



Praveen *et al.*

Assessment of Milk Yield and Milk Quality on Boron Supplemented Groups in Crossbred Karanfries (Holstein Friesian X Tharparker) Cows During Hot Humid Season

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ABSTRACT

Present study was done on Crossbred Karan Fries (Holstein Friesian X Tharparker)cows (n=18) to evaluate the effect of boron supplementation on milk yield, milk composition and somatic cell count of milk during heat stress, cows were allocated to three treatments, viz, group 1 (n=6), group 2 (n=6) and group 3 (n=6) based on bodyweight, parity and estimated producing ability. All cows received basal feed (roughage and concentrate) based on ICAR feeding standards. Additionally, in group 2 and group 3, boron was supplemented at the rate of 250 ppm and 500 ppm, respectively. The plasma boron was estimated to be significantly higher in boron supplemented groups than control. The milk yield, milk Fat% and milk composition on weight basis (kg) has no significant improvement but other milk composition (viz, milk SNF, milk protein, milk lactose and milk total solids %) did differ significantly between the control and boron supplemented groups. Somatic cell count of boron supplemented groups was significantly low ($p<0.05$) than control, but there was no significant relationship between treatment groups. It is concluded that supplementing boron in diet improved the milk composition (%) and reduced the SCC of milk but no considerable improvement in milk yield was observed.

Key words: Boron, Heat stress, Somatic cell count, Milk yield

INTRODUCTION

The bovine population of India is about 302.79 million according to 20th livestock census (2019) and out of that about 51.36 million animals were crossbred. Exotic cattle with average milk production of about 8.09 kg per day which contribute 28% of total milk produced in India DADH (2021). In dairy cattle when the THI exceeds more than 72, heat stress condition arises, where high producing dairy cattle are more vulnerable to heat stress (Zimbelman *et al.*, 2010). Temperature humidity index (THI) is the combined measure of effect of ambient heat and relative humidity which indicates degree of heat stress experienced by dairy cows (Berman, 2005). Prolonged period of high environmental temperature with high relative humidity reduces milk yield due to reduced dry matter intake (Kadzere *et al.*, 2002) about 0.2 kg per unit increase in THI above 72 (West, 2003). Pragna *et al.* (2017) stated that high temperature and humidity reduces feed intake which leads to reduced

reproduction potential and in turn reduces milk yield where high yielding cows are mostly susceptible. Schwartz *et al.* (2009) enacted a significant decrease ($P<0.01$) in Dry matter intake in lactating dairy cows during heat stress condition progressively. Bouraoui *et al.* (2002) observed that with the increase of THI value from 68 to 78 % reduces DMI by 9.6 % and also milk yield by 21 %. Aggarwal and Singh (2006) found an increase in THI from 68 to 78 leads to reduced milk production by 21 % and DMI by 9.6 % and also stated that for each unit increase in THI above 69, 0.41 kg milk production per day per cow is decreased. Increased body temperature during heat stress affects the fat synthesis i.e., milk quality is affected. Increase in somatic cell count during heat stress condition of Holstein cows observed (Smith *et al.*, 2013). McDowell *et al.* (1976) showed a decrease of 39.7, 18.9 and 16.9% of milk fat, solids-not-fat, and milk protein percentages respectively with the air temperature from 18 to 30 in lactating Holstein cows.

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Boron is identified as emergency trace mineral and also as an essential mineral in animals (Nielsen, 1994). Boron supplementation increases lambs' cell-mediated and humoral immunity (Bhasker *et al.*, 2017) and also have potential to increase propionate concentration (Bosaglu *et al.*, 2017) and reduce the incidence of ketosis and negative energy balance by reducing NEFA and BHBA (Kabu and Civelek (2012). Therefore, the present study was undertaken to determine the effects of supplemental boron on milk yield and milk quality of crossbred Karanfries cows.

MATERIALS AND METHODS

Animal care and conduction of this experiment was approved by the Institutional Animal Ethics Committee (IAEC No. 44-IAEC-19-24) as per the article number 13 of the CPCSEA- rules, laid down by the Government of India.

Present study was conducted at Livestock Research Centre of National Dairy Research Institute (N.D.R.I.), Karnal, Haryana, India. NDRI, Karnal is situated at 29° 43' N latitude and 76° 58' E longitudes at an altitude of 245 meters above the mean Sea level. The minimum ambient temperature falls to near freezing point in winter and maximum goes approximately up to 45°C in summer. The average annual rainfall is close to 700 mm, most of which is received from July to September.

