean eosinophil in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
-21	3.45 ± 0.32	2.85 ± 0.27	2.68 ± 0.25	2.98 ± 0.30	NS
-14	2.10 ± 0.33	2.35 ± 0.30	2.10 ± 0.35	2.62 ± 0.35	NS
-7	1.86 ± 0.30	2.51 ± 0.29	1.97 ± 0.36	2.09 ± 0.31	NS
-5	3.17 ± 0.66	2.55 ± 0.25	2.05 ± 0.30	2.25 ± 0.37	NS
-3	3.40 ± 0.94	3.06 ± 0.74	2.60 ± 0.35	2.61 ± 0.42	NS
-1	4.13 ± 0.90	3.79 ± 0.83	3.67 ± 0.71	3.83 ± 0.97	NS
At calving	2.67 ± 0.25	3.97 ± 0.83	4.00 ± 0.81	2.74 ± 0.30	NS
+1	3.67 ± 0.89	3.92 ± 0.94	3.57 ± 1.00	3.24 ± 0.80	NS
+3	3.53 ± 0.84	4.44 ± 1.21	3.63 ± 0.63	2.42 ± 0.30	NS
+5	3.67 ± 0.91	3.92 ± 0.94	2.59 ± 0.30	2.51 ± 0.29	NS
+7	3.87 ± 1.23	3.67 ± 0.81	3.35 ± 0.86	2.07 ± 0.38	NS
+14	3.42 ± 0.82	4.10 ± 1.23	2.55 ± 0.30	2.81 ± 0.35	NS
+21	4.64 ± 1.42	4.14 ± 1.07	2.98 ± 0.71	2.87 ± 0.87	NS
Mean	3.35 ± 0.21 ^{ab}	3.48 ± 0.19 ^a	2.90 ± 0.18 ^{bc}	2.69 ± 0.13 ^b	*
Prepartum Mean	3.01 ± 0.35	2.85 ± 0.21	2.51 ± 0.26	2.73 ± 0.25	NS
Postpartum Mean	3.80 ± 0.17 ^a	4.03 ± 0.10 ^a	3.11 ± 0.19 ^b	2.65 ± 0.16 ^b	**

^{a,b,c}- Mean with different superscript in a row differ significantly *(P<0.05) and **(P<0.01)

Basophil profile

Prepartum mean basophil values were higher than that of postpartum mean but the variation were found to be statistically non significant (Table 9). Overall,

values of mean basophil had not shown any pattern throughout transition period among the entire groups.

Table 9. Mean values (\pm SE) of basophil (%) in Sahiwal cows

Periods of observations Days relative to calving	Mean basophil in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
-21	0.96 \pm 0.11	1.17 \pm 0.10	0.79 \pm 0.10	0.95 \pm 0.11	NS
-14	0.81 \pm 0.16	0.91 \pm 0.17	0.73 \pm 0.12	0.90 \pm 0.14	NS
-7	0.78 \pm 0.14	0.80 \pm 0.11	0.73 \pm 0.06	0.78 \pm 0.13	NS
-5	0.64 \pm 0.13	0.88 \pm 0.10	0.78 \pm 0.10	0.76 \pm 0.10	NS
-3	0.71 \pm 0.15	0.72 \pm 0.0.14	0.75 \pm 0.16	0.79 \pm 0.14	NS
-1	0.90 \pm 0.17	0.65 \pm 0.09	0.67 \pm 0.10	0.69 \pm 0.10	NS
At calving	0.73 \pm 0.10	0.72 \pm 0.17	0.75 \pm 0.22	0.77 \pm 0.21	NS
+1	0.65 \pm 0.15	0.64 \pm 0.18	0.63 \pm 0.22	0.61 \pm 0.15	NS
+3	0.53 \pm 0.17	0.77 \pm 0.19	0.82 \pm 0.23	0.70 \pm 0.16	NS
+5	0.76 \pm 0.13	0.67 \pm 0.22	0.73 \pm 0.19	0.80 \pm 0.17	NS
+7	0.61 \pm 0.17	1.54 \pm 0.83	0.82 \pm 0.20	0.76 \pm 0.16	NS
+14	0.75 \pm 0.15	0.76 \pm 0.18	0.75 \pm 0.15	0.77 \pm 0.16	NS
+21	0.71 \pm 0.18	0.63 \pm 0.17	0.63 \pm 0.19	0.68 \pm 0.19	NS
Mean	1.14 \pm 0.40	0.83 \pm 0.07	0.73 \pm 0.01	0.76 \pm 0.02	NS
Prepartum Mean	0.8 \pm 0.04	0.85 \pm 0.07	0.74 \pm 0.01	0.81 \pm 0.03	NS
Postpartum Mean	0.66 \pm 0.03	0.83 \pm 0.14	0.73 \pm 0.03	0.72 \pm 0.02	NS

Our results are in agreement with Aswal et al. (2021), where, they observed increased level of neutrophil and monocyte and decreased level of lymphocyte was found on the day of calving suggesting a positive relationship between TLC and neutrophil and monocyte and a negative relationship with lymphocyte. There was non-significant change in an eosinophil and basophil. Increase in neutrophil count on the day of calving may be stimulated by cortisol release in the highly stressful condition was

reported by Kulberg et al. (2002).

