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Effect of supplementation of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 probiotic cultures on physiological responses in Murrah buffalo calves

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Abstract
The present experiment was carried out from November 2017 to April 2018 i.e. during the winter and summer season at ICAR-National Dairy Research Institute, Karnal (Haryana) to investigate the effect of supplementation of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 probiotic cultures on physiological responses in Murrah buffalo calves. The new born Murrah buffalo calves were divided into four groups viz., T_1, T_2, T_3, and T_4. The calves of control group (T_1) were fed according to ICAR, 2013 feeding standard. Calves of T_2 were fed according to control group (T_1) except *Lactobacillus plantarum* CRD 2 supplementation 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day). The calves T_3 were fed according to control group (T_1) except *Lactobacillus rhamnosus* CRD 9 supplementation 10^8 cfu/ml (As a fermented milk @ 100 ml/calf/day), also the calves of T_4 fed according to control group (T_1) except combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8 cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) supplementation. During the winter season the overall mean± SE values of maximum and minimum temperature were 21.49±0.88 and 08.18±0.85 respectively while during the summer season the overall mean± SE values of maximum and minimum temperature were 30.22±1.74 and 13.45±1.38 respectively. The mean temperature-humidity index (THI) at morning and afternoon sessions during the winter season were 58.49±1.43 and 60.54±3.45 respectively while 75.07±1.72 and 59.20±1.29 respectively during the summer seasons. There were no significant ($P<0.05$) differences observed in rectal temperature, respiration rate and pulse rate between different treatment groups at the morning and afternoon session during the winter as well as summer season also during the pre-ruminant as well as post-ruminant phase while the rectal temperature, respiration and pulse rate in buffalo calves at afternoon session was slightly higher in all groups during the winter as well as summer season. There were also no significant ($P<0.05$) differences observed in rectal temperature, respiration rate and pulse rate between different treatment groups of female and male buffalo calves in the morning as well as afternoon session. The rectal temperature and respiration rate of treatment groups in the morning and at afternoon session in the both sexes was slightly lower than control group. The all observation were falls in the normal physiological range so these findings indicated that probiotic supplementation had no adverse effect on physiology of calves resulted in normal physiological responses in buffalo calves, so probiotic supplementation advantageous in the removal of stress in the young buffalo calves during the stress period which are more prone to cold as well as heat stress.

Keywords: Physiological responses, summer, winter, pre-ruminant, post-ruminant, female

Introduction
Dairying has been prominent supplementary enterprise and regular source of income to the farmers (Mahida *et al.*, 2018) [6], with respect to dairy farming or dairy business is concern buffaloes play an important role in the livelihood of millions of small and marginal farmers in the country. It contributes 76.19 million tonnes of milk (49%) and 1.61 million tonnes of meat (23%) annually to Indian food (DAHDF, 2016) [4]. Physiological responses or vital body sign i.e. rectal temperature, pulse rate and respiration rate in dairy calves as important clinical indicators for monitoring health status of the dairy calves and which form the important basis for the monitoring effect of undergoing treatment in various diseases. As far as dairy farming or dairy business is concern calf health is a very critical factor affecting the welfare and economics of young stock in dairy enterprise. The productivity of buffalo is mainly influenced by the care and management during young stage. Young calves being the foundation of replacement stock can be an important asset for the farmer to generate income. Hence, healthy young stock is indispensable for a successful and profitable dairy enterprise.

But, buffalo calves health is a huge setback limiting the achievement of the dairy industry and farmers. Physiological responses helps in the understanding of the illness exhibited by the change in the norms of rectal temperature, respiration rate and pulse rate and which ultimately helps in the diagnosis of certain diseases and treatment effect. Deviation from the normal physiological parameters considered as discomfort to the animals. However, numerous research studies have been conducted to investigate the health benefit and positive effect of probiotics supplementation on physiological responses in dairy calves. The present experiment was carried out from September 2017 to June 2018 at ICAR-National Dairy Research Institute, Karnal (Haryana). The institute is situated at 29°42"20N Latitude and 76°58"52.5˙ E Longitude, at an altitude of 227 m above mean sea level. The maximum and minimum tempratue in summer and winter goes up to 450C and 4-50C. The extreme climatic conditions are responsible for imbalance in thermoregulatory and homeostasis mechanisms in calves resulted in poor growth and development, poor performance and poor health. Heat or cold stress results in direct economic losses because of increased calf mortality and morbidity, as well as indirect costs caused by reduced weight gain, performance, and long-term survival (Roland *et al.*, 2016) [12]. Extreme heat stress can damage the gastrointestinal mucosa, which protects the internal environment of the body from bacteria and bacterial endotoxins (lipopolysaccharides), dysfunction of this protective barrier results in increased intestinal permeability and diffusion of toxic bacterial components from the gut lumen to the blood and for that gut microbiota play key role in keeping mucosal barrier function (Sorokulova *et al.*, 2015) [13]. Thermic stress is observed in association with extreme temperatures and large temperature variations, but other variables such as relative humidity and wind speed can also contribute to thermic stress (Roland *et al*., 2016) [12]. By modulating the intestinal microbiota one might offer a novel and non-invasive therapeutic approach for promoting host well-being by prevention and treatment of the adverse effects of stress in the gut (Sorokulova *et al.*, 2015) [13]. Therefore, the present experiment aimed to investigate the effect of supplementation of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 probiotic cultures on physiological responses in pre and post ruminant Murrah buffalo calves.

Materials and Methods

Calves Resources and Housing Management

The new born Murrah buffalo calves of 5-7 days old, just after colostrum feeding period was finished were taken from Livestock Research Centre (LRC), NDRI, Karnal. The calves were divided into four groups with ten calves in each group. The calves were housed individually in well-ventilated pens. All the pens were of similar size. The housing condition for all calves was similar during entire period of experiment. The floor of the shed was pukka with cement concrete. In winter, to avoid excessive cold and inclement weather condition, sufficient precautionary measures were taken. The pens were cleaned and washed daily and also after milk and probiotic feeding and all necessary care taken throughout the experimental period. The calves were daily brushed in the morning hours to remove faeces and dirt etc. from the body and also to stimulate blood circulation. The calf pens were maintained under proper hygienic condition. The faeces were removed from both indoor and outdoor housing two times daily. All the calves were exposed to regular contact with the animal attendant and researchers to ensure that they become adapted to the experimental conditions. All experimental calves during the Pre as well as Post ruminant phase were gone through daily routine health checkup with utmost care throughout the experimental period to diagnose diseases and other problems so as to follow proper treatment. Deworming of experimental calves was followed as per standard procedure of NDRI, Karnal. The first deworming was done on 15 days and then subsequently 30, 60, 90, 120, 150, 180 day of experiment by Albbendazole 5 mg/kg and Piperazine 200 mg/kg.

Feeding management of experimental calves, treatment details and Ethical permission

The calves of T_1 (control) were provided green fodder like maize and sorghum and concentrate mixture containing maize grain, bajra grain, groundnut cake, deoiled mustard oilseed cake (DMOC), soyabean meal (SBM), deoiled rice bran (DORB) and wheat bran during entire experiment period. Roughage and concentrate were divided into two halves and offered at morning and evening in group. The feeding of milk was carried out twice a day. Whole milk fed to the calves @ $1/10^{th}$ of BW up to the four weeks, $1/15^{th}$ of BW during four to eight weeks and $1/25^{th}$ of BW during the eight to ten weeks. Calves of T_2 were fed according to control group (T_1) except *Lactobacillus plantarum* CRD 2 supplementation 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day). The calves T_3 were fed according to control group (T_1) except *Lactobacillus rhamnosus* CRD 9 supplementation 10^8 cfu/ml (As a fermented milk @ 100 ml/calf/day), also the calves of T_4 fed according to control group (T_1) except combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8 cfu/ml [1:1 Ratio] (As a fermented milk @ 100 mL/calf/day) supplementation. Experiment was approved by the institutional animal ethics committee (IAEC) constituted as per the article no.13 of the CPCSEA rules, laid down by Govt. of India.