For the present study, 18 pregnant peri-parturient Karan Fries (Holstein Friesian X Tharparker) cows were selected from Livestock Research Centre of NDRI, Karnal and divided into three groups of each six animals on the basis of Expected Production Ability (EPA), parity and body weight (Table 1). It was made sure that the animals used for research were devoid of any anatomical, physiological, or viral diseases.

Table 1. Details of experimental animals (Mean $\pm$ S.E)

Groups	Body weight	Parity	EPA
1	447.5 $\pm$ 24.39	2.25	3514.50 $\pm$ 159.00
2	418.16 $\pm$ 22.01	2.00	3524.66 $\pm$ 84.21
3	458.66 $\pm$ 41.02	2.66	3644.33 $\pm$ 384.44

The experimental animals were kept in separate pens during the course of experiment. Experimental animals were maintained as per the standard feeding and management followed at NDRI. All the animals were fed a ration consisting of concentrate mixture and roughage maize. The concentrate mixture consists of maize, oats, wheat bran, GNC, mustard cake, cotton seed cake, gram chunai, rice bran, common salt, mineral mixture and sodium bicarbonate was offered in morning with 20 % crude protein and 70 % total digestible nutrient and the chaffed green fodder (maize) was offered at 10.00 AM. Free access to fresh tap water was made all the times to the animals.

The study was conducted out from month of July to October at the livestock research centre of ICAR-National Dairy Research Institute, Karnal, Haryana (India). Meteorological variables in terms of dry and wet bulb temperatures and relative humidity were recorded throughout the experiment during morning and evening hours and temperature humidity index

was calculated by the formula,

$THI = 0.72 (T_{db} + T_{wb}) + 40.6$, Where, T_{db} = dry bulb temperature ($^{\circ}C$), T_{wb} = wet bulb temperature ($^{\circ}C$)

The respective quantities of Boron were weighed accurately based in dry matter intake at the dose of 250 ppm and 500 ppm for two treatment groups and mixed with small quantity of concentrate to be fed to the animals. Boron was supplemented daily as a form of Boric Acid (food grade) 30 days before and 60 days after calving. Digestion of plasma samples was carried out in microwave digester (multiwave 3000, Anton Parr) using polypropylene vessels to avoid possible contamination with boron from borosilicate tubes. One mL of Plasma was added to vessel along with 1 mL of hydrochloric acid (HCl) and 5 mL of nitric acid (HNO₃) and digested in closed vessel microwave. After cooling, sample volume was increased to 25 mL using distilled water. Concentration of Boron in the digested plasma

samples was determined via Inductively Coupled Argon Plasma Optical Emission Spectrophotometer (Optima 9000, multitype, Shimadzu, Japan).

Milk samples were collected at weekly intervals in 50 ml sterilized milk sampling bottles from individual animal of each milking for analysis of Somatic Cell Count (SCC) and milk composition. First few strips of milk samples were discarded as it contains higher amount of microbial population and the subsequent stepping from each quarter was collected and pooled to get 50 ml for analysis.

Milk composition viz milk fat (%), milk SNF (%), milk protein (%), milk lactose (%) and total solids was estimated by lactoscan milk analyser. Somatic cell count in milk was estimated by

ekomilk scan machine. 10 ml of milk and 5 ml ekoprime solution was added in flask of ekomilk scan machine. After few minutes of adding sample, results were noted. The results were noted down in ($\times 10^5$ cells/ml).

Statistical analysis was done by one way ANOVA using statistical package for the Social Sciences (SPSS for windows, V21.0; SPSS Inc., Chicago, USA) The data reported as Mean $\pm$ SE, and differences were regarded as significant at 5% significant level.

RESULTS AND DISCUSSION

The values of THI at fortnight interval were recorded and given in the table 2.

Table 2. Mean fortnightly temperature humidity index (THI) during the experimental period

Month	Fortnights	THI _{min}	THI _{max}	THI _{avg}
July	I	76.02	83.22	79.62
	II	79.14	82.67	80.90
August	III	80.08	84.15	82.06
	IV	79.58	83.37	81.48
September	V	79.79	85.00	82.40
	VI	76.36	82.25	79.30
October	VII	70.68	80.46	75.57
	VIII	66.83	79.09	72.96

Alhussien and Dang (2017) enacted that THI is the most common indicator to assess heat stress on growth of dairy heifers which is due to combined effect of environmental temperature and relative humidity. THI is common method of analysis for heat stress assessment and value of less than 72 indicate no heat stress, value ranging from 72 to 76 indicate moderate heat stress, 76 to 80 denote severe heat stress and the value more than 82 indicate deadly heat stress (NRC, 1971). The experiment was conducted during July to October and THI calculated during those periods indicated heat stress condition to animals. Kumar *et al.* (2018) stated that the major problem for dairy farmers causing economic loss to animal is heat stress which in most severe form can cause death of animal and also Thatcher *et al.* (2010) observed that when the THI is above 72 and 78, it was stressful and severe stress in dairy cows.