Present findings are in accordance with Kumar et al. (2018) where, they observed that the levels of basophil, eosinophil and monocyte per cent were found similar among all groups, however, concentration of neutrophil was lower compared to control group in zinc methionine and zinc sulphate group. While, concentration of lymphocyte was higher in zinc methionine and zinc sulphate groups.

Body condition score (BCS)

Mean BCS during prepartum period were higher

than that of postpartum period among all groups (Table 10).

Table 10 Mean values (\pm SE) of Body condition score in Sahiwal cows

Periods of observations	Mean BCS in Sahiwal cows				Level of Significance
	Control	Zn ₄₀	nanoZn ₂₀	nanoZn ₄₀	
Days relative to calving					
-21	3.29 \pm 0.19	3.20 \pm 0.10	3.25 \pm 0.25	3.33 \pm 0.13	NS
-14	3.33 \pm 0.22	3.37 \pm 0.24	3.41 \pm 0.10	3.45 \pm 0.19	NS
-7	3.41 \pm 0.10	3.45 \pm 0.13	3.58 \pm 0.16	3.54 \pm 0.15	NS
-5	3.45 \pm 0.17	3.45 \pm 0.19	3.58 \pm 0.20	3.54 \pm 0.19	NS
-3	3.45 \pm 0.10	3.50 \pm 0.21	3.62 \pm 0.12	3.58 \pm 0.17	NS
-1	3.5 \pm 0.17	3.58 \pm 0.25	3.70 \pm 0.31	3.62 \pm 0.19	NS
At calving	3.62 \pm 0.10	3.70 \pm 0.19	3.75 \pm 0.26	3.66 \pm 0.15	NS
+1	3.20 \pm 0.17	3.33 \pm 0.13	3.33 \pm 0.05	3.29 \pm 0.11	NS
+3	3.16 \pm 0.17	3.20 \pm 0.11	3.29 \pm 0.16	3.25 \pm 0.15	NS
+5	3.04 \pm 0.11	3.16 \pm 0.15	3.16 \pm 0.10	3.12 \pm 0.12	NS
+7	3.04 \pm 0.07	3.12 \pm 0.14	3.16 \pm 0.08	3.12 \pm 0.08	NS
+14	2.83 \pm 0.08	2.87 \pm 0.10	2.95 \pm 0.10	2.91 \pm 0.12	NS
+21	2.66 \pm 0.10	2.70 \pm 0.07	2.79 \pm 0.11	2.75 \pm 0.11	NS
Mean	3.22 \pm 0.07	3.27 \pm 0.07	3.35 \pm 0.08	3.32 \pm 0.07	NS
Prepartum Mean	3.40 \pm 0.03	3.42 \pm 0.05	3.52 \pm 0.06	3.46 \pm 0.04	NS
Postpartum Mean	2.98 \pm 0.08	3.06 \pm 0.09	3.11 \pm 0.08	3.07 \pm 0.08	NS

The BCS increased linearly over the time till the day of calving then gradually decreased with the progress of lactation. Control group showed the maximum reduction in BCS from 21 days prepartum to 21 days postpartum. This drop in BCS is usually associated with negative energy balance which normally occurs after parturition. These results are in agreement with the findings of Chandra et al. (2013), where animals exhibited significantly ($P < 0.05$) higher decline in body condition score during prepartum and postpartum period of control group, which might be due to tissue mobilization is usually most active from 14 days prepartum to 35 days postpartum. A similar trend in changes in values of BCS was also observed by Maurya (2011) in transition dairy cows. However, Cope et al. (2009) reported that there was no effect of either level or form of dietary Zn (ZnO or chelated Zn) on body condition score in dairy cows

CONCLUSION

Transition period is the utmost stressful condition in the life of a dairy animal. Therefore evaluation of hematological parameters and body condition score is of eminent importance to find out the physiological

changes and status of health of animals. The results of present study revealed that animals supplemented with 20 ppm of nanoZn showed better performance compared to animals of other groups. A significant increase in TEC, Hb and PCV and significant decrease in eosinophil in nanoZn₂₀ during prepartum compared to postpartum period. Better body condition score of Sahiwal cow was found in nanoZn₂₀ group. There were noticeable changes in hematological parameters and body condition score in Sahiwal cows during transition period. This information may be of great use for better management of dairy cows during transition period.

ACKNOWLEDGMENT

Authors are thankful to Dean, College of Veterinary Science and Animal Husbandry, Anjora, Durg for providing the facilities for conducting this research work and are also sincerely acknowledge to cattle keepers in BMEF, Anjora for their kind participation in the study.

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