Climatic Conditions and Measurement of Temperature Humidity Index (THI)

Daily Weather Phenomenon data were adapted from the recording made at Central Soil and Salinity Research Institute (C.S.S.R.I.), Karnal, Haryana which was situated at 29°43'N Latitude and 76°58'E Longitude, at an altitude of 245 Mtrs. above mean sea level and located 5 km away from the experimental site. The weather phenomenon recorded daily at 7.22 Hrs. IST and 14.22 Hrs. IST. Using the climatic data Thermal comfort level of an animal environment was measured by calculating ambient THI using the following formula (McDowell, 1972) [7] in the whole experimental period and represented as fortnightly average.

THI=0.72 (wet bulb temperature + dry bulb temperature) + 40.6

Physiological Responses

Physiological responses were recorded during the summer as well as winter season and also recorded for pre-ruminant, post-ruminant calves and for different sexes i.e. for male and female calves in the morning and afternoon at fortnight interval. The rectal temperatures (°C) of experimental calves were recorded by using a digital thermometer by inserting 3 inches in the rectum so that it remained in contact with rectal mucus membrane for at least 2 minutes. The observation was recorded in degree Celsius. Respiration rate of experimental calves were recorded by observing flank movement for one

minute in which each inward and outward movement of the flank was counted as one complete respiration. The respiration rate was recorded as breaths per minute (bpm). The Pulse rate was measured by indirect auscultation of heart through stethoscope. Auscultation site for the heart was on fourth and fifth intercostal space on left side. The systolic and diastolic sounds were together counted as one heartbeat. The pulse rate was expressed as beats per minute.

Results and Discussion

Climatic Consideration and Temperature Humidity Index (THI)

The Fortnightly average of daily weather phenomenon and the Temperature Humidity Index (THI) values during the winter and summer seasons are presented in the Table-1. During the winter season the overall mean± SE values of maximum and minimum temperature were 21.49±0.88 and 08.18±0.85 respectively while during the summer season the overall mean± SE values of maximum and minimum temperature were 30.22±1.74 and 13.45±1.38 respectively. During the winter season the highest maximum temperature i.e. 25.27±0.60 was recorded in 1st fortnight while during the 5th fortnight lowest minimum temperature was recorded i.e. 05.01±0.55. As far as summer season is concern the highest maximum temperature i.e. 37.60±0.52 was recorded in 6th fortnight while during the 1st fortnight lowest minimum temperature was recorded i.e. 06.70±0.62. The overall mean± SE values of relative humidity (RH) during the winter season at morning and afternoon sessions were 94.79±1.03 and 54.60±3.01 respectively while during the summer season were 69.79±9.98 and 39.24±3.77 respectively. The maximum relative humidity was recorded in the morning session during the both season. The mean temperature-humidity index (THI) at morning and afternoon sessions during the winter season were 58.49±1.43 and 60.54±3.45 respectively while 75.07±1.72 and 59.20±1.29 respectively during the summer seasons. Various scientists reported threshold value of THI in the range of 65 to 70 in various farm animals. No specific threshold value of THI is available with respect to the calves. The threshold for heat stress in dairy cattle is a THI value of 68 (Britni *et al.,* 2015) [2] while Armstrong (1994) [1] stated that heat stress begins when temperature- humidity index is 72 or higher. In present experiment during the summer season, especially in the morning session the average THI exceeded the threshold for heat stress while during the winter season in the morning and afternoon session and in the summer season at the afternoon session, the average THI did not exceed the threshold limit.

Physiological Responses during the Winter and Summer Season

Physiological responses helps in the understanding of the illness exhibited by the change in the norms of rectal temperature, respiration rate and pulse rate which are considered as the vital signs and which ultimately helps in the diagnosis of certain diseases. Deviation from the normal physiological parameters considered as discomfort to the animals.

Rectal Temperature (RT)

The rectal tempratue (^{0}C) values of different treatment groups during the winter and summer season are presented in Table-2. During the winter season the overall mean± SE values of rectal temperature at morning session were 38.39±0.10, 38.39±0.02, 38.42±0.02 and 38.31±0.01 for T_1, T_2, T_3 and T_4 groups respectively and at afternoon session were 38.85±0.05, 38.80±0.01, 38.82±0.02 and 38.79±0.02 for T_1, T_2, T_3 and T_4 groups respectively. While during the summer season the overall mean± SE values of rectal temperature at morning session were 38.49±0.03, 38.36±0.01, 38.36±0.01 and 38.38±0.02 for T_1, T_2, T_3 and T_4 groups respectively and at afternoon session were 38.90±0.04, 38.86±0.01, 38.86±0.01 and 38.86±0.01 for T_1, T_2, T_3 and T_4 groups respectively. There were no significant ($P<0.05$) differences observed in rectal temperature between groups in the morning and afternoon during the winter as well as summer season. The rectal temperature in buffalo calves at afternoon was slightly higher in all groups during the winter as well as summer season. This was may be due to higher environment temperature in afternoon which was recorded during the experimental period. The rectal temperature of treatment group in the morning and at afternoon session during the summer season was slightly lower than control group. This might be due to fact that probiotic maintained the basal metabolic rate (BMR) at an optimum level and there by the rectal temperature of calves supplemented with probiotic group was lower as compared to control group (Chandra, 2006) [3]. The Rectal temperature observations in present study are in general agreement with the observations reported by Moran (1973) [9] who reported that in buffaloes there is an increased in rectal temperature under solar radiation and also with Chandra (2006) [3] who reported that the rectal temperature of calves varied from phase to phase in morning to afternoon observations, the rectal temperature of treatment group in the morning was lower than control group however, afternoon temperature in both groups was higher as compared to morning hour.

Respiration Rate (RR)

The mean± SE values of respiration rate *(*breaths/minute*)* in different treatment groups during the winter and summer season are presented in Table-3. During the winter the overall mean values for respiration rate at morning were 25.61±0.19, 25.28±0.16, 25.25±0.11 and 25.28±0.21 and 27.55±0.22, 27.61 ±0.15, 27.61±0.35 and 27.63±0.19 in the afternoon for T_1, T_2, T_3 and T_4 groups. While During the summer season the overall mean values for respiration rate at morning were 28.69±0.46, 28.37±0.61, 28.86±0.60 and 28.33±0.61 and 29.57±0.22, 29.23±0.26, 29.35±0.23 and 29.10±0.32 in the afternoon for T_1, T_2, T_3 and T_4 groups. There were no significant ($P<0.05$) difference in respiration rate between groups in the morning and afternoon during the winter and summer season. The respiration rate in buffalo calves at afternoon was slightly higher in all groups (T_1, T_2, T_3 and T_4) during the winter as well as summer season. This was mainly due to higher environment temperature in afternoon period. The results of the present study are in agreement with the observations reported by Chandra (2006) [3] who reported that the respiration rate/min was higher in evening as compared to morning in both treatments groups; this may be due to increased ambient temperature in the afternoon due to which the respiration rate of calves in evening was higher. The observation were falls in the normal physiological range so these findings indicated that probiotic supplementation had no adverse effect on physiology of calves resulted in normal respiration rate in morning and afternoon during the winter as well as summer season.

Pulse Rate (PR)

The pulse rate *(*beats/minute*)* values of different treatment groups during the winter and summer season are presented in

Table-4. During the winter season the overall mean values for pulse rate at morning were 54.22±0.08, 54.20±0.19, 53.86±0.14 & 53.84±0.14 and 55.49±0.42, 55.65±0.42, 55.14±0.43 & 54.86±0.27 in the afternoon for T_1, T_2, T_3 & T_4 groups respectively**.** While during the summer season the overall mean values for pulse rate at morning were 56.10±0.31, 56.08±0.35, 55.92±0.45 & 56.00±0.48 and 57.88±0.27, 57.47±0.35, 57.61±0.36 & 57.12±0.30 in the afternoon for T_1, T_2, T_3 & T_4 groups respectively. There were no significant ($P<0.05$) difference observed in pulse rate between groups in the morning and afternoon during the winter as well as summer season. The pulse rate in buffalo calves during afternoon was slightly higher in all groups during the winter as well as summer season. This was may be due to higher environment temperature in afternoon. These findings are in agreement with the observations reported by Chandra (2006) [3], Legha (1994) [5] and Rokde (1998) [11] who reported that, the pulse rate/min was higher in evening as compared to morning in treatment groups who reported for crossbred calves.

