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Article · November 2018

DOI: 10.18805/ijar.B-3497

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B-3497
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Indian J. Anim. Res.,
Print ISSN:0367-6722 / Online ISSN:0976-0555

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Physiological response to transportation stress at different flocking densities in hot humid and winter seasons in goats and assessment of pretreatment of Vitamin C, Electrolyte and Jaggery

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Received: 23-09-2017 Accepted: 30-12-2017 DOI: 10.18805/ijar.B-3497

ABSTRACT

Present study was conducted to investigate the seasonal effects of transportation of goats (Alpine x Beetle) at different flocking densities, supplemented with Vitamin C in group I, Vitamin C + Electrolyte in group II and Jaggery in group III, 3 days before transport of animal, during winter and hot-humid seasons. The goats were selected from LRC, NDRI Karnal and were of 10-12 months old. Each group consisted of 25 goats each, divided into high (15) and low (10) flocking densities, transported for 8h with average speed of 25 Km/h. All the animals were kept off-feed and deprived of water during the transportation period. Physiological responses (Respiration rate, Rectal temperature, Pulse rate and Skin temperature) were recorded before and after transportation. A significant increase ($P<0.05$) in ST, RT, RR and PR were recorded just after unloading in both the flocking density groups and during both seasons which then declined to basal values ($P<0.05$) within 6-12 hours post transportation. Supplementation with Vitamin C, Vitamin C + Electrolyte and Jaggery aided in reducing transportation stress but Vitamin C + Electrolyte combination proved more beneficial in alleviating transportation stress in the goats.

Key words: Electrolyte, Flocking density, Jaggery, Physiological response, Transportation stress, Vitamin C.

INTRODUCTION

Livestock animals are transported from place to place for various reasons which include marketing, slaughtering for meat in abattoirs or replacement of old stocks, exhibitions, fair, sports competition, seasonal tourism, taming, reproduction and health status and from drought-affected areas to better grazing sites etc, which is an inevitable husbandry practices (Fazio and Ferlazzo, 2003) and there is a need for quantitative assessment of their welfare during transportation. Transportation also induces changes in the blood composition as well as other bodily parameters like HR, RR and skin quality (Gregory, 2007). Stress is a common factor responsible for various physiological alterations in the body that indirectly alter the immune system through production of reactive oxygen species (ROS) leading to decreased endurance and increased susceptibility to various infections and reduces overall productivity and fertility leading to economic losses to the farmers. Moreover pre-slaughter stress also affect adversely meat quality (Broom, 2000).

During transportation the physiological alterations primarily includes electrolyte imbalance, increased RR and HR, dehydration, energy deficit, catabolism (Das *et al.,* 2000). The animals should be treated before or after the journey so as to overcome the various physiological changes during transportation and potentiate the immune system to resist the infections. Physiological parameters like RR, PR, RT and ST gives a reliable response of animal welfare status during transportation and consequently the level of discomfort/ comfort to the animal (Bianca, 1965). Road transportation for 12h resulted in variable and statistically significant increases in HR, RR, PR and RT (Ambore *et al.,* 2009). RT is an indicator of thermal balance and adverse effect of thermal environment affect growth, lactation, and reproduction of dairy cows (Hansen and Arechiga,1999). Various reports suggest significant increase in RT after transporting different animals (Minka and Ayo, 2009; Kassab and Mohammed, 2014). RT is the most common indicator of body temperature and is considered a reflection of core body temperature (Al-Tamimi, 2007). Both RT and ST increases after transportation in goats, but RT is a better indicator (Al-Badwi *et al.,* 2012). RR indicate heat stress in a hot environment and this give significant correlation with circulating corticoid concentration. PR reflects primarily the homeostasis of the

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animal body along with the general metabolic status (Kumar and Phrommathed, 2005; Kassab and Mohammed, 2014).

