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ARTICLE



## Minerals response to transportation stress at different flocking densities in hot humid and winter seasons and the assessment of ameliorative effect of pretreatment with vitamin C, electrolyte and jaggery in goats

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### ABSTRACT

The present study evaluated the seasonal effects of transportation of goats (Alpine × Beete) 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 Livestock Research Centre (LRC), National Dairy Research Institute, 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 8 h with an average speed of 25 km/h. All the animals were kept off-feed and deprived of water during the transportation period. Blood samples were taken just before transportation, immediately after transportation, 6 h, 12 h, 24 h and 2 days post-transportation from all the three groups. The blood samples were further analysed for estimation of different minerals (Na, K, Cl<sup>-</sup>, P and Mg). In both the seasons and in both the flocking density groups, higher pre-transportation values ( $P < 0.05$ ) of Na, K, Mg and Cl<sup>-</sup> were observed whereas P values post transportation ( $P < 0.05$ ) were higher in hot humid season in all the treated groups and also in group III. Except for P, minimum values ( $P < 0.05$ ) of all other minerals were recorded just after unloading in both the density groups and in both the seasons, which then increased to basal values ( $P < 0.05$ ) after 12–24 h of 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.

**Abbreviation:** fd: flocking density; Na: sodium; K: potassium; Cl<sup>-</sup>: chloride; P: phosphorous; Mg: magnesium, HH: hot humid; AA: ascorbic acid; lfd: low flocking density; hfd: high flocking density; LRC: Livestock Research Centre

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## Introduction

The intensity and specialization of livestock production and the demand for livestock to be marketed and slaughtered outside places where they are being produced have necessitated transportation of different domestic animal species all over the world (Minka and Ayo 2007). Transportation of food animals is of great concern due to several reasons (Hartung 2003). Stressful transportation may affect adversely meat quality, the risk of spread of infectious diseases and causes injury, reduce performance and cause increased morbidity and mortality rate. Flocking/stocking density during livestock transport is one of the main factors that influence welfare and comfort. From an economic point of view, flocking/stocking densities can increase or reduce unit costs of operation (De la Fuente et al. 2010).

Changes in mineral metabolism triggered off by an adjustment in the original hormonal status as a result of environmental stress factors brought about during animal transportation involves mainly calcium, magnesium, sodium, potassium and chloride (Schaefer et al. 1997; Montane et al. 2002; Parker et al. 2003). The way transport stress affects mineral metabolism chiefly sodium, potassium, chloride and calcium was classified by Klaus-Dietrich (1985). He noted that stress-induced cell stimulation causes an aggressive potential change of the cell from the rest potential to the action potential.

Parker et al. (2003) observed no significant increases in calcium and phosphate ions in both control and experimental groups during animal transportation. There was a trend for the plasma concentration of phosphate ions to be higher in the feed-deprived group than in the control. Galyean et al. (1981) reported plasma phosphate ion concentration to be higher in fasted and transported animals for 32 h. The concentration of magnesium in body tissue decreases during transportation, which led to a change in the activity of mitochondrial membrane of cells (Klaus-Dietrich 1985).

The energy exchange of skeletal and heart muscle is seriously affected by lack of magnesium. Stress together with lack of magnesium causes an increase in the synthesis and release of catecholamines, resulting in an increase in cell permeability (Davidson et al. 2004).

Similarly, plasma concentrations of Ca, Na, Cl<sup>-</sup> and P were observed to remain within the normal values in transported cattle, steer and calves (Atkinson 1992; Parker et al. 2003). However, the value of K was observed to decrease below normal. The decrease may be due to the fact that the stresses induced activation of the hypothalamic-pituitary-adrenal axis and stimulated the secretion of cortisol, resulting in the excretion of potassium (Parker et al. 2003).

(Mg) acts as a co-factor or an activator of many critical enzymes for the reactions involving ATP that energize all major metabolism pathways. Also, magnesium acts as a sedative that reduces the stress induced catecholamine secretion and the inhibited glycogen breakdown (Steinhardt and Hans-Herma 1998).

Mir (2015) found that blood sodium, potassium and magnesium concentration decreased whereas phosphorus concentration increased after transportation during hot humid (HH), winter and hot dry seasons in goats. Steinhardt and Hans-Herma (1998) showed a significant lower concentration of Mg and Fe, in transported calves that were 60 days old and a lower concentration of Ca and P in the blood of calves under physical stress. Even though cattle are known to have a substantial buffering capacity, transportation and handling stress were reported to cause significant changes in electrolyte balance, which includes chloride, potassium, calcium and magnesium (Schaefer et al. 1997).

Non-significant differences in the values of sodium, potassium and chloride ions post-3-h transportation in ascorbic acid supplemented and non-supplemented sheep indicated that ascorbic acid administration as well as 3-h road transportation did not affect these important ions in sheep (Kassab and Mohammed 2014).

The present study investigated the comparative ameliorative effects of vitamin C, vitamin C + electrolyte and jaggery after loading, unloading after transportation on some plasma minerals in goats raised extensively in a tropical country during HH and winter season.

## **Materials and methods**

### ***Experimental site and thermal environment conditions***

Animal experimentation was performed in compliance with regulations set by the Livestock Research Centre (LRC), National Dairy Research Institute (NDRI), India, and approved by the Institutional Animal Ethics Committee. The site is situated at an altitude of 250 m above sea level and at 29° 42' N latitude and 77° 02' E longitude. The maximum temperature recorded goes up to 45°C in summer and minimum temperature from 3.5°C to 4°C in winter. The average rainfall is about 700 mm. The experiment was conducted in two seasons: HH 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 three groups obtained from LRC, NDRI, Karnal. Groups I, II and III consisting of 25 goats were divided into high (15) and low (10) flocking densities 0.14 m<sup>2</sup> (lfd) and 0.20 m<sup>2</sup> (hfd) per goat of 20–25 kg, respectively, and were subject to continuous 8 h 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.body weight/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 amounts of concentrate, 3 days before the start of experiment.

### ***Sampling procedure***

The animals allotted for experimentation were housed at the same place 5 days before the start of transportation trailer during which one blood sample was taken to record the basal values of all the animals. Blood samples were taken just before transportation, immediately after transportation, 6 h, 12 h, 24 h and 2 days post-transportation in heparinized vacutainer tubes and immediately placed in ice packs and brought to the laboratory. Then, blood samples were centrifuged at 2500 rpm for 20 min to separate plasma. Then, the plasma samples were stored in 2 ml microvials and labelled and then stored at –20°C till they were analysed.

### ***Plasma sodium and potassium estimation***

Sodium and potassium were estimated in plasma samples by using Liquimax Sodium (Na+) Potassium (K+) (optimized) colorimetric method kit purchased from Avecon Healthcare Pvt. Ltd.

Sodium is estimated by colorimetric Method based on Modified Maruna and Trinders Method. Sodium and proteins are precipitated together by magnesium uranyl acetate as uranyl magnesium sodium acetate. Excess of uranyl salt reacts with potassium ferrocyanide to produce a brownish colour. The intensity of the colour is inversely proportional to the sodium concentration in the specimen and is measured photometrically at 505 nm (490–550).

Potassium is estimated by turbidometric method. Potassium ions present in the specimen react with sodium tetra phenyl boron (Boron reagent) to produce an insoluble potassium tetra phenyl boron resulting in a turbid suspension. The extent of turbidity is proportional to the potassium concentration and is measured photometrically at 578 nm (570–620).