The threshold level of maximum THI for indigenous and crossbred cattle were 75.12 and 73.67 respectively (Choudary, 2017). However, crossbred cattle showed heat stress that affects growth, production and reproduction negatively at THI of 72 and above (Zewdu *et al.*, 2014).

The plasma boron levels of the experimental animals were analysed by digestion followed by plasma optical emission spectrophotometer to evaluate the plasma boron levels in the blood following supplementation. It was found that plasma boron values significantly ($p < 0.001$) high in 250 ppm and 500 ppm supplemented groups (Group 2 and 3) as compared (Group 1) to control (only with basal diet) with the Mean $\pm$ SE values of control, T1 and T2 groups were 0.57 ± 0.007 , 3.21 ± 0.04 and 3.87 ± 0.02 respectively denoted in figure 1.

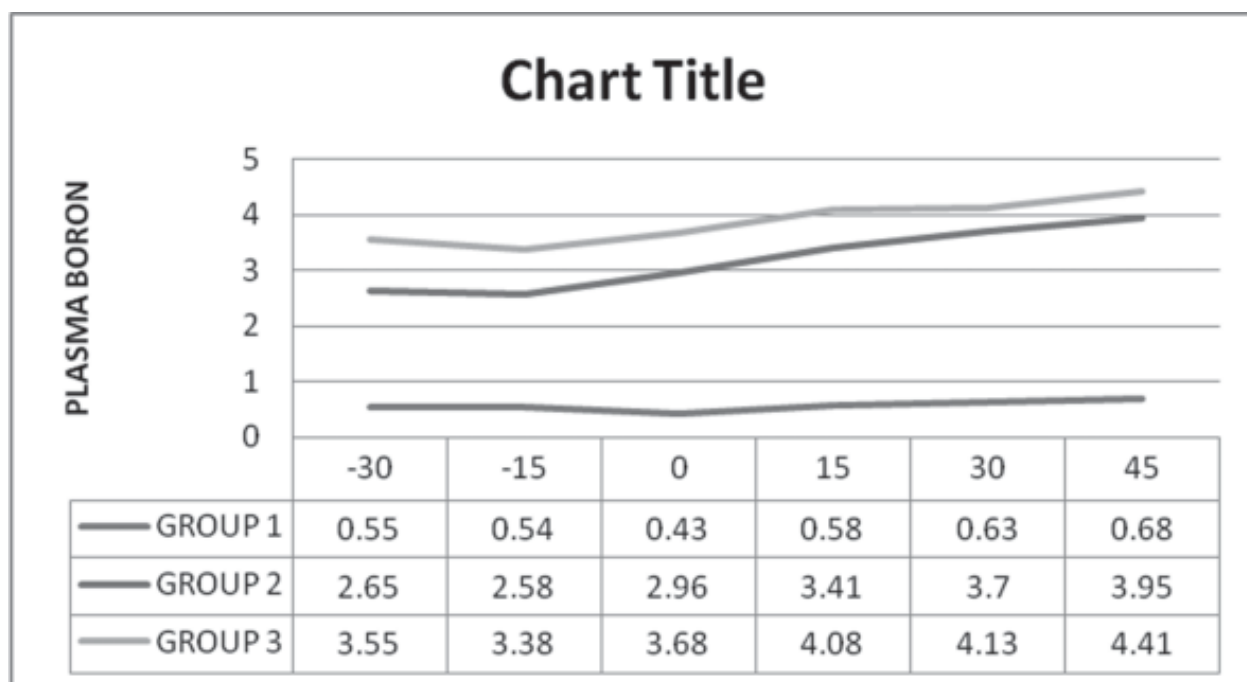


Fig. 1. Average of plasma boron concentration in experimental group of animals

Similar results were reported by Sharma *et al.* (2020), that significantly ($P < 0.05$) higher levels of plasma boron following supplementation of 200 ppm and 400 ppm in the total mixed ration of buffalo diet. Naghii *et al.* (2011) observed that plasma boron concentration increased significantly ($P = 0.002$) within hours after consumption compared to the placebo. Following oral boron supplementation of 250 and 500 ppm boron in the form of boric acid, there was a greater amount of boron in the blood.

seems to be affected the milk composition percentage and milk somatic cell count positively.