Ascorbic acid (AA) is a potent antioxidant that is known to affect mood, and it is the firstknown vitaminergic neurotransmitter (Balz, 2003). It has been established that the administration of AA is beneficial to humans, animals, and poultry under stress situations (Minka and Ayo, 2010; Clero and Grandjean, 2012). Although ruminants may not require AA supplementation in the diet under normal conditions however during stress the requirement of the body for AA exceeds the synthetic capacity of the liver (Lykkesfeldt and Svendsen, 2007). Besides, stress factors are known to deplete adrenal AA (Balz, 2003; Lykkesfeldt and Svendsen, 2007; Gade *et al.*, 2010). Therefore, AA supplementation at the point of stress may provide a potentially important, cheap, and non-toxic alternative treatment to the animal. Electrolyte and fluid supplementation has been shown to be effective in reducing the detrimental effects of transportation stress in cattle (Schaefer *et al.* 1997). Jaggery is nutritious sugarcane product and easily available and rich source of sucrose besides containing good amount of vitamins and minerals (Singh, 2013).

The present study was aimed to assess the effect of Vitamin C (Ascorbic acid) alone, with electrolyte, and jaggery alone supplementation when goats are transported at different flocking densities in tropical region at different seasons.

MATERIALS AND METHODS

Experimental site and thermal environment conditions: Animal experimentation was performed in compliance with regulations set by the Livestock Research Council (LRC), National Dairy Research Institute (NDRI), India and approved by the Institutional Animal Ethics Committee (IAEC). The site is situated at an altitude of 250 meters above sea level and at 29 º42' N latitude and 77º 02' E longitude. The maximum temperature recorded goes up to 45º C in summer and minimum temperature 3.5º to 4º C in winter. The average rainfall is about 700 mm. The experiment was conducted in two seasons: hot humid season (September-October) and winter season (December-January).

Animals and experimental plan: Cross bred goats (Alpine x Beetal) of 10-12 months of age were divided into four groups obtained from LRC, NDRI Karnal. Group I, II and III consisted of 25 goats divided into high (15) and low (10) flocking densities 0.14 mt^2 (lfd) and 0.20 m^2 (hfd) per goat of 20-25 kg respectively, continuous 8 hours transport in a tractor trailer with minimum speed of 25 km/h for 3 days. During the journey all the animals were kept off-feed and deprived of water.

Group I was fed vitamin C at a dose of 180 mg/kg.bwt/day/animal orally. Group II was fed with vitamin C + electrolyte (180 mg/kg.bwt/day/animal of vitamin C + 7 g / animal / day Electral powder) and group III was fed jaggery at dose of 200 g/animal/day orally with small amount of concentrate, 3 days before the start of experiment.

Environmental parameters: These parameters indicates study were conducted in hot humid area in india. Minimum and maximum ambient temperatures, dry bulb and wet bulb temperatures were recorded with respective thermometer for microenvironment of the experimental goats inside shed and vehicle at the time of sampling and recording physiological responses. The THI was calculated from dry bulb and wet bulb temperatures using the equation: THI = 0.72 x (C_{db} + C_{wb}) + 40.6 (McDowell *et al.*, 1976). Daily relative humidity was calculated by difference of dry bulb temperature and wet bulb temperature.

Physiological responses: All the physiological responses measured on before loading, after loading, 6h, 12h, 24h and 48h post-transportation were taken as follows:

1. Rectal Temperature: RT of each animal was recorded by inserting digital thermometer about 3 cm into the rectum for 1 minute ensuring the probe touches the rectal mucosa. The results were expressed in °F.

2. Respiration rate: RR was recorded by observing the flank movement for one minute.One outward and inward movement was counted as one respiration and the results were expressed as breaths per minute.

3. Pulse rate: PR of goats was measured by feeling pulsation of femoral artery and the results were expressed in beats per minute.

4. Skin or peripheral temperature: The ST was measured at three different sites on the body viz. back, head and leg using infrared Tele-thermometer (Raytek, Model Raynger ST2L, M/s. Surrey Scientific, Surrey, U.K.) by keeping it 2-3 inches away from the surface of the desired site. The results were expressed in °C.

Statistical analysis: Data analysis was carried out using SAS 9 software licensed to NDRI. Mean values at different sampling times were compared with respective basal mean values of each group using one way ANOVA with post-test as Dunnets multiple comparison.