### ***Plasma phosphorous estimation***

Phosphorous was estimated in plasma samples by using Liquimax Phosphorous-SLR (molybdate-UV method) kit purchased from Avecon Healthcare Pvt. Ltd.

Inorganic phosphate forms an ammonium phosphomolybdate complex having the formula  $(\text{NH}_4)_3 [\text{PO}_4 (\text{MoO}_3)_{12}]$  with ammonium molybdate in the presence of sulphuric acid. The complex is determined photometrically in the ultraviolet region (340 nm).

### ***Plasma magnesium estimation***

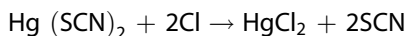
Magnesium was estimated in plasma samples by using Liquimax Magnesium-SLR (optimized) (xylidyl blue method) kit purchased from Avecon Healthcare Pvt. Ltd.

With xylidyl blue, magnesium ions form a blue violet complex whose colour intensity is proportional to the magnesium concentration in the specimen and can be used for the photometric determination.

### ***Plasma chloride estimation***

Chloride was estimated in plasma samples by using Liquimax Chloride-SLR (MTC method) kit purchased from Avecon Healthcare Pvt. Ltd.

The chloride ions react with mercuric thiocyanate to release thiocyanate ions which in turn react with ferric thiocyanate. The absorbance of the red coloured complex at 505 nm is proportional to the chloride concentration.



## Environmental parameters

These parameters indicate that study was conducted in aHH area in India. Minimum and maximum ambient temperatures, dry bulb and wet bulb temperatures were recorded with respective thermometers for microenvironment of the experimental goats inside shed and in the vehicle at the time of sampling and recording physiological responses. The THI (Temperature Humidity index) was calculated from dry bulb and wet bulb temperatures using the equation  $THI = 0.72 \times (C_{db} + C_{wb}) + 40.6$  (McDowell et al. 1976). Daily relative humidity was calculated by the difference of dry bulb temperature and wet bulb temperature.

## Statistical analysis

Data analysis was carried out using SAS 9 software licenced to NDRI. Mean values at different sampling times were compared with respective basal mean values of each group using one-way analysis of variance with post-test as Dunnett's multiple comparison.

## Results and discussion

Meteriological data THI has been estimated 77.14 and 58.74 in the month of Sep - Oct and Dec - Jan respectively. This suggests HH and winter seasons in Karnal, India. Average values of plasma minerals Na, K,  $Cl^-$ , P and Mg (presented in Tables (1–5) and Figures (1–5)) obtained in all the three groups of goats at lfd and hfd, during winter and HH seasons and the analysis of variance in Table 6 for Na, K,  $Cl^-$ , P and Mg levels indicated that there was a significant difference ( $P < 0.05$ ) between seasons, between density and between groups.

**Table 1.** Average values of plasma sodium (mEq/L) 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
<b>Vitamin C (group I)</b>				
Before loading	147.17 ± 0.87 <sup>fw</sup>	147.00 ± 0.99 <sup>ew</sup>	146.17 ± 0.91 <sup>dw</sup>	149.67 ± 0.50 <sup>hx</sup>
After unloading	140.00 ± 1.01 <sup>ay</sup>	137.83 ± 0.62 <sup>aw</sup>	140.33 ± 0.97 <sup>ay</sup>	139.00 ± 0.58 <sup>bx</sup>
6 h Post	140.33 ± 0.36 <sup>aw</sup>	141.00 ± 0.56 <sup>bw</sup>	141.00 ± 0.34 <sup>aw</sup>	140.67 ± 0.52 <sup>cw</sup>
12 h Post	143.17 ± 0.81 <sup>bw</sup>	143.67 ± 0.87 <sup>cw</sup>	143.17 ± 0.81 <sup>cw</sup>	143.83 ± 0.55 <sup>ew</sup>
24 h Post	145.17 ± 0.99 <sup>dw</sup>	145.50 ± 0.89 <sup>dw</sup>	145.17 ± 0.99 <sup>dw</sup>	146.00 ± 0.89 <sup>fw</sup>
48 h Post	146.33 ± 0.62 <sup>ew</sup>	146.17 ± 0.77 <sup>dw</sup>	145.83 ± 0.73 <sup>dw</sup>	149.17 ± 0.77 <sup>hx</sup>
<b>Vitamin C and electrolyte mixture (Group II)</b>				
Before loading	146.00 ± 0.78 <sup>ex</sup>	148.50 ± 1.06 <sup>ez</sup>	145.00 ± 0.79 <sup>dw</sup>	147.00 ± 0.96 <sup>dy</sup>
After unloading	141.33 ± 0.55 <sup>aw</sup>	141.83 ± 0.73 <sup>bw</sup>	142.00 ± 0.42 <sup>bw</sup>	141.00 ± 0.74 <sup>cw</sup>
6 h Post	141.17 ± 0.94 <sup>aw</sup>	140.83 ± 0.68 <sup>bw</sup>	143.00 ± 0.58 <sup>cx</sup>	142.83 ± 0.58 <sup>dx</sup>
12 h Post	142.83 ± 0.63 <sup>bw</sup>	143.50 ± 0.70 <sup>cx</sup>	143.50 ± 0.79 <sup>cx</sup>	143.83 ± 0.62 <sup>ex</sup>
24 h Post	144.17 ± 0.71 <sup>cx</sup>	145.67 ± 0.86 <sup>dy</sup>	143.67 ± 0.27 <sup>cw</sup>	145.83 ± 0.65 <sup>fy</sup>
48 h Post	146.50 ± 0.31 <sup>ex</sup>	147.17 ± 0.86 <sup>ex</sup>	144.50 ± 0.61 <sup>dw</sup>	146.17 ± 1.13 <sup>fx</sup>
<b>Jaggery solution (group III)</b>				
Before loading	148.17 ± 0.45 <sup>fx</sup>	151.17 ± 0.89 <sup>fy</sup>	147.17 ± 0.87 <sup>ew</sup>	147.00 ± 0.99 <sup>gw</sup>
After unloading	140.67 ± 0.89 <sup>ax</sup>	138.00 ± 0.58 <sup>aw</sup>	140.00 ± 1.01 <sup>ax</sup>	137.83 ± 0.62 <sup>aw</sup>
6 h Post	141.33 ± 0.79 <sup>ax</sup>	141.33 ± 0.84 <sup>bx</sup>	140.33 ± 0.36 <sup>aw</sup>	141.00 ± 0.56 <sup>cx</sup>
12 h Post	143.33 ± 0.75 <sup>bw</sup>	146.67 ± 0.94 <sup>ex</sup>	143.17 ± 0.81 <sup>cw</sup>	143.67 ± 0.87 <sup>ew</sup>
24 h Post	145.17 ± 0.99 <sup>dw</sup>	149.17 ± 1.48 <sup>fx</sup>	145.17 ± 0.99 <sup>dw</sup>	145.50 ± 0.89 <sup>fw</sup>
48 h Post	146.33 ± 0.62 <sup>ew</sup>	150.17 ± 1.06 <sup>fx</sup>	146.33 ± 0.62 <sup>dw</sup>	146.17 ± 0.77 <sup>fw</sup>