Physiological Responses in Pre-ruminant and Post-ruminant Calves:

Rectal Temperature (RT)

The rectal temperature (^{0}C) values of different treatment groups during the Pre and Post ruminant phase are presented in Table-5. During the Pre-ruminant phase the overall mean± SE values of rectal temperature at morning were 38.38±0.06, 38.36±0.01, 38.39±0.01 and 38.30±0.01 for T_1, T_2, T_3 and T_4 groups respectively and at afternoon were 38.84±0.03, 38.80±0.01, 38.82±0.01 and 38.79±0.01 for T_1, T_2, T_3 and T_4 groups respectively. While during the Post-ruminant Phase the overall mean± SE values of rectal temperature at morning were 38.45±0.02, 38.34±0.00, 38.34±0.00 and 38.36±0.01 for T_1, T_2, T_3 and T_4 groups respectively and at afternoon were 38.88±0.02, 38.85±0.01, 38.85±0.01 and 38.84±0.00 for T_1, T_2, T_3 and T_4 groups respectively. There were no significant ($P<0.05$) differences observed in rectal temperature between groups in the morning and afternoon during the Pre as well as Post ruminant phase. The rectal temperature in buffalo calves at afternoon was slightly higher in all groups during the Pre as well as Post ruminant phase. This was may be due to higher environment temperature in afternoon which was recorded during the experimental period. The rectal temperature of treatment group in the morning and at afternoon during the Pre as well as Post ruminant phase was lower than control group. This might be due to fact that probiotic maintained the basal metabolic rate (BMR) at an optimum level and there by the rectal temperature of calves supplemented with probiotic group was lower as compared to control group (Chandra, 2006) [3].

Respiration Rate (RR)

The mean± SE values of respiration rate *(*breaths/minute*)* in different treatment groups during the Pre and Post ruminant phase are presented in Table-6. During the Pre-ruminant phase the overall mean values for respiration rate at morning were 25.33±0.09, 25.27±0.09, 25.14±0.06 and 25.20±0.09 and 27.35±0.11, 27.61 ±0.16, 27.23±0.19 and 27.43±0.08 in the afternoon for T_1, T_2, T_3 and T_4 groups. While During the Post-ruminant phase the overall mean values for respiration rate at morning were 28.43±0.34, 28.22±0.41, 28.71±0.43 and 28.38±0.43 and 29.27±0.17, 29.31±0.11, 29.33±0.14 and 29.08±0.20 in the afternoon for T_1, T_2, T_3 and T_4 groups. There were no significant ($P<0.05$) difference in respiration rate between groups in the morning and afternoon during the Pre and Post ruminant phase. The respiration rate in buffalo calves at afternoon was slightly higher in all groups (T_1, T_2, T_3 and T_4) during the Pre as well as Post ruminant phase. This was mainly due to higher environment temperature in afternoon period. The results of the present study are in agreement with the observations reported by Chandra (2006) [3].

Pulse Rate (PR)

The pulse rate *(*beats/minute*)* values of different treatment groups during the Pre and Post ruminant phase are presented in Table-7. During the Pre-ruminant phase the overall mean values for pulse rate at morning were 54.08±0.05, 54.09±0.10, 53.95±0.12 & 53.93±0.14 and 55.38±0.25, 55.36±0.28, 55.12±0.26 & 55.04±0.15 in the afternoon for T_1, T_2, T_3 & T_4 groups respectively. While during the Post-ruminant phase the overall mean values for pulse rate at morning were 56.00±0.22, 56.00±0.31, 55.77±0.30 & 55.74±0.32 and 57.95±0.20, 57.74±0.20, 57.47±0.22 & 57.03±0.17 in the afternoon for T_1, T_2, T_3 & T_4 groups respectively. There were no significant ($P<0.05$) difference observed in pulse rate between groups in the morning and afternoon during the Pre as well as Post ruminant phase. The pulse rate in buffalo calves during afternoon was slightly higher in all groups during the Pre as well as Post ruminant phase. This was may be due to higher environment temperature in afternoon. These findings are in agreement with the observations reported by Chandra (2006) [3], Legha (1994) [5] and Rokde (1998) [11].

Physiological Responses in Female and Male Calves

Rectal Temperature (RT)

The rectal temperature (^{0}C) values of different treatment groups of female and male buffalo calves are presented in Table-8. In the female buffalo calves the overall mean± SE values of rectal temperature at morning were 38.39±0.06, 38.36±0.02, 38.36±0.02 and 38.35±0.02 for T_1, T_2, T_3 and T_4 groups respectively and at afternoon were 38.83±0.04, 38.81±0.02, 38.84±0.02 and 38.84±0.01 for T_1, T_2, T_3 and T_4 groups respectively. While in the male buffalo calves the overall mean± SE values of rectal temperature at morning were 38.37±0.03, 38.36±0.01, 38.33±0.01 and 38.34±0.01 for T_1, T_2, T_3 and T_4 groups respectively and at afternoon were 38.73±0.05, 38.86±0.01, 38.83±0.01 and 38.78±0.03 for T_1, T_2, T_3 and T_4 groups respectively. There were no significant ($P<0.05$) differences observed in rectal temperature between different treatment groups of female and male buffalo calves in the morning and afternoon session. The results of the present study are in agreement with the findings reported by the Pathmasingham *et al.,* (1982) [10] who reported in the Malaysian swamp buffaloes that the mean for males and females was 38.82 and 38.92^0 C, respectively. This difference was insignificant. Similar findings were reported by the Taneja and Bhatnagar (1958) [14] who reported for Indian buffaloes and Mensalvas and Rivera (1951) [8] who reported for Philippine carabaos. The rectal temperature in both the sexes of buffalo calves at afternoon was slightly higher in all groups. This was may be due to higher environment temperature in afternoon which was recorded during the experimental period. The rectal temperature of treatment group in the morning and at afternoon in the both sexes was lower than control group. This might be due to fact that probiotic maintained the basal metabolic rate (BMR) at an optimum level and there by the rectal temperature of calves supplemented with probiotic group was lower as compared to

control group (Chandra, 2006) [3]. The Rectal temperature observations in present study are in general agreement with the observations reported by Moran (1973) [9] who reported that in buffaloes there is an increased in rectal temperature under solar radiation and also with Chandra (2006) [3] who reported that the rectal temperature of calves varied from phase to phase in morning to afternoon observations, the rectal temperature of treatment group in the morning was lower than control group however, afternoon temperature in both groups was higher as compared to morning hour.