RESULTS AND DISCUSSSION

Meteorological data in the month of Sep-Oct and Dec-Jan THI was 77.14 and 58.74 respectively. This suggest a hot humid (HH) and winter seasons in Karnal, India. Maximum values of RT, ST, RR, PR were recorded just after unloading in both density groups and during both seasons. The values of RT remained high 12 h post transportation. Average values physiological parameters RT, RR (presented in Table 1, 2) and ST, PR (presented as figure 1, 2) obtained in all the three groups of goats at low and high floking density, during winter and hot-humid seasons and the analysis of variance in Table 3. In groups I and II, during winter season, higher values of RT ($P<0.05$) were observed in goats in lfd

Table 1: Average values of rectal temperature (^{0}F) in goats transported at different flocking densities during winter and hot humid seasons.

Transportation / Groups	WINTER		HOT HUMID	
	Low Flocking Density	High Flocking Density	Low Flocking Density	High Flocking Density
Vitamin C (Group I)				
Before Loading	100.96±0.10^{bx}	99.96±0.25^{aw}	102.66±0.03^{cz}	102.19±0.02^{ay}
After Unloading	102.07±0.19^{fw}	102.25±0.17^{ew}	103.79±0.02^{gx}	103.77±0.02^{fx}
6h Post	102.12±0.03^{fx}	101.88±0.03^{ew}	103.10±0.02^{ey}	103.12±0.02^{dy}
12h Post	101.47±0.02^{cw}	100.63±0.19^{bx}	102.65±0.02^{cy}	102.63±0.03^{by}
24h Post	101.15±0.02^{cx}	99.97±0.06^{aw}	102.62±0.03^{cz}	102.32±0.05^{by}
48h Post	100.88±0.03^{bx}	99.88±0.05^{aw}	102.52±0.03^{cz}	102.18±0.06^{ay}
Vitamin C and Electrolyte Mixture (Group II)				
Before Loading	100.64±0.19^{ax}	99.73±0.25^{aw}	102.64±0.03^{cy}	102.63±0.03^{by}
After Unloading	101.86±0.14^{fw}	101.83±0.10^{cw}	103.76±0.03^{gx}	103.64±0.10^{bx}
6h Post	102.00±0.05^{fw}	101.97±0.05^{dw}	103.07±0.04^{ex}	103.08±0.04^{dx}
12h Post	101.42±0.01^{cx}	101.18±0.03^{cw}	102.87±0.03^{dy}	102.88±0.03^{cx}
24h Post	100.68±0.03^{aw}	100.68±0.03^{bw}	102.60±0.03^{cx}	102.58±0.03^{bx}
48h Post	100.58±0.02^{ax}	99.68±0.02^{aw}	102.50±0.01^{cy}	102.52±0.02^{by}
Jaggery Solution (Group III)				
Before Loading	101.76±0.06^{dw}	101.78±0.08^{cw}	102.35±0.07^{bx}	102.34±0.05^{bx}
After Unloading	102.81±0.01^{ew}	102.84±0.01^{ex}	103.42±0.03^{fy}	103.48±0.02^{ez}
6h Post	101.82±0.04^{ey}	101.68±0.07^{cx}	102.32±0.11^{bw}	103.10±0.04^{dz}
12h Post	101.88±0.04^{cx}	101.75±0.11^{cw}	102.05±0.07^{ay}	102.57±0.08^{bz}
24h Post	101.58±0.06^{dw}	101.68±0.07^{cx}	102.30±0.04^{by}	102.42±0.06^{bz}
48h Post	101.63±0.08^{dx}	101.73±0.09^{cy}	102.22±0.10 ^{bz}	102.47±0.06^{bw}

Between rows a, b, c and d and between columns w, x, y and z differed significantly (P<0.05)

Table 2: Average values of respiration rate (breaths per minute) in goats transported at different flocking densities during winter and hot humid seasons.