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 plasma potassium (mEq/L) 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
<b>Vitamin C (group I)</b>				
Before loading	3.67 ± 0.11 <sup>dy</sup>	3.43 ± 0.10 <sup>dx</sup>	3.63 ± 0.13 <sup>dy</sup>	3.25 ± 0.12 <sup>dw</sup>
After unloading	2.55 ± 0.10 <sup>ax</sup>	2.27 ± 0.03 <sup>aw</sup>	2.94 ± 0.03 <sup>by</sup>	2.24 ± 0.03 <sup>aw</sup>
6 h Post	3.25 ± 0.13 <sup>cz</sup>	2.75 ± 0.19 <sup>bx</sup>	2.96 ± 0.09 <sup>by</sup>	2.31 ± 0.14 <sup>bw</sup>
12 h Post	3.57 ± 0.12 <sup>dz</sup>	3.37 ± 0.12 <sup>dy</sup>	3.08 ± 0.07 <sup>bx</sup>	2.86 ± 0.14 <sup>cw</sup>
24 h Post	3.58 ± 0.08 <sup>dz</sup>	3.44 ± 0.07 <sup>dy</sup>	3.29 ± 0.13 <sup>cx</sup>	3.10 ± 0.11 <sup>dw</sup>
48 h Post	3.62 ± 0.09 <sup>dy</sup>	3.42 ± 0.10 <sup>dx</sup>	3.53 ± 0.10 <sup>dy</sup>	3.22 ± 0.07 <sup>dw</sup>
<b>Vitamin C and electrolyte mixture (group II)</b>				
Before loading	3.40 ± 0.11 <sup>cx</sup>	3.47 ± 0.12 <sup>dx</sup>	3.06 ± 0.16 <sup>bw</sup>	3.59 ± 0.10 <sup>ey</sup>
After unloading	2.57 ± 0.11 <sup>ax</sup>	2.35 ± 0.13 <sup>aw</sup>	2.71 ± 0.09 <sup>ay</sup>	3.04 ± 0.11 <sup>dz</sup>
6 h Post	3.05 ± 0.15 <sup>bx</sup>	3.15 ± 0.13 <sup>cx</sup>	2.76 ± 0.12 <sup>aw</sup>	3.10 ± 0.14 <sup>dx</sup>
12 h Post	3.30 ± 0.11 <sup>cx</sup>	3.45 ± 0.12 <sup>dy</sup>	2.80 ± 0.10 <sup>aw</sup>	3.30 ± 0.12 <sup>dx</sup>
24 h Post	3.37 ± 0.12 <sup>cy</sup>	2.15 ± 0.40 <sup>aw</sup>	2.89 ± 0.13 <sup>bx</sup>	3.38 ± 0.03 <sup>dy</sup>
48 h Post	3.45 ± 0.10 <sup>cx</sup>	3.47 ± 0.07 <sup>dx</sup>	3.01 ± 0.09 <sup>bw</sup>	3.54 ± 0.14 <sup>ex</sup>
<b>Jaggery solution (group III)</b>				
Before loading	3.69 ± 0.08 <sup>dx</sup>	3.49 ± 0.09 <sup>dw</sup>	3.68 ± 0.10 <sup>dx</sup>	3.42 ± 0.12 <sup>dw</sup>
After unloading	2.58 ± 0.02 <sup>ax</sup>	2.29 ± 0.04 <sup>aw</sup>	2.56 ± 0.09 <sup>ax</sup>	2.26 ± 0.06 <sup>aw</sup>
6 h Post	3.28 ± 0.04 <sup>cx</sup>	3.14 ± 0.07 <sup>cx</sup>	3.28 ± 0.08 <sup>cx</sup>	3.31 ± 0.11 <sup>dx</sup>
12 h Post	3.56 ± 0.05 <sup>dx</sup>	3.35 ± 0.11 <sup>dw</sup>	3.56 ± 0.07 <sup>dx</sup>	3.38 ± 0.08 <sup>dw</sup>
24 h Post	3.54 ± 0.06 <sup>dx</sup>	3.41 ± 0.05 <sup>dw</sup>	3.59 ± 0.06 <sup>dx</sup>	3.45 ± 0.05 <sup>dw</sup>
48 h Post	3.60 ± 0.07 <sup>dx</sup>	3.40 ± 0.11 <sup>dw</sup>	3.61 ± 0.05 <sup>dx</sup>	3.46 ± 0.09 <sup>dw</sup>

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

**Table 3.** Average values of plasma phosphorus (mg/dL) 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
<b>Vitamin C (group I)</b>				
Before loading	7.77 ± 0.24 <sup>cx</sup>	8.65 ± 0.22 <sup>cy</sup>	6.11 ± 0.18 <sup>aw</sup>	6.00 ± 0.19 <sup>bw</sup>
After unloading	6.86 ± 0.37 <sup>cw</sup>	7.21 ± 0.34 <sup>aw</sup>	7.66 ± 0.09 <sup>bw</sup>	8.05 ± 0.14 <sup>fx</sup>
6 h Post	8.60 ± 0.00 <sup>dz</sup>	7.46 ± 0.18 <sup>ax</sup>	7.18 ± 0.17 <sup>bw</sup>	7.71 ± 0.05 <sup>ey</sup>
12 h Post	6.10 ± 0.00 <sup>bw</sup>	7.78 ± 0.04 <sup>az</sup>	6.98 ± 0.09 <sup>ax</sup>	7.28 ± 0.11 <sup>dy</sup>
24 h Post	6.03 ± 0.00 <sup>bw</sup>	8.08 ± 0.05 <sup>bz</sup>	6.75 ± 0.18 <sup>ax</sup>	7.00 ± 0.11 <sup>dy</sup>
48 h Post	7.84 ± 0.00 <sup>cx</sup>	8.54 ± 0.14 <sup>cy</sup>	6.62 ± 0.16 <sup>aw</sup>	6.46 ± 0.14 <sup>cw</sup>
<b>Vitamin C and electrolyte mixture (group II)</b>				
Before loading	6.65 ± 0.21 <sup>cy</sup>	7.32 ± 0.37 <sup>az</sup>	6.09 ± 0.46 <sup>ax</sup>	5.23 ± 0.52 <sup>aw</sup>
After unloading	5.42 ± 0.20 <sup>aw</sup>	7.18 ± 0.52 <sup>ax</sup>	6.95 ± 0.44 <sup>ax</sup>	6.45 ± 0.60 <sup>cx</sup>
6 h Post	5.72 ± 0.11 <sup>aw</sup>	7.22 ± 0.11 <sup>az</sup>	6.73 ± 0.14 <sup>ay</sup>	6.29 ± 0.03 <sup>cx</sup>
12 h Post	6.10 ± 0.03 <sup>bw</sup>	7.25 ± 0.15 <sup>ay</sup>	6.63 ± 0.15 <sup>ax</sup>	6.10 ± 0.01 <sup>bw</sup>
24 h Post	6.17 ± 0.02 <sup>bx</sup>	7.28 ± 0.11 <sup>az</sup>	6.53 ± 0.13 <sup>ay</sup>	5.79 ± 0.08 <sup>aw</sup>
48 h Post	6.34 ± 0.14 <sup>bx</sup>	7.29 ± 0.10 <sup>ay</sup>	6.40 ± 0.07 <sup>ax</sup>	5.44 ± 0.05 <sup>aw</sup>
<b>Jaggery solution (group III)</b>				
Before loading	6.91 ± 0.43 <sup>cw</sup>	7.06 ± 0.24 <sup>aw</sup>	6.99 ± 0.33 <sup>aw</sup>	7.56 ± 0.20 <sup>dw</sup>
After unloading	8.89 ± 0.21 <sup>ex</sup>	8.13 ± 0.30 <sup>bw</sup>	8.99 ± 0.11 <sup>cx</sup>	8.01 ± 0.15 <sup>fw</sup>
6 h Post	7.51 ± 0.23 <sup>cw</sup>	7.62 ± 0.19 <sup>aw</sup>	7.71 ± 0.13 <sup>bw</sup>	7.58 ± 0.14 <sup>ew</sup>
12 h Post	6.15 ± 0.18 <sup>bw</sup>	7.13 ± 0.20 <sup>ax</sup>	6.65 ± 0.14 <sup>aw</sup>	7.19 ± 0.16 <sup>dx</sup>
24 h Post	6.09 ± 0.12 <sup>bw</sup>	7.08 ± 0.21 <sup>ax</sup>	6.19 ± 0.10 <sup>aw</sup>	7.12 ± 0.17 <sup>dx</sup>
48 h Post	6.12 ± 0.11 <sup>bw</sup>	7.05 ± 0.18 <sup>ax</sup>	6.14 ± 0.15 <sup>aw</sup>	7.09 ± 0.15 <sup>dx</sup>