Respiration Rate (RR)

The mean± SE values of respiration rate *(*breaths/minute*)* in different treatment groups of female and male buffalo calves are presented in Table-9. In the female buffalo calves the overall mean values for respiration rate at morning were 27.33±0.54, 26.83±0.52, 27.10±0.62 and 26.92±0.50 and 28.58±0.37, 28.58±0.29, 28.17±0.36 and 28.37±0.27 in the afternoon for T_1, T_2, T_3 and T_4 groups. While in the male buffalo calves the overall mean values for respiration rate at morning were 26.70±0.54, 27.04±0.59, 27.25±0.63 and 27.06±0.58 and 28.25±0.33, 28.44±0.33, 28.44±0.41 and 28.46±0.31 in the afternoon for T_1, T_2, T_3 and T_4 groups. There were no significant ($P<0.05$) difference in respiration rate between groups in the morning and afternoon in the female and male buffalo calves. The respiration rate in female and male buffalo calves at afternoon was slightly higher in all groups (T_1, T_2, T_3 and T_4). This was mainly due to higher environment temperature in afternoon period. The results of the present study are in agreement with the observations reported by Mensalvas and Rivera (1951) [8] for Philippine carabaos who reported that the carabao males had 24.6 whilst the females had 27.7 respiration rates while contrary to the findings reported by Pathmasingham *et al.,* (1982) [10] who reported in the Malaysian swamp buffaloes that males and females having 49.82 and 47.79 respiration rate respectively which were higher than the present findings. Chandra (2006) [3] who reported that the respiration rate/min was higher in evening as compared to morning in both treatments groups; this may be due to increased ambient temperature in the afternoon due to which the respiration rate of calves in evening was higher. The observation were falls in the normal physiological range so these findings indicated that probiotic supplementation had no adverse effect on physiology of calves resulted in normal respiration rate in morning and afternoon.

Pulse Rate (PR)

The pulse rate *(*beats/minute*)* values of different treatment groups of female and male buffalo calves are presented in Table-10. In the female buffalo calves the overall mean values for pulse rate at morning were 55.10±0.30, 55.23±0.39, 54.87±0.35 & 55.12±0.38 and 56.71±0.44, 56.25±0.51, 56.25±0.53 & 56.00±0.45 in the afternoon for T_1, T_2, T_3 & T_4 groups respectively. While in the male buffalo calves the overall mean values for pulse rate at morning were 55.12±0.38, 55.06±0.36, 55.06±0.40 & 54.90±0.38 and 56.52±0.52, 56.69±0.41, 56.13±0.44 & 55.98±0.47 in the afternoon for T_1, T_2, T_3 & T_4 groups respectively. There were no significant ($P<0.05$) difference observed in pulse rate between groups in the morning and afternoon. The results of the present study are in agreement with the findings reported by Mensalvas and Rivera (1951) [8] for Philippine carabaos and Pathmasingham *et al.,* (1982) [10] who reported in the Malaysian swamp buffaloes. The differences between different groups of the females and males buffalo calves are slight and non-significant. The pulse rate in female and male buffalo calves during afternoon was slightly higher in all groups. This was may be due to higher environment temperature in afternoon. These findings are in agreement with the observations reported by Chandra (2006) [3], Legha (1994) [5] and Rokde (1998) [11] who reported that, the pulse rate/min was higher in evening as compared to morning in treatment groups who reported for crossbred calves.

Table 1: Fortnightly Average of Daily Weather Phenomenon* and Temperature Humidity Index (THI) at Morning and Afternoon Session during the Winter and Summer Season

Fort.	Winter Season										Summer Season									
	Temp. (°C)		Morning Session				Afternoon Session				Temp. (°C)		Morning Session				Afternoon Session			
	Max.	Min.	Dry Bulb	Wet Bulb	RH (%)	THI	Dry Bulb	Wet Bulb	RH (%)	THI	Max.	Min.	Dry Bulb	Wet Bulb	RH (%)	THI	Dry Bulb	Wet Bulb	RH (%)	THI
1	25.27 ±0.60	13.51 ±0.56	15.08 ±0.51	14.88 ±0.49	97.87 ±0.93	62.17 ±0.73	24.81 ±0.61	19.73 ±0.34	60.80 ±2.70	72.67 ±0.64	21.40 ±0.49	06.70 ±0.62	08.17 ±0.64	7.68 ±0.58	93.53 ±2.00	66.06 ±0.51	20.64 ±0.47	14.72 ±0.38	44.93 ±3.66	51.62 ±0.63
2	24.21 ±0.35	08.66 ±0.75	10.57 ±0.64	10.65 ±1.07	88.60 ±0.62	55.88 ±1.07	22.63 ±1.11	15.08 ±0.33	36.20 ±3.42	67.75 ±0.86	24-49 ±0.44	10.53 ± 0.69	12.38 ±0.79	11.97 ±0.69	95.54 ±2.11	71.32 ±0.60	24.32 ±0.41	18.34 ±0.47	53.46 ±1.98	56.68 ±0.81
3	22.15 ±0.93	08.56 ±0.52	10.05 ±0.51	09.44 ±0.48	92.47 ±3.28	54.64 ±0.68	20.99 ±0.99	14.41 ±0.43	47.93 ±5.83	66.09 ±0.91	28.23 ±0.48	12.31 ±0.51	15.44 ±0.43	14.45 ±0.37	90.00 ±1.77	74.51 ±0.49	27.47 ±0.48	19.63 ±0.24	45.33 ±1.63	59.52 ±0.47
4	20.67 ±0.56	06.83 ±0.42	08.16 ±0.39	07.88 ±0.39	96.06 ±1.56	52.15 ±0.55	19.89 ±0.63	15.10 ±0.25	59.19 ±4.71	65.79 ±0.54	30.56 ±0.53	13.84 ±0.46	17.31 ±0.46	16.44 ±1.17	80.94 ±2.55	75.78 ±0.78	28.86 ±0.97	20.00 ±0.34	36.75 ±1.59	61.08 ±1.02
5	16.97 ±1.08	05.01 ±0.55	06.31 ±0.50	06.13 ±0.47	97.53 ±1.35	61.37 ±1.14	16.52 ±1.12	12.33 ±0.46	62.20 ±5.84	45.02 ±0.35	33.29 ±0.71	17.25 ±0.39	21.83 ±0.34	18.48 ±0.27	7.53 ±3.13	80.53 ±0.89	32.92 ±0.89	22.53 ±0.73	39.67 ±4.71	63.96 ±0.50
6	19.68 ±0.77	06.48 ±0.53	07.65 ±0.56	07.35 ±0.48	96.19 ±2.17	64.70 ±0.96	18.85 ±0.92	14.63 ±0.47	61.25 ±4.42	45.89 ±0.34	37.60 ±0.52	20.05 ±0.76	24.99 ±0.50	18.25 ±0.49	51.20 ±2.39	82.21 ±0.67	37.16 ±0.52	20.63 ±0.61	15.29 ±2.09	62.34 ±0.73
Mean	21.49 ±0.88	08.18 ±0.85	09.64 ±0.89	09.39 ±0.90	94.79 ±1.03	58.49 ±1.43	20.62 ±0.84	15.21 ±0.70	54.60 ±3.01	60.54 ±3.45	30.22 ±1.74	13.45 ±1.38	16.69 ±1.77	14.55 ±1.20	69.79 ±9.98	75.07 ±1.72	28.56 ±1.71	19.31 ±0.76	39.24 ±3.77	59.20 ±1.29

*Daily Weather Phenomenon data were adapted from the recording made at Central Soil and Salinity Research Institute (C.S.S.R.I.), Karnal, Haryana.

