



Assessment of Summer Stress and Management with Poly Herbal Anti-Stressor Product (Restobal) in Buffaloes

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ABSTRACT

Variations in the different environmental parameters including temperature, wind velocity, pressure and relative humidity had influenced the bovines and reduce the feed intake further leads to the reduction in the milk production. The present study was conducted to evaluate the summer stress and to record the efficacy of polyherbal anti-stressor product (Restobal[®], M/s Ayurved Limited) against the management of summer stress in buffaloes with hyperthermia. Twenty buffaloes with hyperthermia were selected during the summer period and divided into two equal groups. Group I buffaloes (control group) with hyperthermia were treated for hyperthermia alone and buffaloes under group II (treatment group) was treated for hyperthermia along with the oral polyherbal anti stressors product. Whole blood and serum were collected on the 0th day, 3rd day and 5th day of treatment for laboratory analysis. Buffaloes in the treatment group showed the increased total leukocyte, neutrophil and lymphocyte count, elevated serum glutathione peroxidase levels and reduced serum cortisol levels when compared with the buffaloes in the control group. The present study concluded that polyherbal anti stressor product (Restobal which contains *Ocimum sanctum*, *Phyllanthus emblica*, *Mangifera indica* and *Withania somnifera*) is recommended as supportive therapy in the management of summer stress along with the treatment of hyperthermia in buffaloes.

Keywords: Stress, Summer, Buffaloes, Restobal, Management

Summer stress is one of the major influencing factors for the reduced production as well as to reproduction efficacy in animals. Due to variations in the environmental parameters including temperature, relative humidity and the solar radiation influences the vital physiological characters of the animals and reduces the feed intake further leads to the reduction in the milk production (Chaudhary *et al.*, 2015). Nardoen *et al.* (2010) stated that there will be a severe loss to the economy if we fail to adopt the different strategies to counteract the climatic

changes and climatic stress. Literature on the importance of the climatic changes in production of cattle was reviewed during summer (Chauhan and Ghosh, 2014). Studies on the assessment of the summer stress and its management were very limited in bovines. Therefore, the present study was formulated to record the alterations in the clinical, haematological changes during the summer stress and to evaluate the polyherbal anti-stressor product (Restobal[®], M/s Ayurved Limited) in the management of summer stress in buffaloes with hyperthermia.

MATERIALS AND METHODS

The present study was carried out at the College of Veterinary Science, Proddatur, YSR Kadapa District of Andhra Pradesh, India. Twenty buffaloes suffering from hyperthermia during the summer season were selected for the study analysis. Selected buffaloes were examined thoroughly and divided into two equal groups. Buffaloes in group I was treated for hyperthermia without any anti-stressor supplementation and serve as control group. Buffaloes in groups II were treated for hyperthermia along with oral herbal preparation (Restobal, Ayur Vet Ltd @ 50 ml orally BID for 5 days) and is considered as the treatment group. Both the group of buffaloes were treated for hyperthermia with injection oxytetracycline (20 mg/kg body weight, intravenously, BID) for three days, injection phenylbutazone and sodium salicylate, (20 ml/animal, intravenously once in a day) and oral rehydration solution containing electrolytes for two days. Whole blood and serum were collected on the 0th day, 3rd day and 5th day of treatment for laboratory analysis. Whole blood was utilised for the estimation of packed cell volume (PCV), haemoglobin (Hb), red blood cell count (RBC), white blood cell count (WBC) and absolute differential counts (DC). Serum was utilised for the estimation of cortisol and glutathione peroxidase levels as per the commercially available kit (Sivajothi and Reddy, 2017). Feed intake, rectal temperature, pulse rate and respiration rate, milk yield of the individual animal was

recorded before and after 30 days of treatment. Mean ambient temperature, humidity and atmospheric pressure was recorded during the study at the present geographical region. Data were presented as the mean \pm standard error (SE) and were subjected to statistical analysis using one-way analysis of variance by using SPSS version 20.0. If $P > 0.05$ considered as statistically not significant, $P \leq 0.05$ considered as statistically significant and $P \leq 0.01$ considered as statistically highly significant at 95% level of significance.

