ABSTRACT

Chitwan district of Nepal has been known as the dairy kingdom of the country considering its strategic location and potential to easily provision inputs and produce as well as market quality milk across the country. However, the climate especially during summer has been a challenge to farmers that potentially compromise the daily milk yield of cows. Therefore, sixteen crossbred dairy cattle were placed in a RCBD fashion to assess the impacts of cold water bathing in ameliorating effect of heat stress on their performance and body physiology. Routine assessment of the microclimate within the experimental shed and blood parameters was made. The results from the experiment inferred that frequency of bathing (none, once, twice or thrice a day) did not have any significant impact on Haemoglobin, Haematocrit, Sodium, Potassium, Chloride and Bicarbonate levels in the blood plasma(p>0.05). In addition, the animals did not exhibit any signs of physiological distress clinically either. A detailed study scoping temperature humidity index and over a number of other milk and blood parameters are to be tested across a number of other available breeds too in order for the researchers to come to a meaningful strategy to beat the heat stress.

Keywords: Heat Stress, Salt balance, Haematology

INTRODUCTION

There is general agreement that average temperatures in Nepal increased at an annual rate of 0.06°C between 1977 and 2000, with a 0.04°C increases in the terai and 0.08°C increases in the Himalayas (Malla, 2009). Warming patterns have been most pronounced at higher altitudes and more so during the winter months as opposed to the terai and monsoon season. Warming has also been greater in the western half of the country than in its eastern counterpart (Chhetri & Easterling, 2010). The climatic conditions of our country vary from place to place in accordance with the geographical features. In the north, summers are cool and winter are severe, while in the south, summers are very warm and winters are mild. An average temperature drop of 6°C occurs for every 1000 m increase in altitude. In the terai, summer temperatures exceed 37°C and higher in some areas, winter temperatures range from 7°C to 23°C in terai. The average temperature of Chitwan in summer (May, June and July) has been observed to be maximum 33°C and minimum 25.3°C (Anonymous, 2015). Extreme climatic conditions affect the performance of dairy cattle and rather extremities of either hot or cold further exacerbates the condition by affecting the normal physiology of all classes of animals (Paudel & Perrera, 2009). Hot environments affect the performance of dairy cattle in a variety of different ways. In lactating Holstein cows, for example, the comfortable temperature is within the range 4-24°C (Hahn 1981; Roenfeldt, 1998). The effects of heat stress on the cows usually begin to be observed above 24°C, and markedly observable decline in milk yield start to be visible above 27°C (Johnson 1965). In this light, heat stress for dairy cows can be understood to indicate all high temperature related forces that induce adjustments occurring from the sub-cellular to the whole animal level to help the cow cope with physiological dysfunction and to better fit to environment (Yousef, 1985). The endeavour by homeotherms to stabilize body temperature within fairly narrow limits is essential to control biological reactions and physiological processes associated with normal metabolism (Shearer & Beede, 1990). In order to maintain homeothermy, an animal must be within thermal equilibrium which is further affected by factors like radiation, air temperature, air movement and humidity.

A study was, therefore, carried out in one of the tropical climatic set-up of Nepal, to assess the impact of summer heat on body physiology especially reflected by blood parameters and evaluate bathing as a rehabilitation strategy to overcome such stressful condition.

MATERIALS AND METHODS

The study area and period

The study was carried out during the summer months of 2015 (May-July) at experimental station of National Cattle Research Program (NCRP) in Rampur, Chitwan (27º39'N and 84º21'E), 10 km west of district headquarter, Bharatpur. Climatically, Chitwan harbours the tropical climate and yet is one of the most important dairy pockets of Nepal, largely due to the high demand of fluid milk, easy availability of feeds, straw, veterinary services favouring commercialization of dairy farming.
Sample size, design and treatment groups

All together sixteen lactating cattle were used for the study and fit into completely randomized block design (RCBD). Blocking was done on the basis of the parity of the animals selected for the study. The following list explains the treatment arrangement of the experiment.

Treatment group

- **T<sub>1</sub>**: Bathing once in a day (12:00 hours)
- **T<sub>2</sub>**: Bathing twice (12:00 and 15:00 hours)
- **T<sub>3</sub>**: Bathing thrice (9:00, 12:00 and 15 hours)
- **T<sub>4</sub>**: Control (no bathing)

Data collection and analysis

Meteorological data (maximum, minimum, dry bulb and wet bulb temperature in centigrade) were collected from inside the shed every morning at 9:00 hours. Likewise, vital signs were collected three times daily (at 9:00, 12:00 and 15 hours) every fortnight during the trial period. Any aberrations to the vital signs and animal behaving other than their normal behavior were closely monitored. Provisions were made in a way that any animal distressed due to the experiment will be treated and taken out of the experiment considering the study's alignment with the standard animal ethics protocol.