Table 2: Rectal Temperature (0^C) in Morning and Afternoon Session at Fortnightly intervals in different groups during the Winter and Summer Season (N=7)

Fortnight	Winter Season								Summer Season							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	38.17 ±0.18	38.40 ±0.15	38.44 ±0.12	38.29 ±0.04	38.78 ±0.06	38.75 ±0.03	38.93 ±0.15	38.68 ±0.14	38.42 ±0.05	38.34 ±0.02	38.35 ±0.02	38.34 ±0.02	39.13 ±0.18	38.84 ±0.02	38.85 ±0.02	38.84 ±0.02
1	38.05 ±0.17	38.51 ±0.15	38.46 ±0.15	38.31 ±0.02	38.83 ±0.02	38.81 ±0.02	38.82 ±0.02	38.81 ±0.02	38.48 ±0.13	38.33 ±0.02	38.32 ±0.02	38.33 ±0.02	38.84 ±0.02	38.83 ±0.02	38.82 ±0.02	38.83 ±0.02
2	38.56 ±0.21	38.35 ±0.07	38.48 ±0.19	38.27 ±0.08	38.77 ±0.08	38.76 ±0.04	38.76 ±0.08	38.77 ±0.08	38.42 ±0.05	38.35 ±0.02	38.37 ±0.01	38.36 ±0.01	38.86 ±0.01	38.85 ±0.02	38.87 ±0.01	38.86 ±0.01
3	38.87 ±0.51	38.34 ±0.09	38.42 ±0.18	38.30 ±0.07	38.80 ±0.07	38.80 ±0.07	38.78 ±0.07	38.80 ±0.07	38.45 ±0.06	38.44 ±0.13	38.38 ±0.02	38.38 ±0.01	38.88 ±0.02	38.94 ±0.13	38.88 ±0.02	38.88 ±0.01
4	38.28 ±0.06	38.41 ±0.07	38.43 ±0.15	38.28 ±0.06	38.78 ±0.06	38.79 ±0.05	38.79 ±0.04	38.78 ±0.06	38.49 ±0.13	38.34 ±0.03	38.35 ±0.02	38.34 ±0.02	38.84 ±0.02	38.84 ±0.03	38.85 ±0.02	38.84 ±0.02
5	38.35 ±0.02	38.35 ±0.02	38.34 ±0.02	38.35 ±0.02	38.85 ±0.02	38.85 ±0.02	38.84 ±0.02	38.85 ±0.02	38.51 ±0.15	38.36 ±0.02	38.37 ±0.02	38.50 ±0.14	38.87 ±0.02	38.86 ±0.02	38.87 ±0.02	38.86 ±0.01
6	38.42 ±0.05	38.34 ±0.02	38.35 ±0.02	38.34 ±0.02	39.13 ±0.18	38.84 ±0.02	38.85 ±0.02	38.84 ±0.02	38.67 ±0.29	38.38 ±0.01	38.40 ±0.01	38.38 ±0.01	38.88 ±0.01	38.88 ±0.01	38.90 ±0.01	38.88 ±0.01
Overall Mean	**38.39±0.10**	**38.39±0.02**	**38.42±0.02**	**38.31±0.01**	**38.85±0.05**	**38.80±0.01**	**38.82±0.02**	**38.79±0.02**	**38.49±0.03**	**38.36±0.01**	**38.36±0.01**	**38.38±0.02**	**38.90±0.04**	**38.86±0.01**	**38.86±0.01**	**38.86±0.01**

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 3: Respiration Rate (breaths/min) in Morning and Afternoon Session at Fortnightly intervals in different groups during the Winter and Summer Season (N=7)

Fortnight	Winter Season								Summer Season							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	25.43±0.30	25.57±0.48	25.14±0.40	24.71±0.52	26.86±0.46	27.43±0.37	28.86±0.26	27.14±0.26	26.14±0.40	24.86±0.55	25.43±0.30	24.86±0.55	28.43±0.57	27.86±0.59	28.57±0.53	28.29±0.81
1	25.86±0.40	25.43±0.48	25.29±0.61	25.71±0.36	27.43±0.37	27.71±0.42	27.29±0.36	27.86±0.34	28.43±0.37	28.43±0.65	29.57±0.48	28.43±0.37	29.57±0.53	29.29±0.42	29.71±0.75	27.71±0.57
2	26.14±0.40	24.71±0.52	25.71±0.52	24.86±0.55	27.71±0.29	27.14±0.26	28.14±0.26	27.14±0.14	29.43±0.37	29.00±0.65	29.29±0.52	28.29±0.42	29.43±0.48	29.14±0.74	29.86±0.59	29.14±0.70
3	25.71±0.52	25.71±0.36	25.00±0.38	26.14±0.40	28.14±0.26	27.86±0.34	27.14±0.26	27.71±0.29	29.71±0.42	29.57±0.48	29.71±0.29	29.00±0.49	30.00±0.44	29.43±0.65	29.00±0.44	29.29±0.61
4	25.14±0.40	25.71±0.52	24.86±0.55	25.71±0.52	27.14±0.26	28.14±0.26	26.86±0.51	28.14±0.26	28.86±0.34	29.57±0.37	30.14±0.40	29.14±0.40	29.71±0.47	29.43±0.37	29.14±0.51	29.57±0.48
5	24.86±0.55	25.00±0.38	25.29±0.36	25.00±0.38	27.14±0.14	27.14±0.26	26.43±0.48	27.14±0.26	28.86±0.67	28.43±0.37	28.43±0.69	28.71±0.29	29.57±0.53	29.29±0.47	28.86±0.51	29.43±0.53
6	26.14±0.40	24.86±0.55	25.43±0.30	24.86±0.55	28.43±0.57	27.86±0.59	28.57±0.53	28.29±0.81	29.43±0.48	28.71±0.52	29.43±0.53	29.86±0.46	30.29±0.29	30.14±0.34	30.29±0.29	30.29±0.47
Overall Mean	25.61±0.19	25.28±0.16	25.25±0.11	25.28±0.21	27.55±0.22	27.61±0.15	27.61±0.35	27.63±0.19	28.69±0.46	28.37±0.61	28.86±0.60	28.33±0.61	29.57±0.22	29.23±0.26	29.35±0.23	29.10±0.32

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 4: Pulse Rate (beats/min) in Morning and Afternoon Session at Fortnightly intervals in different groups during the Winter and Summer Season (N=7)

Fortnight	Winter Season								Summer Season							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	54.14±0.51	54.57±037.	53.57±0.37	53.86±0.34	55.29±0.52	55.57±0.78	55.14±0.88	54.71±0.42	54.57±0.48	54.14±0.34	53.57±0.53	53.29±0.52	57.71±0.64	57.71±1.04	56.86±1.14	56.00±1.31
1	54.29±0.42	53.86±0.51	54.14±0.34	53.86±0.40	55.43±0.48	55.29±0.75	55.00±0.72	53.86±0.40	56.43±0.37	56.14±0.26	55.57±0.37	56.14±0.26	56.71±0.64	56.00±0.22	56.14±0.40	56.71±0.75
2	54.14±0.51	54.29±0.42	54.14±0.40	53.86±0.34	54.57±0.53	54.29±0.42	54.14±0.40	54.57±0.65	56.29±0.36	56.43±0.20	56.00±0.31	56.43±0.30	57.57±0.72	57.71±0.68	58.86±0.99	57.43±0.78
3	54.43±0.43	54.57±0.53	54.43±0.53	54.29±0.47	55.29±0.47	56.00±0.76	55.57±0.81	55.14±0.86	57.00±0.38	56.29±0.29	56.14±0.26	56.00±0.31	58.14±0.51	57.86±0.83	57.71±0.78	57.14±0.51
4	54.00±0.44	54.71±0.36	53.43±0.48	54.29±0.52	54.29±0.61	54.71±0.36	53.43±0.48	54.29±0.52	55.57±0.43	56.00±0.62	56.00±0.62	56.14±0.55	58.00±0.58	57.14±0.59	57.14±0.91	57.14±0.70
5	54.00±0.53	53.29±0.36	53.71±0.47	53.43±0.43	55.86±0.80	56.00±1.02	55.86±1.16	55.43±1.34	56.14±0.40	56.43±0.48	57.14±0.40	56.86±0.40	57.86±0.63	56.86±0.46	58.14±0.67	56.86±0.40
6	54.57±0.48	54.14±0.34	53.57±0.53	53.29±0.52	57.71±0.64	57.71±1.04	56.86±1.14	56.00±1.31	56.71±0.29	57.14±0.46	57.00±0.44	57.14±0.46	59.14±0.77	59.00±0.82	58.43±0.90	58.57±0.75
Overall Mean	54.22±0.08	54.20±0.19	53.86±0.14	53.84±0.14	55.49±0.42	55.65±0.42	55.14±0.43	54.86±0.27	56.10±0.31	56.08±0.35	55.92±0.45	56.00±0.48	57.88±0.27	57.47±0.35	57.61±0.36	57.12±0.30