RESULTS AND DISCUSSION

Recorded environmental parameters was 18°C, 38°C, 32°C of temperature; 26%, 91%, 68% of humidity and 997 mbar, 1015 mbar, 1008 mbar of pressure (minimum, maximum and average values respectively). Buffaloes with environmental stress showed the elevated rectal temperature, heart rate, respiratory rate and reduction in the milk yield. Haematology revealed the elevated levels of haemoglobin, total erythrocyte count, packed cell volume, total leukocyte count, eosinophil and monocyte count; elevated serum cortisol levels than the mean values of the apparently healthy group of buffaloes. Findings of the study proved that buffaloes had environmental stress during the summer period. Recorded changes were mentioned in the Table 1. Clinical, haematological changes during the summer stress in association with the

Table 1: Clinical, haematological changes and stress parameters during the environment stress in buffaloes (Mean \pm S.E.)

Sl. No	Parameters	Apparently healthy buffaloes (N=10)	Buffaloes with Environment stress (N=20)	P - Value
1	Haemoglobin (g/dl)	11.27 \pm 0.13	13.49 \pm 0.60	0.001**
2	PCV (%)	36.50 \pm 1.1	45.60 \pm 1.30	0.001**
3	TEC $\times 10^6$ /cumm	7.45 \pm 0.09	9.01 \pm 0.66	0.001***
4	TLC /cumm	8378.0 \pm 236.1	9235.6 \pm 132.56	0.002**
5	Neutrophils /cumm	2555.6 \pm 101.22	2853.6 \pm 89.56	0.636 ^{NS}
6	Lymphocytes /cumm	5227.87 \pm 201.21	5505.7 \pm 173.7	0.069 ^{NS}
7	Monocytes /cumm	293.3 \pm 19.3	489.5 \pm 32.4	0.001**
8	Eosinophils /cumm	192.70 \pm 9.16	253.96 \pm 18.4	0.030*
9	Basophil /cumm	67.60 \pm 9.22	55.41 \pm 8.12	0.176 ^{NS}
10	Cortisol (nmol/L)	18.91 \pm 0.71	56.99 \pm 2.53	0.001**
11	Temperature (°F)	99.81 \pm 0.05	102.04 \pm 0.11	0.001**
12	Heart rate (/min)	63.75 \pm 0.94	88.80 \pm 1.24	0.001**
13	Pulse rate (/min)	63.05 \pm 0.73	87.95 \pm 1.12	0.001**
14	Respiratory rate (/min)	17.10 \pm 0.46	29.56 \pm 0.53	0.001**
15	Glutathione peroxidase (u/ml)	18.63 \pm 1.89	32.9 \pm 1.11	0.001**

Table 2: Mean milk yield (Litre/Day) in group I and group II buffaloes (Mean \pm S.E.)

0 th Day		5 th Day		30 th Day		P value
Control (Group I) (N=10)	Treatment (Group II) (N=10)	Control (Group I) (N=10)	Treatment (Group II) (N=10)	Control (Group I) (N=10)	Treatment (Group II) (N=10)	
2.86 \pm 0.19	2.66 \pm 0.24	2.73 \pm 0.31	2.79 \pm 0.18	4.41 \pm 0.49	5.16 \pm 0.69	0.025 *

Table 3: Table showing the changes in the vital, haematological and cortisol levels in control and treatment group of buffaloes (Mean \pm S.E.)