Milk yield of animals were recorded daily for each animal among the groups upto 90 days postpartum. The overall Mean ($\pm$ S.E.) milk yield (Liter/day) were 12.02 ± 0.22 , 12.65 ± 0.25 and 12.79 ± 0.25 in Control, T1 (B250) and T2 (B500) groups respectively showing no significant improvement in treatment groups in compared to control (figure 2).

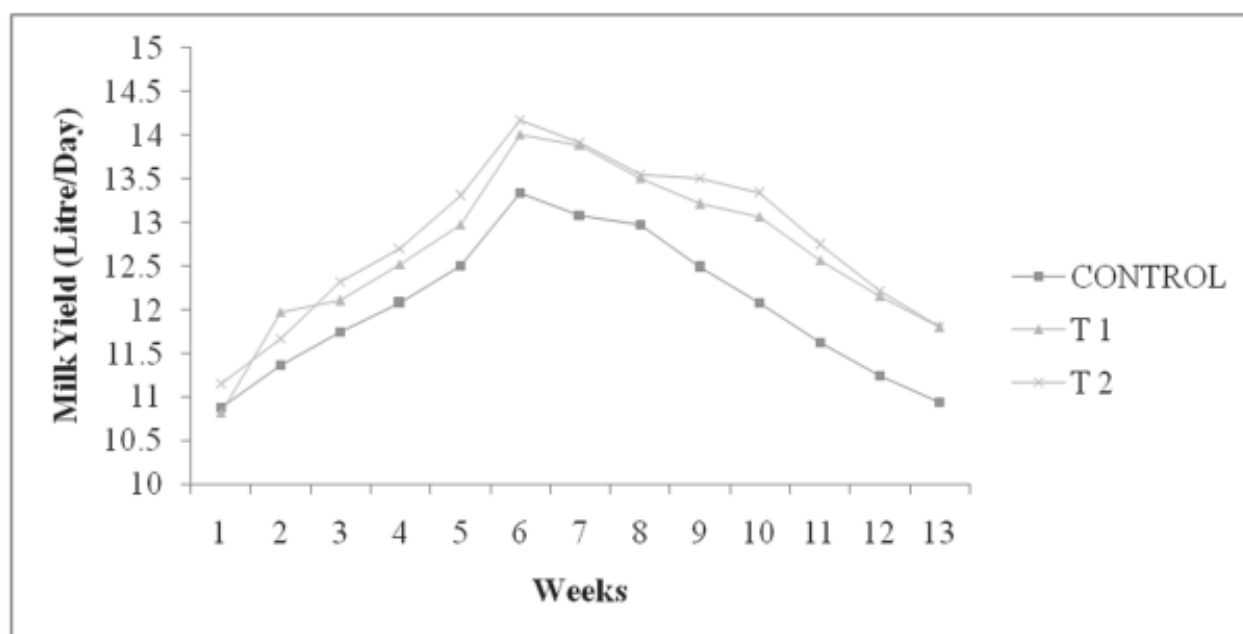


Fig. 2. Average weekly milk yield (litre/day) of experimental groups

The dry period is a critical time for mammary cell turnover in dairy cows. In this period, mammary cells have extensive growth and cell turnover to prepare for the next lactation (Annen *et al.*, 2004). One of the major economic losses resulting from heat stress in dairy industries is a decreased milk yield. Habeeb *et al.* (2014) observed significantly lower DMI at summer compared to winter and the percentage decrease was about 21.96% in purebred and was 24.09% in crossbred calves. Basoglu *et al.* (2017) also observed no change in DMI in Holstein dairy cows supplemented with boron as Borax in proportion of 567, 1134 and 1071 ppm over control groups. The major reason for reduced milk yield in dairy cows was decreased feed intake during heat stress. OACC, 2014 finding is that a drop of 10 % milk production observed at the temperature and RH of about 27-32 °C and 50-90 % respectively and drop of 25 % observed at 32-38 °C and 50-90 % respectively especially in high producers. Singh *et al.* (2019) observed that in Karan Fries cows, from THI of 72.2-91.0 there will be reduction in 131.11 ml of milk per unit rise in THI. Smith *et al.* (2013) observed when THI > 72, Milk yield from 34.8 to 32.9 kg/day in moderate and further to 30.4 kg/day in severe heat stress condition significantly ($P < 0.01$).