The blood samples were collected in vacutainer tube at morning time. Blood parameters sodium (Na), potassium (K), chloride (Cl), bicarbonate, Hemoglobin and Hematocrit were analyzed three times (0, 45 and 90 days) during the experiment at the laboratory in Bharatpur, Chitwan following the method described by Obitsu et al. (2011). The blood samples were collected in vacutainer tubes in mornings.

Collected data were assembled and processed and cleaned with the use of MS Excel (version 16.16.4) and differences among the means using different models has been carried out using SPSS™ Statistical Software (version 23). All the recorded data were subjected to analysis of variance and Duncan's multiple range test (DMRT) for mean separation. The significance differences among the means were tested using least significance difference (LSD) at 5% level of significance (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

**Microclimatic condition inside the cattle shed**

The climatic conditions of inside the shed were presented in table 1. In which the average of maximum, minimum, dry bulb and wet bulb temperature were found 35.36±0.25, 27.39±0.18, 30.69±0.16 and 26.07±0.15 °C respectively. These types of climatic condition are not suitable for the jersey cattle.
Hagiwara et al. (2002) reported that the environmental temperature at which the respiration rate started to increase was lower (17°C) for high-producing cows (>35 kg milk/day) than that (22°C) of low producing cows. The research studied that lactating dairy cows from European breeds, the thermos-neutral zone ranges between -5 and 25°C, and are called lower critical temperature (LCT) and upper critical temperature (UCT) respectively meaning animals require no additional energy above maintenance to cool or heat their body (Johnson, 1987).

**Table 1. Meteorological data of inside the cattle shed during the experimental period**

<table>
<thead>
<tr>
<th>Variable</th>
<th>No. of observation</th>
<th>Mean ± SD (°C)</th>
<th>Minimum (°C)</th>
<th>Maximum (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximum temperature</td>
<td>90</td>
<td>35.36±0.25</td>
<td>30.00</td>
<td>39.00</td>
</tr>
<tr>
<td>Minimum temperature</td>
<td>90</td>
<td>27.39±0.18</td>
<td>23.00</td>
<td>33.00</td>
</tr>
<tr>
<td>Dry bulb temperature</td>
<td>90</td>
<td>26.07±0.15</td>
<td>22.00</td>
<td>28.50</td>
</tr>
<tr>
<td>Wet bulb temperature</td>
<td>90</td>
<td>26.07±0.15</td>
<td>22.00</td>
<td>28.50</td>
</tr>
</tbody>
</table>

**Blood parameter of cross bred cattle**

**Sodium (Na) and Potassium (K) in cattle blood**

The sodium and potassium in blood were analyzed of the experimental animal of different treatment groups in three times (0, 45 and 90 days) during the research period which are presented in table 2. The results of sodium in cattle blood during the summer condition of different treatment group were found statistically non-significant. The result of next important blood parameter potassium was found mid time (40 days) statistically significant and other initial and final days result were found non-significant.

**Table 2. Effect of cold water bathing on blood Sodium and potassium values during summer**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Na (mmol/l) in days</th>
<th>K (mmol/l) in days</th>
<th>F value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>45</td>
<td>90</td>
</tr>
<tr>
<td>T1</td>
<td>144.25±1.03</td>
<td>145.88±0.95</td>
<td>145.70±2.72</td>
</tr>
<tr>
<td>T2</td>
<td>143.40±1.64</td>
<td>143.23±2.00</td>
<td>145.50±1.47</td>
</tr>
<tr>
<td>T3</td>
<td>144.00±1.06</td>
<td>144.75±1.30</td>
<td>143.65±0.97</td>
</tr>
<tr>
<td>T4</td>
<td>144.05±0.61</td>
<td>148.90±2.60</td>
<td>144.90±1.56</td>
</tr>
<tr>
<td>F value</td>
<td>0.10</td>
<td>1.60</td>
<td>0.21</td>
</tr>
</tbody>
</table>

Note: T1: Bathing once in a day (12 noon), T2: Bathing twice (12 noon and 3 pm), T3: Bathing thrice (9 am, 12 noon and 3 pm) and T4: Control (no bathing)

**Chloride and Bicarbonate in cattle blood**

The Chloride and Bicarbonate were analyzed in three times (on 0, 45 and 90 days) during the experiment. Results are presented in table 3 and both chloride and bicarbonate values across treatment did not show significant differences.
### Table 3. Effect of cold water bathing on blood Chloride and bicarbonate levels during summer