Sl. No.	Parameters	0 th Day		3 rd Day		5 th Day		P-value
		Control (Group I) (N=10)	Treatment (Group II) (N=10)	Control (Group I) (N=10)	Treatment (Group II) (N=10)	Control (Group I) (N=10)	Treatment (Group II) (N=10)	
1	Haemoglobin (g/dl)	13.4 \pm 0.25	13.64 \pm 0.22	13.5 \pm 0.2	13.71 \pm 0.17	13.79 \pm 0.23	13.89 \pm 0.18	0.456 ^{NS}
2	PCV (%)	45.6 \pm 0.94	43.8 \pm 0.2	44.3 \pm 1.26	44.5 \pm 1.4	44.8 \pm 1.12	45.3 \pm 1.21	0.766 ^{NS}
3	TEC $\times 10^6$ /cumm	8.37 \pm 0.16	8.18 \pm 0.2	8.14 \pm 0.15	8.16 \pm 0.17	8.19 \pm 0.14	8.18 \pm 0.11	0.965 ^{NS}
4	TLC /cumm	9378.8 \pm 115.6	9091.2 \pm 147.5	8972.2 \pm 285.5	9289.9 \pm 128.6	8766.1 \pm 299.6	9480.0 \pm 114.4	0.040*
5	Neutrophils /cumm	2738.4 \pm 210.56	2963.76 \pm 198.42	2610.8 \pm 105.4	2814.6 \pm 98.39	2726.3 \pm 98.65	2769.6 \pm 108.7	0.300*
6	Lymphocytes /cumm	5748.7 \pm 98.56	5463.7 \pm 132.46	5411.1 \pm 122.5	5619.8 \pm 109.34	5049.3 \pm 102.6	5962.9 \pm 101.3	0.000**
7	Monocytes /cumm	487.65 \pm 38.25	490.9 \pm 39.16	466.5 \pm 56.14	473.8 \pm 76.27	482.2 \pm 71.6	464.6 \pm 48.1	0.080 ^{NS}
8	Eosinophils /cumm	253.2 \pm 26.15	254.5 \pm 18.24	260.6 \pm 27.28	269.9 \pm 24.23	315.6 \pm 19.22	284.4 \pm 11.21	0.060 ^{NS}
9	Basophil /cumm	56.3 \pm 2.16	54.6 \pm 3.16	44.8 \pm 1.16	65.02 \pm 1.15	70.12 \pm 4.13	37.92 \pm 3.17	0.070 ^{NS}
10	Cortisol (nmol/L)	56.62 \pm 3.68	57.15 \pm 3.64	52.9 \pm 3.4	42.58 \pm 3.10	47.74 \pm 3.24	30.28 \pm 3.12	0.000**
11	Temperature ($^{\circ}$ F)	102.0 \pm 0.17	102.4 \pm 0.11	100.6 \pm 0.21	100.1 \pm 0.13	100.2 \pm 0.33	99.4 \pm 0.13	0.060 ^{NS}
12	Heart rate	88.2 \pm 1.64	89.4 \pm 1.9	82.6 \pm 1.34	79.3 \pm 1.5	75.6 \pm 0.94	71.3 \pm 1.7	0.040 *
13	Pulse rate	87.6 \pm 1.43	88.3 \pm 1.77	82.3 \pm 1.29	79.0 \pm 1.44	75.4 \pm 0.96	71.1 \pm 1.62	0.035 *
14	Respiratory rate	29.7 \pm 0.71	25.5 \pm 0.8	25.5 \pm 0.5	24.3 \pm 0.4	22.4 \pm 0.69	19.0 \pm 0.60	0.002**
15	Glutathione peroxidase (u/ml)	32.9 \pm 1.11	29.67 \pm 1.19	31.40 \pm 1.19	33.60 \pm 1.01	30.9 \pm 1.24	35.11 \pm 1.92	0.000**

previous studies in cattle and different breeds of buffaloes (Fagiolo *et al.*, 2004; Chaudhary *et al.*, 2015). Clinical parameters including respiration rate, heart rate and rectal temperature are indicative of climatic stress parameters. Due to the elevation of the rectal temperature 1 $^{\circ}$ C than normal causes the reduction in the performance in most of the animals (Fagiolo *et al.*, 2004). The elevated PCV levels and haemoglobin concentration was recorded in buffaloes with environmental stress and the increase in PCV might

be due to haemoconcentration and dehydration of plasma. The elevated levels of TLC might be indicative of changes in the immune system. Serum cortisol is suggestive of stress marker and levels of serum cortisol increased in the study, which is an indication of that the buffaloes had environmental stress (Reddy *et al.*, 2015). The recorded result of stress parameters during the summer stress in buffaloes was supported by the previous literature in cattle exposed to thermal stress (Yadav *et al.*, 2015).