In our experiment, peak milk yield for all the groups were found to be at 6th week of lactation

and after that reduced reduction in milk yield were noted. Supplementation of Boron has no significant effect on milk yield of different groups of animals which are in line with previous researchers who found no improvement in milk yield by B supplementation (Kabu and Uyarlar 2015; Basoglu *et al.*, 2017). Also, by Owen (1994) that B supplementation at 620 mg boron/100 kg bodyweight per day had no effect on production performance of animals.

Table 3, represents the effect of boron supplementation on milk composition. Results showed that, Milk fat (%) ranges from 3.90 to 4.31 in Control; 3.98 to 4.48 in T-1 and 3.92 to 4.52 in T-2 groups and Milk SNF (%) ranges from 8.13 to 8.50 in Control; 8.48 to 8.86 in T-1 and 8.22 to 8.80 in T-2 groups. Milk protein (%) ranges from 3.10 to 3.23 in Control; 3.28 to 3.38 in T-1 and 3.20 to 3.38 in T-2 groups. Milk Lactose (%) ranges from 4.31 to 4.43 in Control; 4.42 to 4.46 in T-1 and 4.40 to 4.56 in T-2 groups. Total Solids (%) ranges from 12.16 to 12.81 in Control; 12.58 to 13.34 in T-1 and 12.44 to 13.40 in T-2 groups. from the findings shown that there is significant increase in overall average milk SNF %, protein %, lactose % and total solids % but no significance was observed in Milk fat % and milk composition on weight basis of boron supplemented groups.

Table 3. Mean ($\pm$ S.E.) of milk composition of different treatment groups

Milk parameter	B0 Group 1	B250 Group 2	B500 Group 3	Significance
DMI (kg/100kg bwt)	10.98 $\pm$ 0.63	11.23 $\pm$ 0.67	11.56 $\pm$ 0.62	0.495
4% FCM	12.48 $\pm$ 3.65	13.15 $\pm$ 2.81	13.36 $\pm$ 1.67	0.312
Milk fat %	4.12 $\pm$ 0.19	4.18 $\pm$ 0.12	4.20 $\pm$ 0.15	0.372
Milk SNF %	8.25 $\pm$ 0.11 ^a	8.65 $\pm$ 0.25 ^b	8.47 $\pm$ 0.09 ^b	0.001
Milk protein %	3.18 $\pm$ 0.06 ^a	3.32 $\pm$ 0.09 ^b	3.30 $\pm$ 0.04 ^b	0.000
Milk lactose %	4.37 $\pm$ 0.07 ^a	4.49 $\pm$ 0.11 ^b	4.50 $\pm$ 0.05 ^b	0.011
Total solids %	12.42 $\pm$ 0.23 ^a	12.91 $\pm$ 0.27 ^b	12.83 $\pm$ 0.16 ^b	0.002
Milk fat (kg)	0.50 $\pm$ 0.07	0.53 $\pm$ 0.12	0.54 $\pm$ 0.15	0.309
Milk SNF (kg)	1.00 $\pm$ 0.18	1.10 $\pm$ 0.10	1.09 $\pm$ 0.12	0.054
Milk protein (kg)	0.38 $\pm$ 0.03	0.42 $\pm$ 0.03	0.42 $\pm$ 0.10	0.060
Milk lactose (kg)	0.53 $\pm$ 0.03	0.57 $\pm$ 0.05	0.57 $\pm$ 0.06	0.114
Total solids (kg)	1.52 $\pm$ 0.13	1.66 $\pm$ 0.15	1.63 $\pm$ 0.43	0.154

Our results were similar with Bosaglu *et al.* (2017) observed no change in milk fat % compared to treatment groups but contradictory to his findings that non significant in Milk Protein % in periparturient Holstein dairy cows supplemented with boron as Borax in proportion of 567, 1134 and 1701 ppm was over control groups. But there were not traceable research findings to support the results of boron supplementation on SNF and Lactose percentage and weight in kgs. Pragna *et al.* (2017) stated that high temperature and humidity reduces feed intake which leads to reduced reproduction potential and in turn reduces milk yield where high yielding cows are mostly susceptible. Increased body temperature during heat stress affects the fat synthesis i.e., milk quality is affected. Smith *et al.* (2013) observed Milk protein percentage decreased in moderate HS from 3.2 to 3.1% ($P < 0.05$) in Holstein cows. Spiers *et al.* (2004) stated that the reduced milk fat, milk protein, milk solids and lactose percentage during

heat stress is due to elevated core body temperature of animals.