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Chloride (mmol/l) in days</th>
<th>Bicarbonate (mmol/l) in days</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>45</td>
</tr>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>107.70±1.08</td>
<td>103.95±0.79</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>106.00±2.04</td>
<td>102.33±0.48</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>104.98±1.99</td>
<td>106.80±1.64</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>104.26±0.83</td>
<td>105.66±1.47</td>
</tr>
</tbody>
</table>

F value | 0.85 | 2.98 | 0.15 | 0.27 | 1.94 | 0.37 |

Note: T<sub>1</sub>: Bathing once in a day (12 noon), T<sub>2</sub>: Bathing twice (12 noon and 3 pm), T<sub>3</sub>: Bathing thrice (9 am, 12 noon and 3 pm) and T<sub>4</sub>: Control (no bathing)

Coppock et al. (1982) was perhaps the first to report the effects of heat stress on electrolyte balance of dairy cows ultimately affecting the buffering capacity of the body and thereby the entire body physiology. However, as reported, the current study did not show any decline in the electrolyte levels both in terms of Chloride and Bicarbonate across all sampling days. Likewise, the excess drain of these salt has not been clinically evidenced during the experiment since excess draining are often associated with conditions like alkanosis or acidotic conditions (Afzaal et al., 2004). The other associated risks of kidneys reabsorbing or liberating the ions under excessive flushing of these electrolytes were not assessed (Gaynor et al., 1989).

### Haemoglobin (Hb) and Haematocrit (Ht) in cattle blood

The blood haemoglobin and haematocrit of the experimental animals with the interval of 45 days during the research period of the different treatment groups of normal water bathing in summer condition are presented in table 4. The results of Hb and Ht analysis also statistically were found non-significant.

### Table 4. Effect of cold water bathing on Haemoglobin (Hb) and Haematocrit (Ht) values in blood of cattle during summer

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Hb% in days</th>
<th>Ht % in days</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>45</td>
</tr>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>10.53±1.31</td>
<td>8.28±3.33</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>9.53±0.50</td>
<td>7.66±1.25</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>10.28±0.05</td>
<td>8.58±0.40</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>10.08±0.46</td>
<td>7.85±0.75</td>
</tr>
</tbody>
</table>

F value | 0.47 | 0.68 | 0.82 | 0.49 | 0.82 | 0.59 |

Note: T<sub>1</sub>: Bathing once in a day (12 noon), T<sub>2</sub>: Bathing twice (12 noon and 3 pm), T<sub>3</sub>: Bathing thrice (9 am, 12 noon and 3 pm) and T<sub>4</sub>: Control (no bathing)

Ali-Gholi et al. (2007) described that Ht value, Hb concentration and erythrocyte number rise, especially at the beginning of hot period, evidenced a rise of blood concentration. This hemocoencentration was initially induced by an increase in erythrocyte number, later by plasma dehydration (Obitsu et al., 2011; Lamp et al., 2015). Toharmat and Kume (1997) did not find any significant difference between Ht value and Hb concentration in hot and cool weather. In further work, however, Toharmat et al. (1998) described an increase in hematocrit values and hemoglobin concentration in summer. They also described a significant decrease in Ht value in.
dairy cows exposed to high temperatures. Ht and red blood cells decrease were perhaps caused by a rise in erythrocyte destruction, hemodilution effect could also participate here, because more water was transported in circulatory system for evaporative cooling. Another similar result was found that, cooling by sprinkling prevented marked changes in red blood picture, only non-significant increase in Ht value and erythrocyte number occurred at the beginning. However, it is also reported that cooling of heat stressed dairy cows increased blood hemoglobin concentration (Aboulnaga et al., 1989).

CONCLUSION

This study assessed the clinical and sub-clinical impacts of heat stress, among others, on the animal performances especially in terms of blood parameters. The results obtained from the study did not, however, confer to the recommendation and hypothesis that extremities of climate are expressed in terms of body physiology and can be measured through electrolyte balance in the blood. This could perhaps be due to the reason that either the animals have already been more or less suited to local harshness of summer or the temperature humidity index could perhaps be the right parameter to get more impressive results as part of the study (Ganter et al., 2011). On the other hand, the thermoneutral zone of different breeds vary and a more detailed study scoping the breed effect is also deemed essential.

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Author Contributions

T.P.P. designed and performed experiments, analyzed data and wrote the paper; B.R.A. helped during conducting the experiments and recording observations. D.B.K. and B.S.S. helped to revise the manuscript.

Conflicts of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

REFERENCES