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

In both the seasons and in both the flocking density groups, higher pre-transportation values ( $P < 0.05$ ) of Na, K, Mg and Cl<sup>-</sup> were observed whereas  $P$  values post-transportation ( $P < 0.05$ ) were higher in HH season in all the treated groups and

**Table 4.** Average values of plasma magnesium (mg/dL) 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
<b>Vitamin C (group I)</b>				
Before loading	2.65 ± 0.04 <sup>bx</sup>	2.42 ± 0.05 <sup>cw</sup>	2.38 ± 0.03 <sup>bw</sup>	2.46 ± 0.03 <sup>cw</sup>
After unloading	2.54 ± 0.02 <sup>ax</sup>	2.22 ± 0.03 <sup>aw</sup>	2.27 ± 0.03 <sup>aw</sup>	2.25 ± 0.02 <sup>aw</sup>
6 h Post	2.83 ± 0.05 <sup>cx</sup>	2.83 ± 0.05 <sup>ex</sup>	2.29 ± 0.03 <sup>aw</sup>	2.27 ± 0.03 <sup>aw</sup>
12 h Post	2.93 ± 0.06 <sup>dx</sup>	2.93 ± 0.06 <sup>fx</sup>	2.30 ± 0.07 <sup>aw</sup>	2.33 ± 0.06 <sup>bw</sup>
24 h Post	2.96 ± 0.07 <sup>dx</sup>	2.96 ± 0.07 <sup>fx</sup>	2.33 ± 0.06 <sup>bw</sup>	2.37 ± 0.06 <sup>bw</sup>
48 h Post	2.98 ± 0.05 <sup>ey</sup>	2.98 ± 0.05 <sup>fy</sup>	2.34 ± 0.07 <sup>bw</sup>	2.43 ± 0.05 <sup>cx</sup>
<b>Vitamin C and electrolyte mixture (group II)</b>				
Before loading	2.62 ± 0.05 <sup>bx</sup>	2.36 ± 0.03 <sup>bw</sup>	2.58 ± 0.10 <sup>cx</sup>	2.90 ± 0.05 <sup>ey</sup>
After unloading	2.53 ± 0.04 <sup>ax</sup>	2.21 ± 0.02 <sup>aw</sup>	2.50 ± 0.08 <sup>cx</sup>	2.78 ± 0.07 <sup>ey</sup>
6 h Post	2.83 ± 0.05 <sup>cx</sup>	2.83 ± 0.05 <sup>ex</sup>	2.51 ± 0.05 <sup>cw</sup>	2.80 ± 0.02 <sup>ex</sup>
12 h Post	2.93 ± 0.06 <sup>dy</sup>	2.93 ± 0.06 <sup>fy</sup>	2.52 ± 0.06 <sup>cw</sup>	2.82 ± 0.02 <sup>ex</sup>
24 h Post	2.96 ± 0.07 <sup>dy</sup>	2.96 ± 0.07 <sup>fy</sup>	2.54 ± 0.03 <sup>cw</sup>	2.85 ± 0.02 <sup>ex</sup>
48 h Post	2.98 ± 0.05 <sup>ey</sup>	2.98 ± 0.05 <sup>fy</sup>	2.57 ± 0.03 <sup>cw</sup>	2.87 ± 0.04 <sup>cx</sup>
<b>Jaggery solution (group III)</b>				
Before loading	2.64 ± 0.05 <sup>bw</sup>	2.79 ± 0.13 <sup>ex</sup>	2.69 ± 0.03 <sup>dw</sup>	2.89 ± 0.10 <sup>ex</sup>
After unloading	2.58 ± 0.14 <sup>bw</sup>	2.44 ± 0.03 <sup>cw</sup>	2.59 ± 0.07 <sup>cw</sup>	2.64 ± 0.05 <sup>cw</sup>
6 h Post	2.61 ± 0.10 <sup>bw</sup>	2.52 ± 0.03 <sup>dw</sup>	2.60 ± 0.08 <sup>dw</sup>	2.62 ± 0.08 <sup>dw</sup>
12 h Post	2.63 ± 0.11 <sup>bw</sup>	2.67 ± 0.08 <sup>ew</sup>	2.62 ± 0.10 <sup>dw</sup>	2.68 ± 0.07 <sup>ew</sup>
24 h Post	2.64 ± 0.09 <sup>bw</sup>	2.73 ± 0.05 <sup>ew</sup>	2.63 ± 0.04 <sup>dw</sup>	2.75 ± 0.04 <sup>ew</sup>
48 h Post	2.67 ± 0.06 <sup>bw</sup>	2.76 ± 0.07 <sup>ex</sup>	2.65 ± 0.05 <sup>dw</sup>	2.79 ± 0.06 <sup>ex</sup>

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

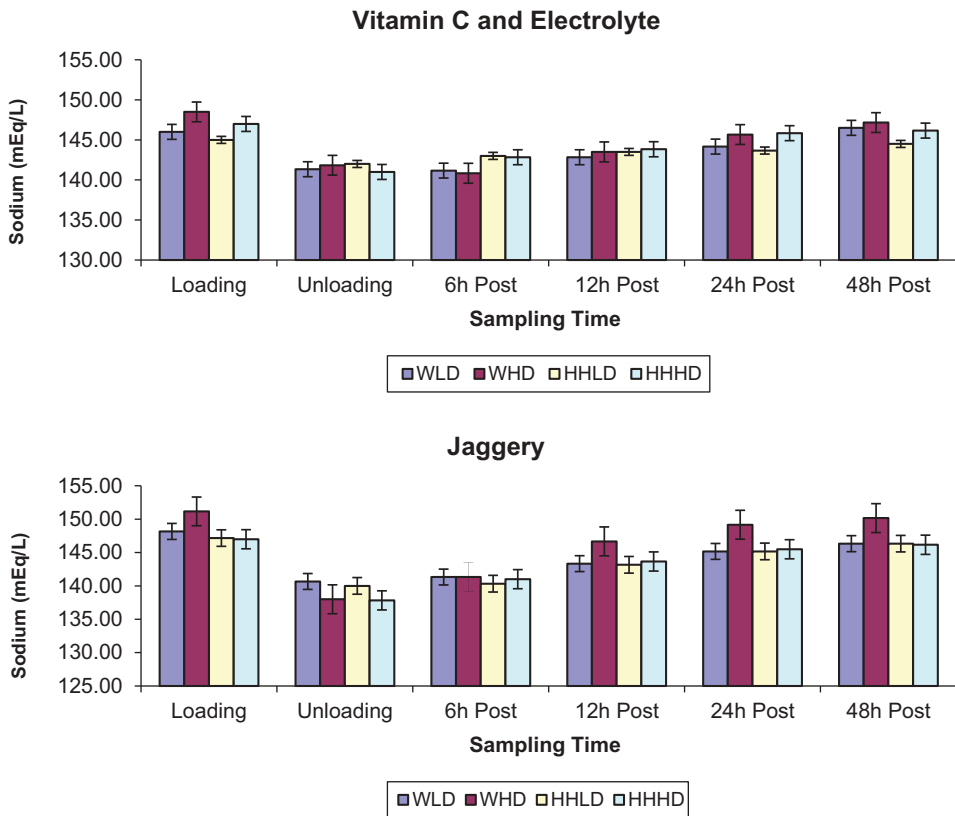
**Table 5.** Average values of plasma chloride (mMOI/L) in goats transported at different flocking densities during winter and hot humid seasons.