Transportation / Groups	WINTER		HOT HUMID	
	Low Flocking Density	High Flocking Density	Low Flocking Density	High Flocking Density
Vitamin C (Group I)				
Before Loading	23.11±0.20^{cx}	20.94±0.21^{aw}	28.00±0.20^{bz}	24.00±0.40^{ay}
After Unloading	29.94±0.19^{fx}	28.83±0.29^{gw}	42.83±0.32^{gy}	46.72±0.18^{ez}
6h Post	24.50±0.12^{ex}	22.67±0.18^{cw}	29.00±0.20^{dz}	25.33±0.18^{by}
12h Post	23.67±0.11^{dx}	22.17±0.17^{cw}	28.33±0.27^{cz}	25.17±0.43^{by}
24h Post	23.33±0.18^{cx}	21.17±0.17^{bw}	23.67±2.57^{ax}	24.33±0.41^{ax}
48h Post	22.83±0.17^{cx}	20.83±0.17^{aw}	27.83±0.26^{bz}	24.17±0.22^{ay}
Vitamin C and Electrolyte Mixture (Group II)				
Before Loading	23.00±0.20^{cw}	23.06±0.19^{cw}	24.39±0.42^{ax}	28.11±0.21^{cy}
After Unloading	29.00±0.20^{fx}	27.00±0.20^{fw}	40.89±0.18^{fy}	47.78±0.17^{fz}
6h Post	24.17±0.29^{ew}	24.00±0.20^{ew}	25.50±0.34^{ax}	28.83±0.17^{cy}
12h Post	23.50±0.12^{cw}	23.50±0.23^{dw}	25.33±0.39^{ax}	28.50±0.19^{cy}
24h Post	23.17±0.26^{cw}	23.17±0.17^{cw}	24.67±0.33^{ax}	28.17±0.33^{cy}
48h Post	22.83±0.17^{cw}	22.83±0.35^{cw}	24.00±0.14^{ax}	28.00±0.28^{cy}
Jaggery Solution (Group III)				
Before Loading	22.78±0.29^{cw}	22.17±0.38^{cw}	24.28±0.24^{ax}	24.50±0.32^{ax}
After Unloading	32.33±0.24^{gw}	32.44±0.24^{hw}	35.50±0.10^{ex}	36.61±0.15^{dy}
6h Post	23.00±0.31^{cx}	22.33±0.36^{cw}	23.67±0.27^{ay}	24.50±0.23^{az}
12h Post	22.00±0.20^{bw}	22.17±0.47^{cw}	24.17±0.17^{ay}	23.83±0.29^{ax}
24h Post	21.50±0.31^{aw}	22.33±0.18^{cx}	24.33±0.23^{ay}	25.17±0.33^{bz}
48h Post	23.33±0.23^{cx}	21.50±0.31 ^{bw}	23.83±0.29^{ay}	24.11±0.16^{az}

Between rows a, b, c and d and between columns w, x, y and z differed significantly (P<0.05)