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 5: Rectal Temperature (0^C) in Morning and Afternoon Session at Fortnightly intervals in different groups during the Pre-Ruminant and Post-Ruminant Phase (N=9)

Fortnight	Pre-Ruminant Phase								Post-Ruminant Phase							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	38.20±0.13	38.38±0.11	38.40±0.09	38.26±0.03	38.79±0.04	38.77±0.02	38.89±0.12	38.67±0.10	38.38±0.04	38.32±0.02	38.32±0.02	38.32±0.02	39.07±0.10	38.82±0.02	38.82±0.02	38.82±0.02
1	38.09±0.13	38.46±0.11	38.42±0.11	38.31±0.01	38.81±0.01	38.81±0.01	38.81±0.01	38.81±0.01	38.44±0.10	38.33±0.01	38.33±0.02	38.33±0.01	38.83±0.01	38.83±0.01	38.83±0.02	38.83±0.01
2	38.51±0.16	38.34±0.05	38.44±0.14	38.28±0.06	38.78±0.06	38.78±0.03	38.78±0.06	38.78±0.03	38.39±0.04	38.35±0.01	38.35±0.01	38.35±0.01	38.85±0.01	38.85±0.01	38.85±0.01	38.85±0.01
3	38.74±0.39	38.33±0.06	38.41±0.13	38.30±0.05	38.80±0.05	38.80±0.05	38.80±0.05	38.80±0.05	38.42±0.05	38.36±0.11	38.36±0.01	38.36±0.01	38.86±0.01	38.86±0.11	38.86±0.01	38.86±0.01
4	38.40±0.12	38.37±0.05	38.40±0.11	38.29±0.05	38.79±0.04	38.79±0.04	38.79±0.02	38.79±0.05	38.45±0.10	38.33±0.01	38.33±0.02	38.33±0.01	38.83±0.01	38.83±0.01	38.83±0.02	38.83±0.01
5	38.34±0.01	38.34±0.01	38.34±0.01	38.34±0.01	38.84±0.01	38.84±0.01	38.84±0.01	38.84±0.01	38.47±0.11	38.35±0.01	38.35±0.01	38.46±0.10	38.85±0.01	38.85±0.01	38.85±0.01	38.85±0.01
6	38.38±0.04	38.32±0.02	38.32±0.02	38.32±0.02	39.07±0.10	38.82±0.02	38.82±0.02	38.82±0.02	38.59±0.22	38.37±0.01	38.37±0.01	38.37±0.01	38.87±0.01	38.88±0.01	38.88±0.01	38.87±0.01
Overall Mean	38.38±0.06	38.36±0.01	38.39±0.01	38.30±0.01	38.84±0.03	38.80±0.01	38.82±0.01	38.79±0.01	38.45±0.02	38.34±0.00	38.34±0.00	38.36±0.01	38.88±0.02	38.85±0.01	38.85±0.01	38.84±0.00

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8 cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8 cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 6: Respiration Rate (breaths/min) in Morning and Afternoon Session at fortnightly intervals in different groups during the Pre-Ruminant and Post-Ruminant Phase (N=9)

Fortnight	Pre-Ruminant Phase								Post-Ruminant Phase							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	25.22±0.27	25.77±0.46	25.00±0.33	25.00±0.44	26.88±0.35	27.22±0.32	26.88±0.20	27.22±0.22	25.77±0.40	24.88±.053	25.22±0.27	24.88±0.53	28.11±0.51	28.88±0.56	28.66±.44	27.78±0.70
1	25.44±0.41	25.33±0.40	25.00±0.50	25.55±0.33	27.33±0.28	27.55±0.37	27.11±0.30	27.77±0.27	28.00±0.40	28.22±0.52	29.66±0.40	29.00±0.47	29.11±0.51	29.22±0.36	29.88±0.58	28.55±0.70
2	25.66±0.47	25.00±0.44	25.33±0.47	24.88±0.53	27.44±0.29	27.22±0.22	27.55±0.44	27.00±0.28	29.11±0.35	28.88±0.51	29.00±0.44	28.44±.37	29.22±0.40	29.22±0.57	29.44±.60	29.22±0.54
3	25.33±0.47	25.55±0.33	25.00±0.33	25.77±0.40	27.55±0.44	27.77±0.27	27.11±0.26	27.55±0.24	29.22±0.46	29.22±0.46	29.44±0.29	29.11±.38	29.55±0.47	29.33±0.52	29.00±0.37	29.55±0.50
4	25.00±0.33	25.33±0.47	24.88±0.53	25.33±0.47	27.11±0.26	27.55±0.44	27.00±0.47	27.55±0.44	29.00±0.37	29.33±0.40	30.11±0.35	29.55±.41	29.33±0.44	29.11±0.38	29.22±0.40	29.11±0.48
5	24.88±0.53	25.00±0.33	25.55±0.33	25.00±0.33	27.00±0.28	27.11±0.26	26.33±0.40	27.11±0.26	28.55±0.55	28.11±0.38	28.44±0.52	28.22±.40	29.33±0.44	29.22±0.36	29.00±0.40	29.22±0.43
6	25.77±0.40	24.88±.053	25.22±0.27	24.88±0.53	28.11±0.51	28.88±0.56	28.66±.44	27.78±0.70	29.33±0.40	28.88±0.42	29.11±0.48	29.44±0.44	30.22±0.27	30.22±0.27	30.11±0.26	30.11±0.38
Overall Mean	25.33±0.09	25.27±0.09	25.14±0.06	25.20±0.09	27.35±0.11	27.61±0.16	27.23±0.19	27.43±0.08	28.43±0.34	28.22±0.41	28.71±0.43	28.38±0.43	29.27±0.17	29.31±0.11	29.33±0.14	29.08±0.20

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8 cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8 cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 7: Pulse Rate (beats/min) in Morning and Afternoon Session at Fortnightly intervals in different groups during the Pre-Ruminant and Post-Ruminant Phase (N=9)

Fortnight	Pre-Ruminant Phase								Post-Ruminant Phase							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	54.00±0.40	54.55±0.29	53.88±0.38	54.11±0.35	54.88±0.48	55.33±0.62	55.11±0.69	55.22±0.81	54.33±0.40	53.66±0.55	53.44±0.41	53.22±0.40	57.22±0.64	57.33±0.83	56.77±1.05	56.11±1.16
1	54.11±0.35	54.11±0.42	54.00±0.37	53.77±0.40	55.33±0.31	55.22±0.40	55.11±0.57	54.77±0.56	56.33±0.40	55.55±0.28	55.66±0.29	55.66±0.28	56.77±0.59	56.33±0.28	56.00±0.33	56.44±0.60
2	53.88±0.42	54.00±0.40	54.44±0.37	54.22±0.36	54.55±0.47	54.00±0.40	54.44±0.37	54.77±0.52	56.33±0.28	56.55±0.24	56.00±0.23	56.33±0.23	58.00±0.74	58.11±0.58	58.22±0.86	57.11±0.63
3	54.33±0.33	54.22±0.46	54.66±0.44	54.55±0.41	55.00±0.40	55.33±0.72	55.55±0.62	55.22±0.66	56.66±0.37	56.66±0.37	56.11±0.26	56.11±0.26	58.55±0.50	58.33±0.72	57.88±0.73	57.00±0.40
4	53.88±0.42	54.55±0.33	53.66±0.40	54.33±0.40	54.66±0.55	54.55±0.33	53.66±0.40	54.33±0.40	55.55±0.33	56.00±0.50	55.77±0.52	55.55±0.50	58.00±0.44	57.77±0.64	57.88±0.67	57.44±0.58
5	54.00±0.44	53.55±0.41	53.55±0.41	53.33±0.37	56.00±0.62	55.77±0.79	55.22±0.99	54.88±1.09	56.11±0.35	56.33±0.37	56.66±0.44	56.44±0.41	58.11±0.53	57.66±0.68	57.44±0.68	57.11±0.61
6	54.33±0.40	53.66±0.55	53.44±0.41	53.22±0.40	57.22±0.64	57.33±0.83	56.77±1.05	56.11±1.16	56.66±0.23	57.22±0.36	56.77±0.36	56.88±0.38	59.00±0.72	58.66±0.66	58.11±0.73	58.00±0.68
Overall Mean	54.08±0.05	54.09±0.10	53.95±0.12	53.93±0.14	55.38±0.25	55.36±0.28	55.12±0.26	55.04±0.15	56.00±0.22	56.00±0.31	55.77±0.30	55.74±0.32	57.95±0.20	57.74±0.20	57.47±0.22	57.03±0.17