Transportation/groups	Winter		Hot humid	
	Low flocking density	High flocking density	Low flocking density	High flocking density
<b>Vitamin C (group I)</b>				
Before loading	127.05 ± 0.52 <sup>fw</sup>	133.55 ± 0.76 <sup>hx</sup>	133.29 ± 1.07 <sup>fx</sup>	134.13 ± 0.82 <sup>fx</sup>
After unloading	118.19 ± 0.36 <sup>cw</sup>	121.48 ± 1.84 <sup>dx</sup>	123.83 ± 0.54 <sup>ay</sup>	119.36 ± 1.47 <sup>aw</sup>
6 h Post	119.72 ± 1.72 <sup>dw</sup>	119.72 ± 1.72 <sup>cw</sup>	124.47 ± 0.42 <sup>bz</sup>	122.77 ± 0.28 <sup>by</sup>
12 h Post	120.35 ± 0.80 <sup>dw</sup>	120.35 ± 0.80 <sup>cw</sup>	127.79 ± 0.74 <sup>cy</sup>	126.55 ± 0.67 <sup>dx</sup>
24 h Post	124.17 ± 0.99 <sup>ew</sup>	124.17 ± 0.99 <sup>ew</sup>	129.65 ± 1.40 <sup>dx</sup>	128.39 ± 1.33 <sup>ex</sup>
48 h Post	124.25 ± 1.28 <sup>ew</sup>	124.25 ± 1.28 <sup>ew</sup>	131.15 ± 1.30 <sup>ex</sup>	130.70 ± 1.06 <sup>ex</sup>
<b>Vitamin C and electrolyte mixture (group II)</b>				
Before loading	128.17 ± 0.72 <sup>gw</sup>	129.46 ± 0.41 <sup>fx</sup>	133.47 ± 0.81 <sup>fy</sup>	133.24 ± 0.59 <sup>fy</sup>
After unloading	123.94 ± 1.98 <sup>ex</sup>	118.35 ± 1.58 <sup>cw</sup>	126.66 ± 1.76 <sup>cy</sup>	123.08 ± 0.87 <sup>bx</sup>
6 h Post	119.72 ± 1.72 <sup>dw</sup>	119.72 ± 1.72 <sup>cw</sup>	127.59 ± 0.32 <sup>cy</sup>	124.04 ± 0.73 <sup>cx</sup>
12 h Post	120.35 ± 0.80 <sup>dw</sup>	120.35 ± 0.80 <sup>cw</sup>	128.66 ± 0.15 <sup>dy</sup>	126.41 ± 0.81 <sup>dx</sup>
24 h Post	124.17 ± 0.99 <sup>ew</sup>	124.17 ± 0.99 <sup>ew</sup>	129.57 ± 1.15 <sup>dx</sup>	128.37 ± 0.57 <sup>ex</sup>
48 h Post	124.25 ± 1.28 <sup>ew</sup>	124.25 ± 1.28 <sup>ew</sup>	132.02 ± 0.68 <sup>ex</sup>	131.89 ± 0.57 <sup>ex</sup>
<b>Jaggery solution (group III)</b>				
Before loading	119.72 ± 1.72 <sup>dw</sup>	118.32 ± 1.24 <sup>cw</sup>	140.67 ± 1.02 <sup>gy</sup>	132.33 ± 0.58 <sup>ex</sup>
After unloading	110.38 ± 1.32 <sup>aw</sup>	110.33 ± 1.24 <sup>aw</sup>	128.82 ± 1.62 <sup>dx</sup>	127.27 ± 0.93 <sup>dx</sup>
6h Post	115.07 ± 2.09 <sup>bw</sup>	130.05 ± 1.40 <sup>gx</sup>	139.25 ± 1.37 <sup>gy</sup>	130.05 ± 1.40 <sup>ex</sup>
12 h Post	116.65 ± 1.28 <sup>bw</sup>	115.23 ± 2.06 <sup>cw</sup>	140.25 ± 1.37 <sup>gy</sup>	133.57 ± 1.20 <sup>fx</sup>
24 h Post	118.32 ± 1.24 <sup>cw</sup>	117.47 ± 1.05 <sup>cw</sup>	141.25 ± 1.37 <sup>gy</sup>	133.23 ± 1.57 <sup>fx</sup>
48 h Post	120.05 ± 1.65 <sup>dw</sup>	119.72 ± 1.72 <sup>cw</sup>	140.92 ± 1.36 <sup>gy</sup>	132.17 ± 1.56 <sup>ex</sup>

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

also in group III. Except for P, minimum values ( $P < 0.05$ ) of all other minerals were recorded just after unloading in both the density groups and in both the seasons, which then increased to basal values ( $P < 0.05$ ) after 12–24 h of post-transportation.

