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Haemato-biochemical response in grey donkeys working with pack loads in southern Agro-climatic zone of Tamil Nadu

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Abstract

Haemato-biochemical alterations were evaluated in adult Grey donkeys (n=30) working with pack loads equivalent to 50% of live body weight to estimate their draught power. Before and after work haematological (Hb, PCV, TEC, TLC and platelets) and biochemical parameters (BUN, creatinine, total protein, albumin, ALT, AST, Total bilirubin, calcium, phosphorus, glucose, cholesterol, triglycerides, sodium, potassium and chloride) were assessed. All haematological parameters were significantly increased (p<0.01) post work. Biochemical examination revealed a significantly (p<0.05) increased in serum BUN, creatinine, ALT, AST, ALP and triglycerides concentration; on the other hand, significant decrease was observed in blood total protein, albumin, glucose, cholesterol, sodium, potassium and chloride concentration. This study concluded the characterization of normal haemato-biochemical values in Grey donkeys in southern districts of Tamil Nadu along with the effect of pack load on haemato-biochemical indices.

Keywords: Haematological, biochemical indices, grey donkey, pack loads, Southern agro-climatic zone

Introduction

Donkeys (*Equus asinus*) were traditionally used as pack working equids for transport and riding in India. Their effectiveness as pack animals in areas where roads conditions are bad or non-existent, in hilly and mountainous terrain, in river banks and in muddy and marshy areas (Hassan and Ibitoye 1993; Pearson *et al.*, 1998) ^[4, 15]. In the Indian subcontinent donkeys are used for carrying bricks at brick kilns, sand from the river bank and transporting clothes by washer men as pack loads from ancient times. In recent times, the growing use of donkey milk especially in paediatric patients allergic to cow's milk proteins and cosmetic industry (Monti *et al.*, 2007; Cavallarin *et al.*, 2015) ^[10, 2] has seen an increasing number of donkey farms throughout Tamil Nadu.

Donkey owners use them for livelihood without minimum care and management for their health as they are often from poor and backward socio-economic status (Pal *et al.*, 2001)^[14]. Lack of awareness among donkey owners regarding the identification of stress, pain, and fatigue symptoms overloading at the brick kilns and sand on river bank often leads to the development of multiple health problems (Pritchard *et al.*, 2005)^[17].

To avoid cruelty and extracting optimum work, they require to be loaded optimally for an optimum duration along with local awareness and benefits of optimum workload have to be established in the donkey rearing management. Few studies (Pal *et al.*, 2001; Pal *et al.*, 2012; Olaifa *et al.*, 2012; Legha *et al.*, 2018) ^[14, 13, 12, 8] are available regarding to work under pack load. Further, normal reference haemato-biochemical values of Grey donkeys were not established in the Southern agro-climatic zone. Hence, the present study was designed to establish the haemato-biochemical reference values and effect of 50% pack load on the haemato-biochemical indices in the Grey Donkey in the Southern agro-climatic zone of the Indian Subcontinent.

Materials and Methods Location of study

The study was conducted at the villages of Tirunelveli (8.7139° N, 77.7567° E) and Thoothukudi (8.7642° N, 78.1348° E) districts and Veterinary College and Research Institute, Tirunelveli (8.7288° N, 77.7061° E) of Tamil Nadu, India during the period from January 2022 to August 2023.

Animal management and feeding module

Grey donkeys were reared in an extensive system of farming. They were fed with wheat bran, *ad-lib* roughage and grazed in the open terrains along with freely accessed potable drinking water except during the working period.

Experimental design

Thirty indigenous Grey adult Donkeys of 6-10 years of age weighing ~ 90 kg were used to carry pack load (sand from river bank) equivalent to 50% of their live body weight for a maximum of 6 hrs or until the animal exhibits signs of fatigue, whichever was earlier. The donkeys were walked for 10 km at @ 1.5 km/hr on Kachcha road in early morning or late evening hours with ambient temperature ranging from 26-36 °C. intermittent rest was provided at every 500 mts distance during which the unloading and loading were done. Fatigue was assessed on the basis of symptoms like incoordination of legs movement, forward movement, excessive salivation and frothing in mouth, increase respiration, excitement, tongue protrusion and unwillingness to continue the work after which they were given rest.

Blood Collection and Haemato-biochemical analysis

5ml of blood was collected aseptically from the jugular vein with minimum pain to the animal just before the start of work (before work) and after work in sterile EDTA tubes. About 2.0 ml of the whole blood was used for evaluating haematological parameters (haemoglobin g/dl, packed cell volume %, red blood cell $10^{6}/\mu$ l, white blood cell count $10^{3}/\mu$ l and platelet count x/µl) in an automated Haemoanalyser

(Mindray, BC-2800). Plasma was separated by centrifugation at 3000 rpm for 15 minutes and biochemical parameters were estimated by Bio Systems A15, Biochemistry Analyzer at Centralised Clinical Laboratory, Veterinary Clinical Complex, Veterinary College and Research Institute, Tirunelveli.

Statistical Analysis

Mean and SEM of before and after work parameters were compared by t-test in each treatment as per the methods detailed in Snedecor and Cochran (1967)^[18].