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8 cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8 cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 8: Rectal Temperature (0^C) in Morning and Afternoon Session at Fortnightly intervals in Female and Male Calves (N=4)

Fortnight	Female Calves								Male Calves							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	38.84±0.03	38.22±0.04	38.32±0.05	38.27±0.06	38.84±0.03	38.72±0.04	39.04±0.27	38.77±0.06	38.29±0.06	38.30±0.03	38.24±0.06	38.30±0.06	38.40±0.06	38.80±0.03	38.75±0.06	38.63±0.05
1	37.83±0.26	38.54±0.26	38.31±0.03	38.30±0.03	38.83±0.03	38.79±0.02	38.81±0.03	38.80±0.03	38.31±0.03	38.37±0.05	38.32±0.03	38.32±0.03	38.81±0.03	38.84±0.03	38.82±0.02	38.82±0.03
2	38.34±0.03	38.41±0.11	38.31±0.20	38.35±0.02	38.84±0.03	38.75±0.06	38.69±0.13	38.85±0.02	38.73±0.36	38.30±0.05	38.36±0.02	38.31±0.04	38.43±0.05	38.80±0.05	38.87±0.02	38.56±0.09
3	38.36±0.02	38.23±0.11	38.23±0.11	38.34±0.02	38.86±0.02	38.73±0.11	38.73±0.11	38.84±0.02	38.34±0.02	38.44±0.09	38.36±0.02	38.36±0.02	38.61±0.09	38.87±0.03	38.86±0.02	38.58±0.07
4	38.34±0.03	38.40±0.13	38.52±0.28	38.34±0.02	38.84±0.03	38.71±0.07	38.77±0.06	38.84±0.02	38.31±0.04	38.37±0.02	38.31±0.04	38.28±0.06	38.44±0.08	38.87±0.02	38.81±0.04	38.66±0.09
5	38.34±0.03	38.34±0.03	38.35±0.03	38.33±0.02	38.84±0.03	38.84±0.03	38.85±0.03	38.83±0.02	38.36±0.02	38.37±0.03	38.33±0.02	38.35±0.03	38.86±0.02	38.87±0.03	38.83±0.02	38.85±0.03
6	38.46±0.09	38.31±0.03	38.34±0.04	38.32±0.04	39.09±0.21	38.81±0.03	38.84±0.04	38.82±0.04	38.35±0.02	38.37±0.03	38.33±0.04	38.34±0.04	38.85±0.02	38.87±0.03	38.83±0.04	38.84±0.04
7	38.35±0.02	38.31±0.03	38.33±0.03	38.34±0.02	38.85±0.02	38.81±0.03	38.83±0.03	38.84±0.02	38.32±0.03	38.37±0.03	38.32±0.03	38.33±0.03	38.82±0.03	38.87±0.03	38.82±0.03	38.83±0.03
8	38.46±0.09	38.34±0.03	38.37±0.01	38.34±0.02	38.86±0.02	38.84±0.03	38.87±0.01	38.84±0.02	38.36±0.02	38.37±0.03	38.35±0.02	38.37±0.02	38.86±0.02	38.87±0.03	38.85±0.02	38.87±0.02
9	38.38±0.02	38.46±0.24	38.40±0.01	38.36±0.02	38.38±0.02	38.96±0.24	38.90±0.01	38.86±0.02	38.34±0.05	38.40±0.02	38.34±0.02	38.40±0.01	38.86±0.02	38.90±0.02	38.84±0.02	38.90±0.01
10	38.59±0.23	38.34±0.01	38.35±0.03	38.34±0.02	38.84±0.03	38.84±0.01	38.85±0.03	38.84±0.02	38.35±0.02	38.34±0.05	38.31±0.04	38.34±0.03	38.85±0.02	38.84±0.05	38.82±0.04	38.84±0.03
11	38.36±0.02	38.38±0.01	38.38±0.02	38.61±0.25	38.86±0.02	38.88±0.01	38.88±0.02	38.86±0.01	38.37±0.03	38.34±0.02	38.34±0.02	38.36±0.02	38.87±0.03	38.84±0.02	38.85±0.02	38.86±0.02
12	38.37±0.02	38.38±0.02	38.42±0.01	38.37±0.02	38.87±0.02	38.88±0.02	38.92±0.01	38.87±0.02	38.39±0.02	38.39±0.02	38.38±0.02	38.38±0.02	38.89±0.02	38.89±0.02	38.89±0.02	38.88±0.02
Overall Mean	38.39±0.06	38.36±0.02	38.36±0.02	38.35±0.02	38.83±0.04	38.81±0.02	38.84±0.02	38.84±0.01	38.37±0.03	38.36±0.01	38.33±0.01	38.34±0.01	38.73±0.05	38.86±0.01	38.83±0.01	38.78±0.03

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 9: Respiration rate (breaths/min) in morning and afternoon session at fortnightly intervals in Female and Male Calves (N=4)

Fortnight	Female Calves								Male Calves							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	25.75±0.25	25.75±0.48	25.50±0.65	24.75±0.48	26.50±0.65	27.50±0.29	26.75±0.25	27.25±0.25	24.75±0.48	25.25±0.75	24.50±0.29	25.00±0.91	27.25±0.48	27.25±0.63	26.75±0.48	27.25±0.48
1	26.25±0.48	25.50±0.65	25.00±0.91	25.25±0.48	27.50±0.65	28.50±0.25	27.00±0.41	27.50±0.29	25.00±0.58	25.50±0.65	25.25±0.75	25.75±0.63	27.25±0.25	27.25±0.63	27.25±0.63	28.00±0.58
2	26.00±0.71	25.50±0.87	25.00±0.71	25.25±0.75	27.50±0.29	27.50±0.29	28.00±0.41	27.25±0.25	25.5±0.87	24.25±0.25	26.00±0.71	25.00±0.91	27.50±0.65	26.75±0.25	27.75±0.63	27.25±0.25
3	26.00±0.71	25.75±0.63	25.00±0.71	25.75±0.63	28.50±0.29	28.25±0.48	27.25±0.25	27.75±0.48	25.00±0.71	25.75±0.25	25.25±0.25	26.00±0.71	27.00±0.71	27.50±0.29	27.25±0.48	27.50±0.29
4	25.00±0.41	25.00±0.71	25.00±0.91	25.5±0.87	27.25±0.25	27.50±0.87	27.75±0.25	27.25±0.48	25.00±0.71	26.00±0.71	25.25±0.75	25.50±0.65	26.75±0.48	28.00±0.41	26.50±0.96	28.50±0.29
5	24.75±0.75	24.25±0.25	25.00±0.58	25.75±0.48	27.00±0.00	26.75±0.25	26.00±0.71	27.50±0.50	24.05±0.87	25.50±0.50	26.00±0.41	24.50±0.29	26.75±0.63	27.25±0.48	26.50±0.65	27.00±0.00
6	26.00±0.71	24.75±1.03	25.50±0.29	25.25±0.75	28.25±0.63	29.00±0.91	27.75±0.25	28.75±1.31	26.00±0.41	24.50±0.50	25.25±0.48	25.00±0.91	28.50±0.87	29.25±0.85	29.75±0.63	28.50±1.04
7	28.5±0.29	28.75±0.48	28.75±0.48	28.5±0.87	29.50±0.50	30.00±0.41	29.00±1.08	28.50±1.26	27.75±0.85	28.00±1.08	30.75±0.25	29.00±0.41	29.25±0.95	28.75±0.48	30.75±0.63	28.00±0.91
8	29.00±0.41	28.25±0.48	29.25±0.85	28.5±0.65	29.75±0.48	29.25±0.48	30.00±1.08	29.75±1.18	29.05±0.65	29.75±0.95	29.00±0.58	28.50±0.65	29.00±0.71	29.25±1.31	29.25±0.48	28.75±0.48
9	30.00±0.41	28.50±0.65	29.75±0.48	28.75±0.63	30.25±0.75	28.50±0.65	28.50±0.29	29.50±0.87	29.00±0.71	30.00±0.71	29.50±0.29	29.50±0.65	29.50±0.29	30.00±0.91	29.75±0.63	29.50±0.87
10	29.25±0.48	29.25±0.48	29.75±0.63	29.00±0.71	29.75±0.63	29.25±0.85	28.75±0.63	29.25±0.75	29.00±0.71	30.00±0.41	30.75±0.25	29.75±0.48	29.25±0.75	29.00±0.41	29.50±0.65	29.50±0.65
11	29.00±1.08	28.50±0.29	28.75±0.63	28.00±0.71	29.50±0.75	29.50±0.65	29.00±0.91	29.00±0.58	28.25±0.75	28.25±0.63	28.00±1.08	28.75±0.48	29.25±0.63	29.00±0.58	28.75±0.25	29.50±0.87
12	29.75±0.75	29.00±0.71	30.00±0.71	29.75±0.75	30.25±0.48	30.00±0.41	30.50±0.29	29.50±0.50	28.75±0.48	28.75±0.75	28.75±0.48	29.50±0.65	30.00±0.41	30.50±0.50	30.00±0.41	30.75±0.63
Overall Mean	27.33±0.54	26.83±0.52	27.10±0.62	26.92±0.50	28.58±0.37	28.58±0.29	28.17±0.36	28.37±0.27	26.70±0.54	27.04±0.59	27.25±0.63	27.06±0.58	28.25±0.33	28.44±0.33	28.44±0.41	28.46±0.31