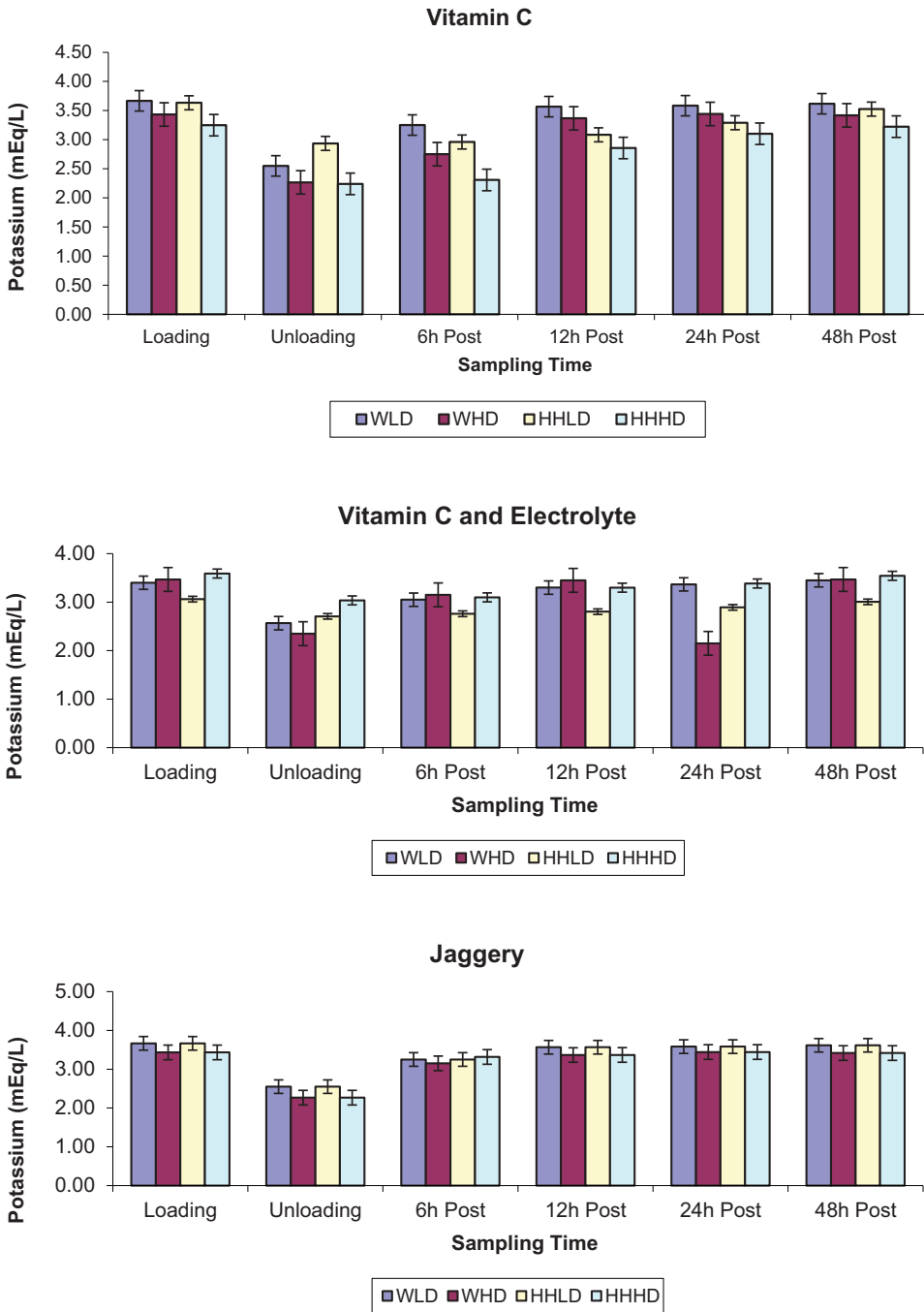




**Figure 1.** Average values of plasma sodium (mEq/L) in goats transported at different flocking densities during winter and hot humid seasons.

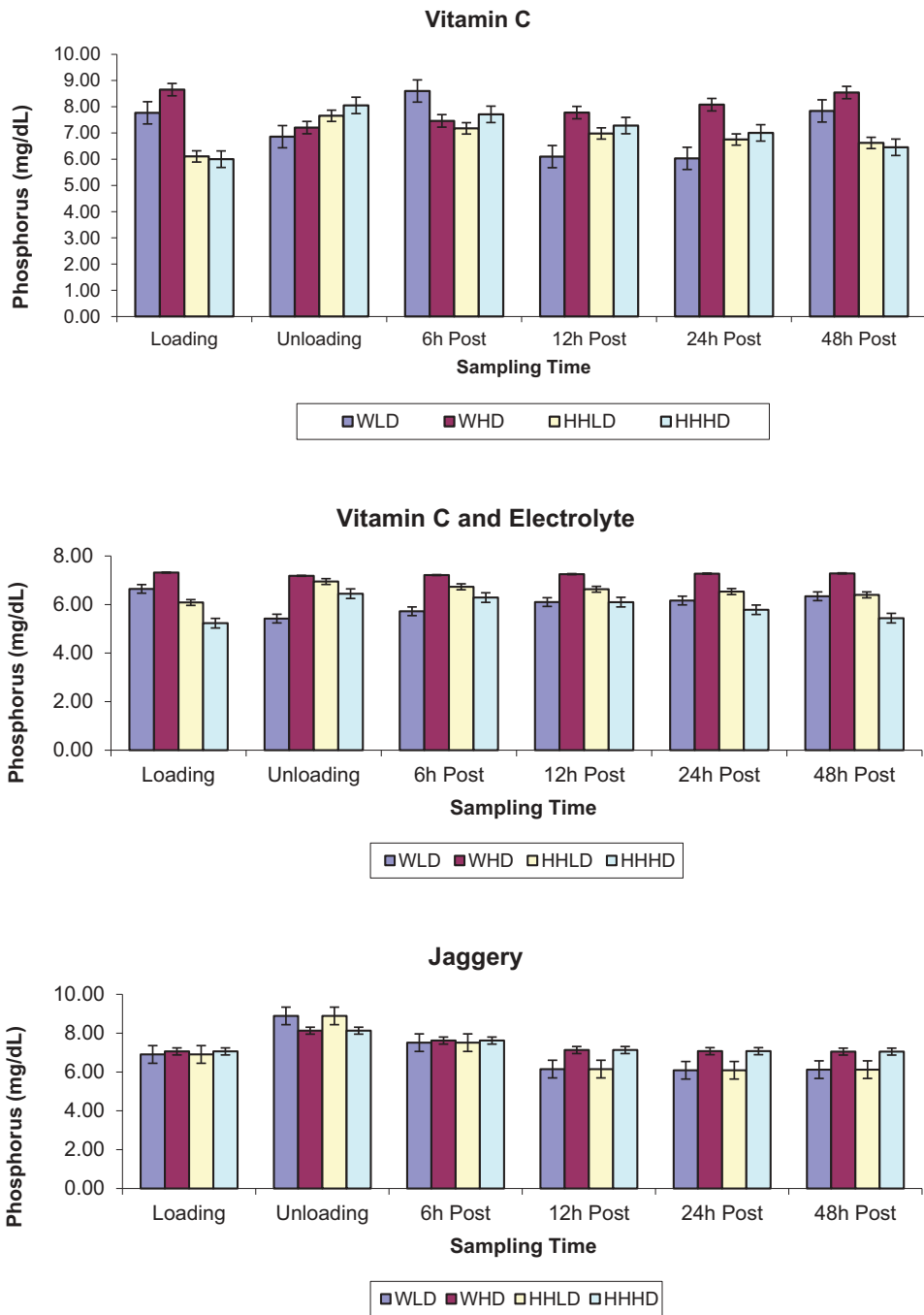
In winter season, in group I, minimum values of sodium were recorded just after unloading in the hfd group, which then increased to attain basal values ( $P < 0.05$ ) 24 h post-transportation. A similar increasing trend was observed for goats in HH season also. The values of sodium remained significantly ( $P < 0.05$ ) low even after 12 h post-transportation in all the groups in both seasons. In winter season, hfd goats of group II and group III showed higher values ( $P < 0.05$ ) of sodium as compared to lfd goats, which then increased to basal values ( $P < 0.05$ ) between 24 and 48 hours of post-transportation. The values of sodium in group III remained lower ( $P < 0.05$ ) even after 12 h post-transportation in both flocking densities during both the seasons. The group III goats showed higher variations in pre- and post-transportation sodium values in both flocking densities during both seasons as compared to the other two groups.