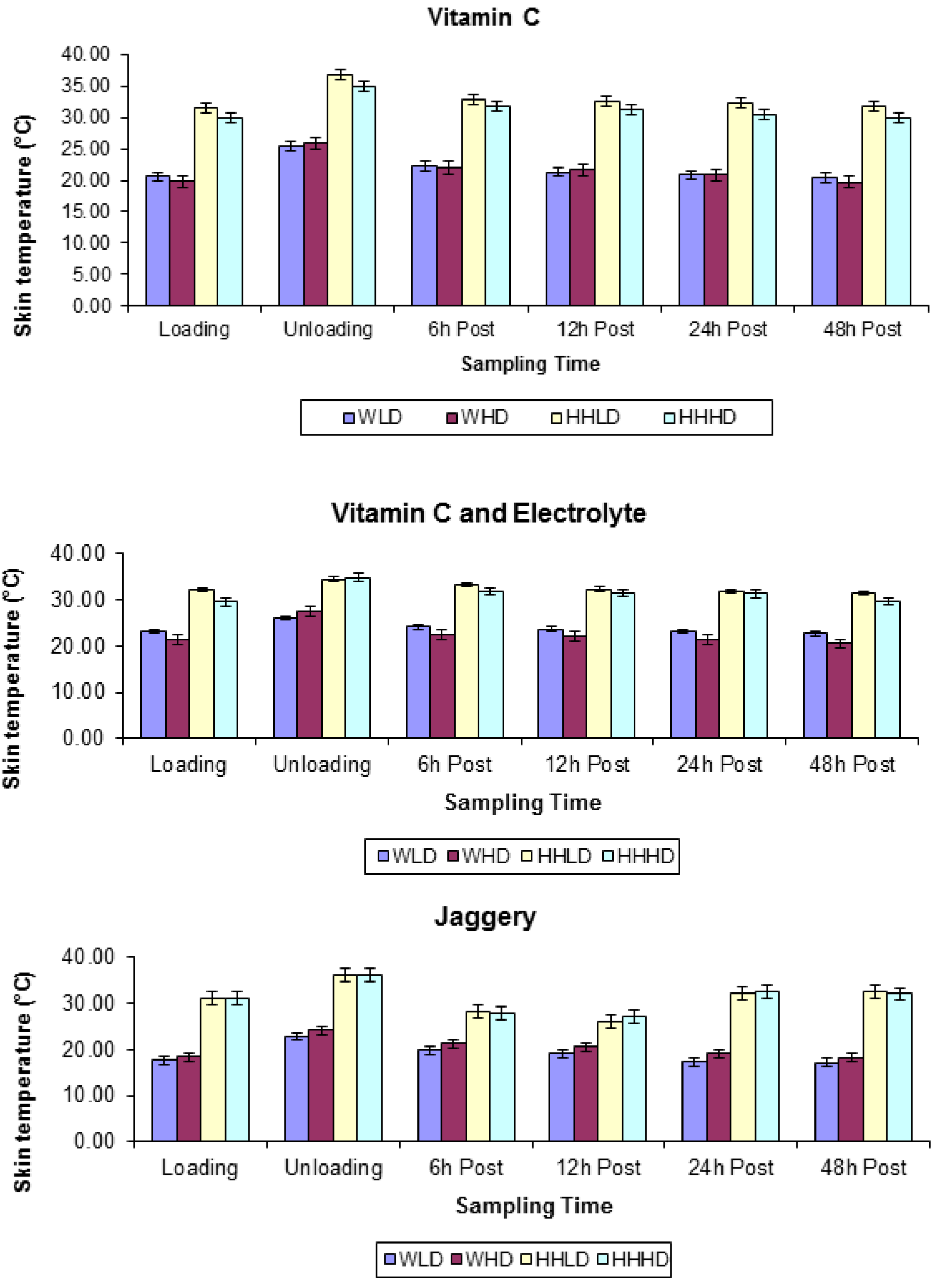

Fig. 1: Average values of skin temperature (^{0}C) in goats transported at different flocking densities during winter and hot humid seasons.