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Table 10: Pulse rate (beats/min) in morning and afternoon session at fortnightly intervals in Female and Male Calves (N=4)

Fortnight	Female Calves								Male Calves							
	Morning Session				Afternoon Session				Morning Session				Afternoon Session			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
0	54.25±0.85	54.50±0.29	53.50±0.29	54.50±0.29	54.75±0.75	54.50±0.29	54.25±0.63	54.50±0.29	53.75±0.48	54.50±0.65	53.75±0.63	53.25±0.25	55.25±0.85	56.25±1.31	55.75±1.44	54.75±0.75
1	54.00±0.71	54.50±0.65	53.75±0.25	54.50±0.65	55.75±0.75	54.50±0.65	55.25±1.25	54.50±0.65	54.50±0.29	53.50±0.65	54.75±0.48	53.5±0.29	54.75±0.25	56.00±1.08	54.75±0.48	53.50±0.29
2	54.50±0.65	53.50±0.65	54.00±0.58	54.50±0.65	55.25±0.48	53.50±0.65	54.00±0.58	54.50±0.65	53.50±0.65	54.50±0.65	54.75±0.63	53.75±0.48	54.25±0.85	54.50±0.65	54.75±0.63	55.00±1.08
3	55.00±0.41	53.75±0.85	54.75±0.48	54.25±0.85	55.75±0.48	56.00±1.47	54.75±0.48	54.50±1.04	53.75±0.48	55.00±0.41	54.25±0.85	54.5±0.29	54.50±0.65	55.25±0.63	56.25±1.31	55.75±1.11
4	54.00±0.71	54.00±0.58	53.25±0.75	54.25±0.48	54.50±1.04	54.00±0.58	53.25±0.75	54.25±0.48	53.50±0.65	55.00±0.41	53.75±0.48	54.25±0.85	54.75±0.85	55.00±0.41	53.75±0.48	54.25±0.85
5	53.75±0.75	54.00±0.58	53.50±0.50	53.00±0.71	56.75±1.25	55.25±1.03	57.25±1.65	56.50±2.33	54.00±0.58	53.00±0.41	53.50±0.87	53.5±0.50	55.00±0.41	56.50±1.55	53.50±0.87	53.50±0.50
6	53.75±0.48	54.00±0.82	54.25±0.48	53.00±0.71	58.25±0.75	56.00±1,41	55.75±1.11	54.75±1.89	55.00±0.71	53.00±0.41	52.75±0.63	53.5±0.65	57.00±0.82	59.00±0.71	57.00±2.04	58.25±1.25
7	56.00±0.41	57.00±0.41	55.25±0.48	56.25±0.48	56.75±1.03	56.75±0.48	56.25±0.63	56.00±0.41	56.25±0.85	55.75±0.25	55.75±0.48	55.75±0.25	56.25±0.75	55.75±0.25	55.75±0.48	57.00±1.35
8	56.25±0.25	56.50±0.29	55.75±0.48	56.50±0.29	56.25±0.25	57.50±0.65	58.75±1.38	56.50±0.29	56.25±0.63	56.25±0.25	56.25±0.25	56.25±0.48	58.75±0.85	58.25±1.11	58.25±1.44	58.00±1.35
9	56.50±0.29	56.25±0.48	56.50±0.29	56.00±0.41	57.50±0.65	57.75±1.38	57.25±1.11	57.00±0.71	57.00±0.82	56.50±0.29	55.75±0.25	56.00±0.41	59.50±0.65	58.75±1.03	57.75±1.03	57.00±0.71
10	55.75±0.48	55.75±1.11	55.50±0.29	56.00±1.00	57.75±0.48	58.00±1.00	57.00±0.91	58.75±0.48	55.50±0.65	56.00±0.41	56.25±1.11	56.00±0.41	58.25±0.95	56.75±0.48	58.00±1.58	55.75±0.25
11	56.00±0.71	56.75±0.48	56.75±0.63	56.50±0.65	57.75±0.95	58.25±1.31	58.50±1.19	58.00±1.08	56.50±0.29	56.00±0.71	57.00±0.71	56.75±0.63	58.00±0.11	56.75±0.75	57.00±0.71	56.75±0.63
12	56.50±0.29	57.50±0.65	56.50±0.29	57.25±0.75	60.25±0.85	59.25±0.85	59.00±1.47	58.25±0.85	57.00±0.41	56.75±0.48	57.25±0.75	56.75±0.48	58.50±1.04	58.25±1.31	57.25±0.75	58.25±1.31
Overall Mean	55.10±0.30	55.23±0.39	54.87±0.35	55.12±0.38	56.71±0.44	56.25±0.51	56.25±0.53	56.00±0.45	55.12±0.38	55.06±0.36	55.06±0.40	54.90±0.38	56.52±0.52	56.69±0.41	56.13±0.44	55.98±0.47

Standard diet without supplementation (T_1) or supplemented with *Lactobacillus plantarum* CRD 2 10^8 cfu/ml (As fermented milk @ 100 ml/calf/day) (T_2), *Lactobacillus rhamnosus* CRD 9 supplementation 10^8cfu/ml (As a fermented milk @ 100 ml/calf/day) (T_3) and combination of *Lactobacillus plantarum* CRD 2 and *Lactobacillus rhamnosus* CRD 9 10^8cfu/ml [1:1 Ratio] (As a fermented milk @ 100 ml/calf/day) (T_4).

Conclusions

From the findings of this research study, it might be concluded that probiotic supplementation advantageous in the removal of stress in the young buffalo calves during the stress period which are more prone to cold as well as heat stress. Also a baseline data on physiological responses during the different seasons, different phases and also in different sexes has been established which were lacking especially for Indian buffalo calves so that future research strategies should be focussed along these lines to manage the young buffalo calves during the stress period effectively and profitably.

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