The decrease in blood sodium concentration can be (1) due to the inhibitory effect of cortisol on antidiuretic hormone, thus inhibiting its water retentive effect and hence diuresis and (2) because cortisol increases the secretion and efficiency of atrial natriuretic peptide, there is enhanced water excretion. However, some reports show that plasma concentrations of Na remain within the normal values in transported cattle, steer and calves (Atkinson 1992; Parker et al. 2003).



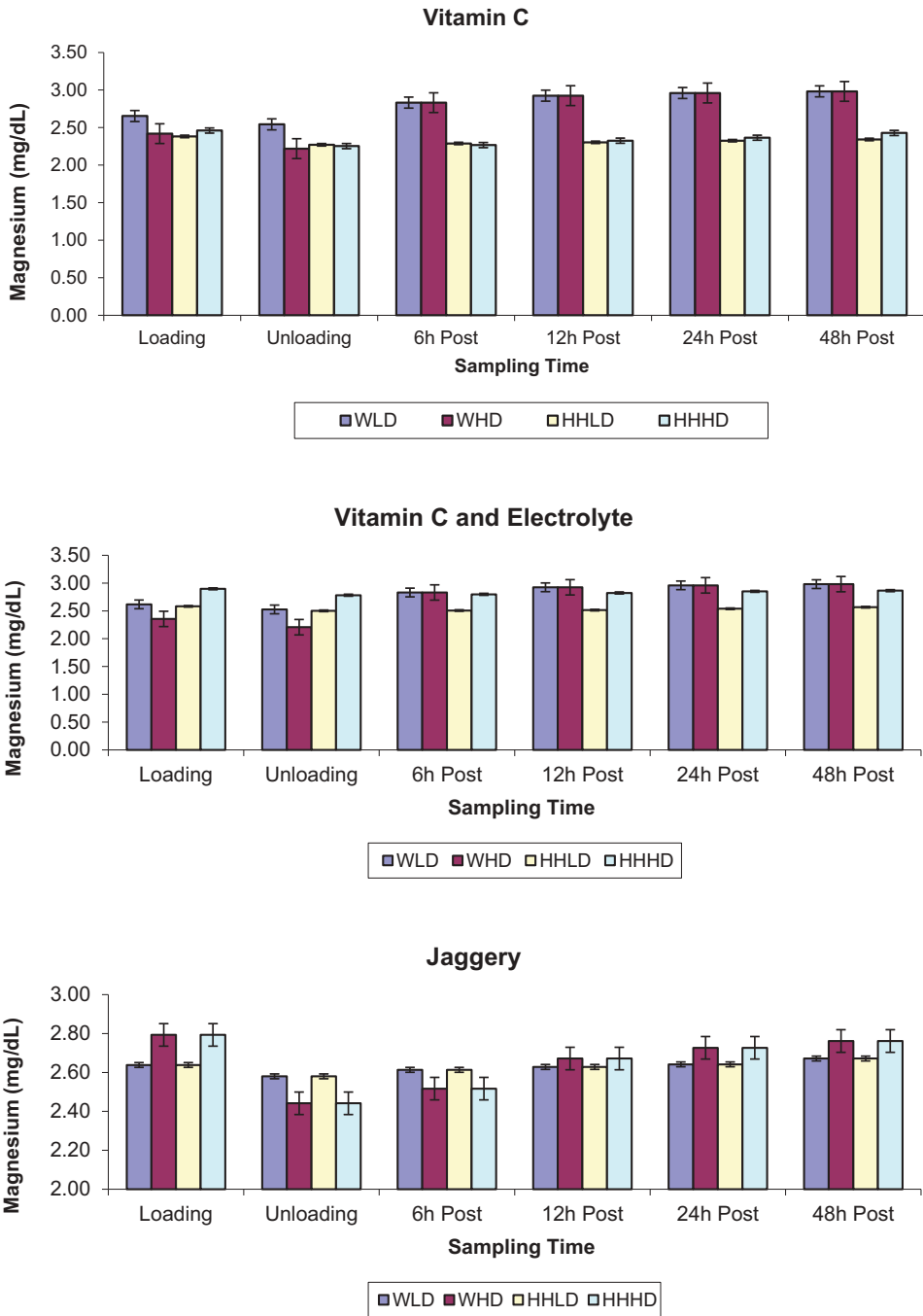
**Figure 2.** Average values of plasma potassium (mEq/L) in goats transported at different flocking densities during winter and hot humid seasons.

In all the three groups of goats and in both the seasons, higher pre-transportation values of potassium ( $P < 0.05$ ) were observed in all flocking density groups. In high-density group



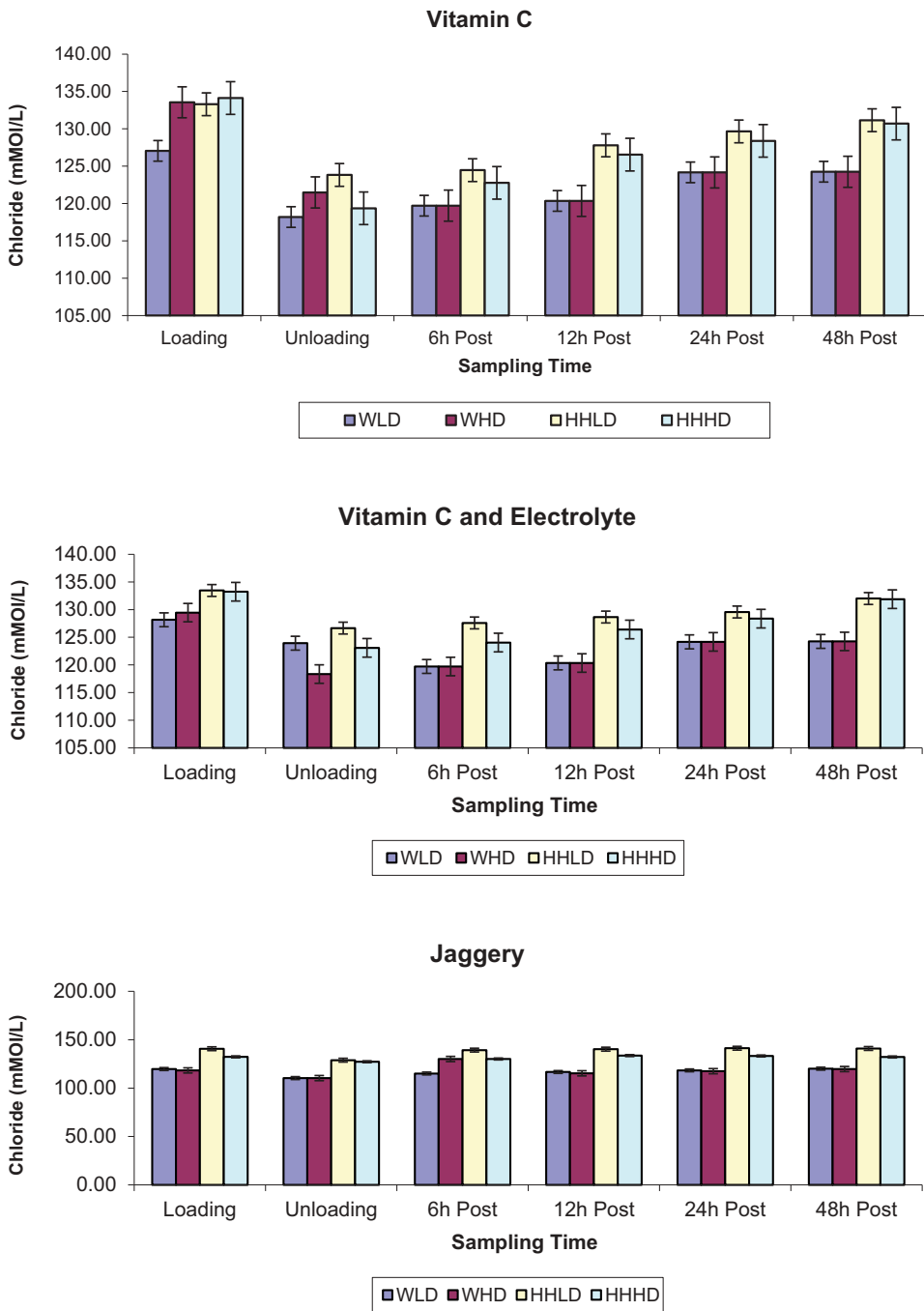
**Figure 3.** Average values of plasma phosphorus (mg/dL) in goats transported at different flocking densities during winter and hot humid seasons.