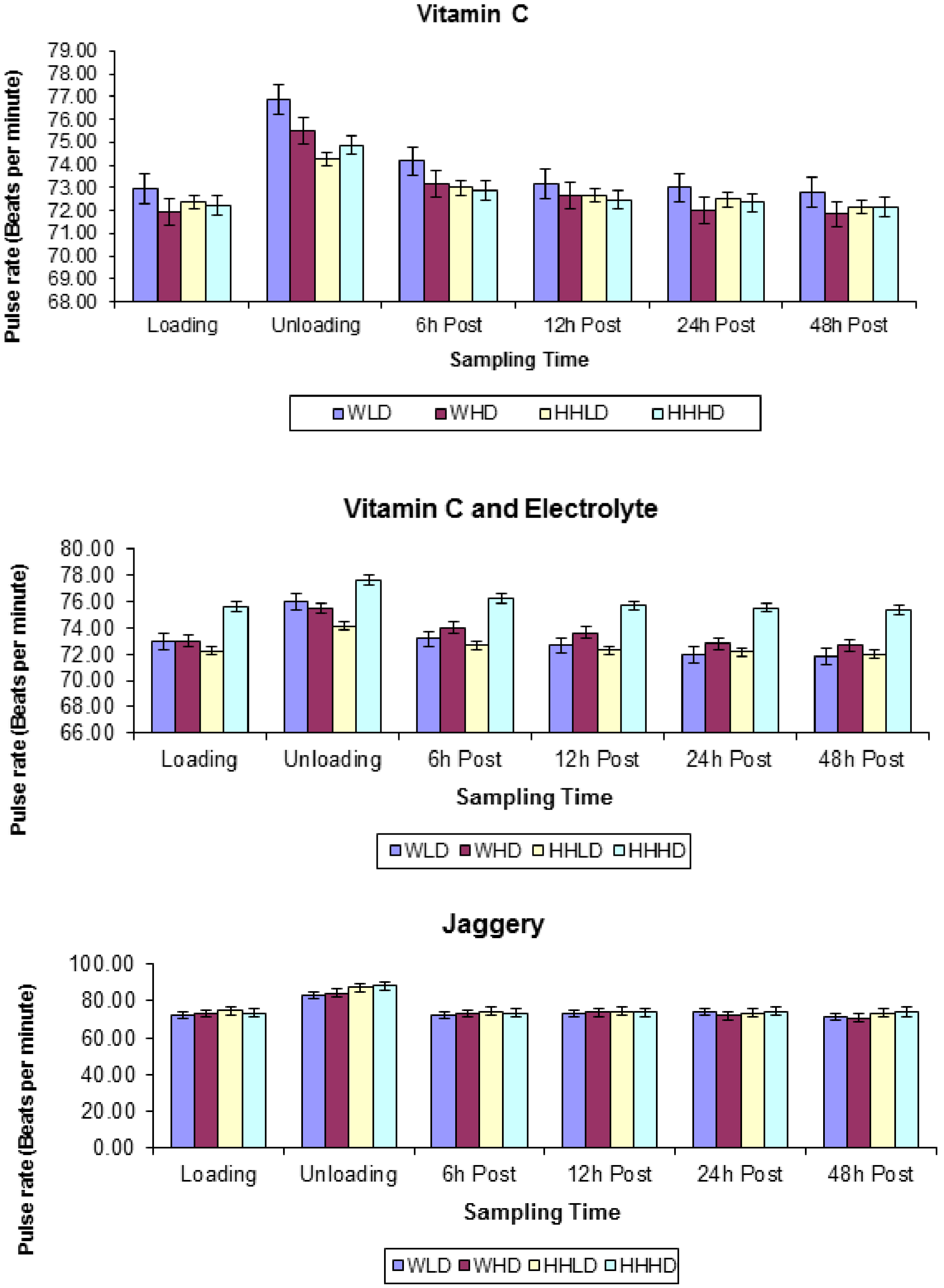

Fig. 2: Average values of pulse rate (beats per minute) in goats transported at different flocking densities during winter and hot humid seasons

in comparison to hfd goats. Although values of RT in group II in both flocking densities during HH season were significantly higher than winter season. In group III, lower RT values were observed in winter season in both flocking densities as compared to HH season. The overall variation of 3.16 °F in RT was observed between the groups in both flocking densities during winter and hot humid seasons. Less significant increase of RT in Vitamin C supplemented group (I and II) can be due to fact that Vitamin C and Electrolyte directly alters thermal set point by decreasing prostaglandin output, especially PGE series (Hadden *et al.,* 1987), which increases during stress and has direct effect on hypothalamic thermoregulatory zone (Ganong, 2001).

The increase in RT due to transportation in goats is corroborated by other workers (Minka *et al.,* 2009; Zulkifi *et al.,* 2010). The insignificant increase of RT in vitamin C supplemented groups is also supported by the study of Kassab and Mohammed (2014) in sheep. They reported insignificant increase of RT in vitamin C supplemented group as compared to control group when transported for 3 hours.

Higher values of ST ($P<0.05$) were observed in goats with lfd in comparison to hfd goats during winter season in group I. A similar decreasing trend was also observed for goats in HH season. In winter season, lfd goats in group II and hfd goats in group III, showed higher values ($P<0.05$) of ST. Maximum variation of 5-6°C in ST was observed in groups I and III goats at unloading as compared to pre loading values in both flocking densities during winter and HH seasons.

In the present study the lower ST during winter season indicated that the peripheral circulation was minimum due to vasoconstriction of blood vessels in the skin which did not allow the heat transfer from the core body to peripheral area. Das et al. (2000) reported a significantly positive correlation between air temperature and ST during transportation. Higher ST in goats indicated that the heat transfer process from the core body to peripheral area was actively operating, as blood vessels in the skin vasodilated during hot conditions mainly to bring body heat to the skin surface for dissipation of heat by radiation and convection (Bianca, 1965).

The respiration rate of goats in HH season were significantly ($P<0.05$) higher than in winter in both the flocking density groups. During HH season, maximum value of respiration rate 46.72±0.18 breaths/min was observed at unloading in hfd group as compared to lfd (group I). The values of RR remained elevated 24 hours post transportation. Moreover in groups I and II, during winter season, higher values of RR ($P<0.05$) were observed in goats with lfd in comparison to hfd goats whereas in HH season the values fluctuated. The RR in HH season between two densities varied significantly ($P< 0.05$) and the highest value of 47.78 ±0.17 breaths/min was reported in hfd goats after unloading. The values of RR in group II in both flocking densities during HH season were significantly higher than the winter season. In group III, lower RR were observed in winter season in both flocking densities as compared to HH season.