Observed clinical, haematological findings, stress parameters and milk yield in group I and group II were (0th day, 3rd day, 5th day of treatment) mentioned in Table - 2 and 3. By the 5th day of therapy buffaloes in group II showed the significant variation from the buffaloes in group I by elevation of the total leukocyte, neutrophil and lymphocyte count, serum glutathione peroxidase levels; reduced serum cortisol levels. Recorded milk yield was less in the buffaloes during the period of summer stress. No significant variation was recorded in between the control and treatment group buffaloes during the time of trial period but, significant improvement in the milk production was recorded in the treatment group after the 30 days of therapy when compared to the control group buffaloes. During the stress associated with hyperthermia causes impairment of the intestinal barrier function which results in bacterial infections (Wheelock *et al.*, 2010). Cortisol is considered as the stress assessing hormone which secreted in higher levels during the stress response to the body. In the present study, stress might be due to hyperthermia during the environmental stress. Environmental stress causes the alteration of the immunity levels and makes animals more prone to the susceptibility to other disease conditions. Increased levels of serum glutathione peroxidase noticed in the group II buffaloes which indicative of presence of anti oxidants levels higher than the control group. In buffaloes of group II (treatment) showed the reduced levels of cortisol when compared to the group I (control).

Present study product Restobal[®] (M/s Ayurvet Limited) contains *Ocimum sanctum*, *Phyllanthus emblica*, *Mangifera indica* and *Withania somnifera*. Glycosides present in the *Withania somnifera* exhibited significant anti stress activity. Antioxidant activity assessed by free-radical scavenging enzymes, superoxide dismutase and glutathione peroxidase levels. Reduction in the level of enzymes leads to accumulation of the toxic oxidative free radicals which further result in degenerative changes in the cellular levels. If enzymes level increased it indicative of increased antioxidant activity which will protect the cells form stress (Bhattacharya and Muruganandam, 2003).

Ocimum sanctum has been traditionally used for its anti-stress effect (Bathala *et al.*, 2012). It modulates the humoral-immune response which leads to antibody production, the release of mediators of hypersensitivity and

it scavenges free radicals in the brain which causes induce increased nonspecific resistance. It will have cortisol sparing effect and act as on the central nervous system as a stimulant. Tabassum *et al.* (2010) stated *Ocimum sanctum* potentially regulates the HPA axis in stress-related disorders. *Phyllanthus emblica* knew in India and its anti stressor effect due to the presence of polyphenols, which will protect cell constituents against destructive oxidative damage from the oxidative stress and potent free radical scavengers (Saha and Verma, 2015). It had defensive antioxidant mechanisms and increases the levels of GSH, antioxidant capacity and activities of SOD, CAT, GSH peroxidase, GSH reductase and GSH Stransferase (Patel and Goyal, 2012). *Withania somnifera* is widely known as ashwagandha and it is a broad broad-spectrum remedy in India. It is considered as an anti-inflammatory and antioxidant herbal supplement (Mishra *et al.*, 2000). It shows the antioxidant activity by and regulation of antistress activities via the hypothalamic-pituitary-adrenal (HPA) axis. Adaptogens are herbs which will improve an individual's ability to with stand to the stress and *Ashwagandha* is one of the adaptogen (Chrousos, 2009).

During the initiation of stress, it induces different autonomic, visceral, immunological, and neurobehavioral responses and activates the hypothalamic-pituitary-adrenal axis (HPA) which results in elevation of serum corticosterone levels. The present study recommended the polyherbal anti stressor product (Restobal) as supportive therapy in the management of summer stress along with the treatment of hyperthermia in buffaloes.

CONCLUSION

In conclusion, polyherbal anti stressor product Restobal[®] (M/s Ayurvet Limited) is recommended as a supportive therapy for the management of summer stress in buffaloes with hyperthermia during the summer season.

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