I goats, just after unloading, minimum values of potassium were recorded during winter ( $2.27 \pm 0.03$  mEq/L) and HH ( $2.24 \pm 0.03$  mEq/L) seasons, which then increased to normal



**Figure 4.** Average values of plasma magnesium (mg/dL) in goats transported at different flocking densities during winter and hot humid seasons.

basal values ( $P < 0.05$ ) 12 h post transportation. Similar increasing pattern in both the seasons was also observed for goats of groups II and III. The values of potassium remained significantly ( $P < 0.05$ ) low even after 12 h post transportation in all the groups in both seasons.



**Figure 5.** Average values of plasma chloride (mMOI/L) in goats transported at different flocking densities during winter and hot humid seasons.

Lower potassium concentration post transportation as indicated by our results is also in agreement with other authors. Parker et al. (2003) reported that transported *Bos indicus* steers animals had lower concentrations of plasma K compared with the control animals. It is

**Table 6.** Analysis of variance for mineral profile in transported goats.

Source of variation	df	Mean sum squares					
		Na	K	Cl <sup>-</sup>	P	Mg	F crit
Between seasons	1	51.36**	1.11**	28625.96**	15.48**	8.60**	3.85
Between densities	3	61.8**	2.62**	10235.20**	35.13**	3.61**	2.61
Between treatments	17	595.08**	9.69**	715.37**	25.30**	1.29**	1.63
Season × treatments	17	25.73*	1.03*	690.30*	6.45*	1.41*	1.63
Densities × treatments	51	24.92*	1.07*	309.34*	5.44*	0.60*	1.36
With in treatments	1206	10.88	0.24	26.88	0.97	0.08	
Total	1295	–	–	–	–	–	

\* $P < 0.05$ , \*\* $P < 0.01$ .

well recognized that stressor-induced activation of the hypothalamo-pituitary-adrenal axis stimulates the secretion of cortisol, resulting in the excretion of K (Parker et al. 2003). The hypokalemia associated with the transported group may also be the result of a lack of feed intake.

In group I, in winter season, minimum values of phosphorous ( $6.03 \pm 0.00$ ) were recorded at 24 h after unloading in the low-density group, that increased to basal values ( $P < 0.05$ ) after 24 h of post transportation. A similar decreasing trend was observed for goats in HH season also. In winter season, in group II, lfd goats showed lower values ( $P < 0.05$ ) of phosphorous as compared to hfd goats, whereas lower ( $P < 0.05$ ) values were recorded in hfd goats in HH season which then increased to basal values ( $P < 0.05$ ) after 24 h post transportation. Maximum values of phosphorous were recorded in each flocking density goats among all the groups just after unloading in H season, which then declined to lower values ( $P < 0.05$ ) after 24 h of post transportation. The group III goats exhibited highest phosphorous values as compared to groups I and II in both the flocking density goats during winter and HH seasons. The values of phosphorous in group III remained elevated ( $P < 0.05$ ) up to 6 h post-transportation in two flocking densities during both the seasons.

In the present study, the levels of phosphorus remained within the normal limits as reported by different workers. The results are in agreement with Galyean et al. (1981) who reported plasma phosphate ion concentration to be higher in fasted and transported animals for 32 h. However, some workers have reported no change in phosphate concentration in transported animals. Plasma concentrations of Ca, Na, Cl and P were observed to remain within the normal values in transported cattle, steer and calves (Atkinson 1992; Parker et al. 2003).

The values of magnesium remained significantly ( $P < 0.05$ ) low at unloading time and then gradually increased to  $2.98 \pm 0.05$  from  $2.42 \pm 0.05$  mg/dL between 6 and 48 hours in winter season in group I. In group II and III goats, no significant differences were observed between hfd goats as compared to lfd goats. In HH season, higher magnesium ( $P < 0.05$ ) values were recorded in hfd goats in group II. The values of magnesium in group III remained lower ( $P < 0.05$ ) till 6 h post-transportation in both flocking densities during both the seasons. Group III goats showed the lowest variations in pre- and post-transportation magnesium values in both flocking densities during both seasons as compared to the other two groups.

The decrease in magnesium concentration in goats due to transportation in our study is corroborated by other workers also. Transportation of desert goats during months

of March–April with an ambient temperature range of about 30–34°C for 2 h significantly reduced the plasma concentration of magnesium by about 21%, i.e. from an average of 0.82 to 0.65 m mol. (Ali et al. 2006). The concentration of magnesium in body tissue decreased during transportation, which led to a change in the activity of mitochondrial membrane of cells (Klaus-Dietrich 1985). Steinhardt and Hans-Herma (1998) also reported a significant lower concentration in magnesium of transported calves. Blood magnesium level of jaggery supplemented goats was not affected due to transportation in our study, which can be attributed to the fact that jaggery being a good source of minerals especially magnesium. As jaggery was fed for 5 days before transportation, goats may have built up higher blood magnesium concentration.

The values of chloride remained significantly ( $P < 0.05$ ) low at unloading and then gradually increased up to 48 h post-transportation but still remained significantly low when compared with pre-transportation values in group I. The post-transportation values in low and hfd goats could not attain pre-transportation (basal) values even after 48 h post-transportation. In HH season, higher chloride ( $P < 0.05$ ) values were recorded in lfd goats as compared to hfd goats in groups II and III and minimum post-transportation chloride values were reported in hfd goats. Group III goats exhibited significantly lower chloride values after 12 h post-transportation in low and hfd goats during winter seasons as compared to other groups.

Chloride is an essential electrolyte, trafficking in and out of cells through chloride channels and playing a key role in maintaining cell homeostasis and transmitting action potentials in neurons. Being the main extracellular anion, the concentration of this element creates an electrochemical gradient across cell membranes which is essential for nutrient transport, nerve conduction, muscle contraction and energy generation, indirectly aiding in maintaining pH balance. Plasma concentrations of Ca, Na,  $\text{Cl}^-$  and P were observed to remain within the normal values in transported cattle, steer and calves (Atkinson 1992; Parker et al. 2003).

## Conclusion

Transportation of animals causes physiological changes induced by handling, loading and actual transport. Supplementation of vitamin C, vitamin C + electrolyte and jaggery aided in reducing transportation stress individually but vitamin C + electrolyte combination proved more beneficial in alleviating transportation stress in goats.

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## Disclosure statement

No potential conflict of interest was reported by the authors.

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