The PR in hot humid season in group II, between two densities varied significantly ($P< 0.05$) with highest value of 77.65 ±0.12 beats/min been reported for hfd goats after unloading. In comparison with the other groups, the values of PR in group I wasthe lowest ($P<0.05$) in both the flocking densities and seasons. The values of PR ranges from 71.00±0.67 to 88.10±0.22 beats/min in both flocking densities during winter and HH seasons. The maximum value of PR with76.89±0.20 beats/min was reported at unloading in hfd group as compared to lfd group during hot humid season. In groups I and II, in winter season, higher values of PR ($P<0.05$) were observed in goats with lfd in comparison to hfd.

RR and PR have been used to determine comfort and adaptability of animals to adverse condition and they reflect a degree of stress imposed on animals by transportation. Road transportation for 12h resulted in variable and statistically significant increases in RR and PR in goats but after 5 days of transportation no significant difference was observed (Ambore et al., 2009). Respiration is a physiological response in which transport of oxygen from the outside air to the cells within tissue and the transport of carbon dioxide in the opposite direction takes place (Nilsson *et al.,* 2010). In mammals, respiration is directed to eliminate CO_2 from the tissues of the body with the provision of O_2 under thermo-neutral conditions and there is evaporation of moisture from the respiratory tract in order to prevent hyperthermia under high ambient temperature (Marai *et al.,* 2007). The PR reflects primarily the homeostasis of circulation along with the general metabolic status. Increased heart rate on exposure to high environmental temperature increases the blood flow from the core to the surface for more heat to be lost by sensible (loss by conduction, convention and radiation) and insensible (loss by diffusion of water from skin) means**.** RR increased significantly after transportation in sheep (Kassab and Mohammed, 2014) and in camel (Khasmi *et al.,* 2013) in control group and compared to ascorbic acid administered group and camels. RR indicates heat stress in hot environment and this results into significant correlation with circulating corticoid concentration (Kumar, 2005) There was significant increase in RR in camels ($P < 0.05$) after transportation (Khasmi *et al.,* 2013). When comparing different seasons, maximum RR was found during HH season and lowest during winter season. Normal respiration rate was not attained by all groups of goats during all seasons post transportation and there was a significant difference in respiratory rate 48 hours post transportation except in group II goats that were supplemented with vitamin C and electrolyte. This is an indication that group II

Table 3: Analysis of variance for physiological reactions in transported Goats.

Source of variation	Df	Mean Sum Squares				
		Peripheral Temp.	**Pulse Rate**	**Respiration Rate**	**Rectal Temperature**	**F crit.**
Between Seasons	1	32638.40**	198.34**	5963.27**	576.27**	3.85
Between Densities	3	10907.82**	136.82**	2102.75**	195.96**	2.61
Between Treatments	17	328.40**	654.37**	1512.73**	21.06**	1.63
Season x Treatments	17	98.01*	29.18*	331.49*	7.80*	1.63
Densities x treatments	51	43.90*	22.31*	138.51*	3.50*	1.36
With in	1206	1.26	1.95	2.82	0.10	
Total	1295	-	-	-	-	

P< 0.01**

supplemented goats recovered basal respiration values earlier than the other transported animal groups.

Analysis of variance (Table 3) for rectal temperature, skin temperature, respiration rate and pulse rate indicated that there was a significant difference (P<0.05) between seasons, between density and between groups.

CONCLUSION

Transportation of goats upto 8 hours during HH and winter seasons induced stress as is reflected by increased values of ST, RT, RR and PR just after unloading. Supplementation of vitamin C + electrolyte showed lesser deviations from the normal values as compared to other groups, indicating that a combination of vitamin C and electrolyte was more beneficial in reducing the transportation effect.

ACKNOWLEDGEMENT

The authors are thankful to Head, Animal physiology Division and Director, NDRl, Karnal for providing necessary facilities for carrying out this study.

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