Bosaglu *et al.* (2017) found that boron helps in energy balance and ketogenesis of postpartum milking cows, he also observed that propionate and postpartum lactate concentration was higher than control groups over boron supplemented groups. Intraruminal infusion of propionate increased milk lactose % and reduced milk fat % (Fisher and Eliot, 1966) and milk protein % (Rook and Balch, 1961). High propionate induced improvement in milk composition in aspect of Protein %, lactose % that in turn showed significant improvement of SNF% and total solids %.

Somatic cell count was estimated upto 9th week of lactation. The overall Mean ($\pm$ S.E.) somatic cell count ($\times 10^5$ cells/mL) were 3.73 ± 0.43 , 1.67 ± 0.58 and 1.34 ± 0.34 in control, T1 and T2 groups respectively showing significant ($p < 0.01$) decrease in treatment groups in comparison to control (Table 4).

Table 4. weekly Mean ($\pm$ S.E.) milk somatic cell (SCC) ($\times 10^5$ cells/mL) of different groups of KF cows

WEEKS Group 1	B0 Group 2	B250 Group 3	B500
1	4.07 ± 0.65	2.32 ± 0.65	2.28 ± 0.78
2	4.01 ± 0.34^b	1.81 ± 0.64^a	1.55 ± 0.37^a
3	4.58 ± 0.73^b	2.05 ± 0.86^a	1.08 ± 0.19^a
4	3.73 ± 0.44^b	1.53 ± 0.45^a	1.33 ± 0.47^a
5	3.44 ± 0.30^b	1.78 ± 0.77^a	1.30 ± 0.32^a
6	4.27 ± 0.65^b	2.00 ± 0.87^a	1.39 ± 0.43^a
7	3.62 ± 0.40^b	1.64 ± 0.59^a	0.97 ± 0.07^a
8	3.12 ± 0.19^b	1.14 ± 0.28^a	1.28 ± 0.36^a
9	2.71 ± 0.20^b	0.77 ± 0.07^a	0.85 ± 0.08^a

^{a,b} Means having different superscripts within a row differ significantly ($p < 0.05$)

Milk somatic cell count was reduced significantly by 55.22 % and 64.07 % in 250 ppm and 500 ppm boron supplemented groups, respectively compared to control. Reduced trend of SCC count was observed in all groups of animals from 1st week postpartum upto 9th week postpartum. Somatic cells in the milk are primarily the leucocytes and also the shredded epithelial cells from the lining of the epithelial cells. Paape *et al.* (2002) and Burvenich *et al.* (2007) stated that epithelial cells shedding from the udder and

leukocytes as a defence mechanism of infection or injury forms the Milk Somatic Cells. Harmon (1994) stated that milk Somatic cell counts (SCC) is a universal indicator of milk quality and mammary gland health. Smith *et al.* (2013) observed increase in Somatic cell score in milk during heat stress condition of Holstein cows but reduction in SCS from 4.18 to 3.84 during mild to severe heat stress. Alhussien and Dang (2017) found increased significantly ($P < 0.05$) SCC during summer months in comparison with Thermo neutral

zone and Winter season and also increased SCC is pronounced in multifarious (above 4th parity) than first lactating animal.

Supplementation of Boron has effectively reduced somatic cell count in 250 ppm and 500 ppm of Boron supplemented groups of animals over control which may be due to immune-modulatory action that is cell mediated and humoral immunity (Bhasker *et al.*, 2017) and antioxidant effect of boron that decreased the somatic cell in milk. We could not compare the results due to unavailability of literature where supplemental Boron has been used in relation to somatic cell count of cattle.

CONCLUSION

During high THI during summer season, the somatic cell count was calculated at weekly interval upto 9 weeks postpartum have found significantly reduced SCC of milk in boron supplemented group than control groups and milk composition percentage but no significant effect on milk yield and milk composition. The reduced somatic cell counts due to immunomodulatory defence mechanism of boron on body which is compromised during heat stress condition due to elevated cortisol level and hormonal imbalances. Boron supplemented groups at the dose of 250 ppm and 500 ppm makes the transition cows adaptable to heat stress by improving the somatic cell count and thus supplementation of boron helps in improving the keeping quality of milk through reducing SCC. These findings will be helpful in developing suitable feeding strategies to combat the harsh environmental climatic condition and its associated stress to improve health and productivity of crossbred animals under the changing climatic scenario.

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