Result and Discussion

Haematological indices

The mean (\pm SE) haemoglobin (mg/dl) concentration before and after work is presented in Figure 1. The mean Hb concentration was significantly (p<0.01) increased after work in Grey donkeys at the Southern agro-climatic zone.

The mean (\pm SE) Packed Cell Volume (PCV) percentage before and after work is presented in Figure 1. The mean PCV value was significantly (p<0.01) increased after work in Grey donkeys at the Southern agro-climatic zone.

The mean (\pm SE) Total Erythrocytes Count (TEC) (x10⁶/µl) before and after work is presented in Figure 1. The mean TEC was significantly (*p*<0.01) increased after work in Grey donkeys at the Southern agro-climatic zone.

The mean (\pm SE) Total Leucocyte Count (TLC) (x10³/µl) before and after work is presented in Figure 1. The mean TLC was significantly (*p*<0.01) increased after work in Grey donkeys at Southern agro-climatic zone.

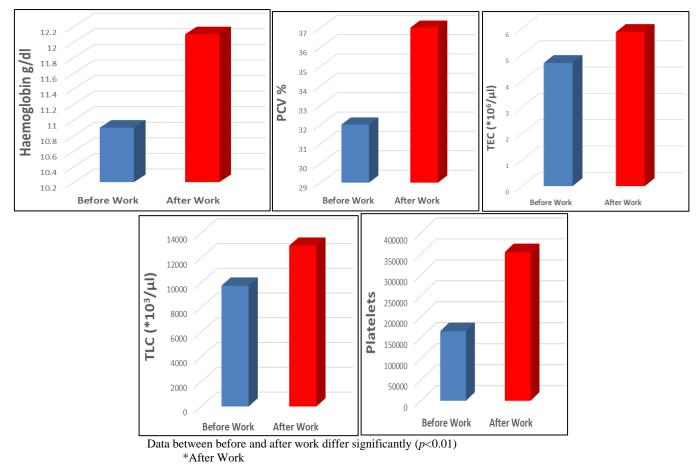


Fig 1: Mean Haematological response during working with pack load in Grey Donkeys in Southern Agro Climatic Zone.

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The mean (\pm SE) Platelets Count (x/µl) before and after work is presented in Figure 1. The mean platelet count was significantly (*p*<0.01) increased after work in Grey donkeys at the Southern agro-climatic zone.

Legha *et al.* (2018)^[8] reported a significant increase in WBC count, RBC count, HB and haematocrit values after work in pack loads with 40%, 50% and 60% of live body weight which agrees with present study. The increase in haematological indices might be due to the release of catecholamine from the adrenals which causes an increase in the fluxing of erythrocytes from the spleen into circulating blood during exercise in equines (Hodgson and Rose, 1994; Piccione *et al.*, 2008; Allaam *et al.*, 2014) ^[5, 16, 1].

Post-work PCV and haemoglobin increase progressively as haemoconcentration occurs due to dehydration and sweating during working and exercising in equines (Allaam *et al.*, 2014)^[1]. The cause of leucocytosis might be the influence of corticosteroids which are released from the adrenals in response to stress in working and exercising equines (Allaam *et al.*, 2014)^[1].

Serum Biochemical indices Protein Metabolites

The mean (\pm SE) BUN and Creatinine before and after work is presented in Figure 2. The mean serum BUN (mg/dl) and creatinine (mg/dl) was significantly (p<0.05) and non-significantly increased after work in Grey donkeys at the Southern agro-climatic zone.

The mean (\pm SE) Total Protein and Albumin before and after work is presented in Figure 2. The mean serum Total Protein (mg/dl) and albumin (mg/dl) was significantly (p<0.05) reduced after work in Grey donkeys at the Southern agroclimatic zone.

Gupta *et al.* (2016) ^[3] reported, urea, creatinine and uric acid contents in serum generally reflect the status of kidney functioning. Like other biochemical indices, mean blood urea content also ranged significantly from 25.25 ± 1.34 mg/dl to 39.96 ± 1.613 mg/dl in donkeys. The present study agrees with Moolchandani and Sareen (2015) ^[11], who observed a significant increase in BUN and Creatinine after work with the findings in horses following submaximal and maximal exercise.

Gupta *et al.* (2017) ^[19] reported a significant reduction in albumin concentration in donkeys to post-stress which agrees with the present study and might be due to albumin serving as an osmotic equilibrium regulator in body fluid, while total protein is an important for various regulatory mechanisms; their value differed significantly after work due to thermoregulatory and increased metabolism and also due to non-availability of nutritious grass covering Southern agroclimatic zone.

Liver Enzymes

The mean (\pm SE) ALT and AST before and after work is presented in Figure 3. The mean serum ALT (U/L) and AST (U/L) was significantly (p<0.05) increased after work in Grey donkeys at the Southern agro-climatic zone.

Ingvartsen (2006) ^[6] reported that AST and ALT are hepatocellular leakage enzyme used as a marker efficiently for hepatic lipidosis. AST and ALT concentration depends on the function of the liver, skeletal muscle and cardiac muscle of animals which might be the reason of an increase in AST and ALT concentration in response to the work in Grey donkeys.

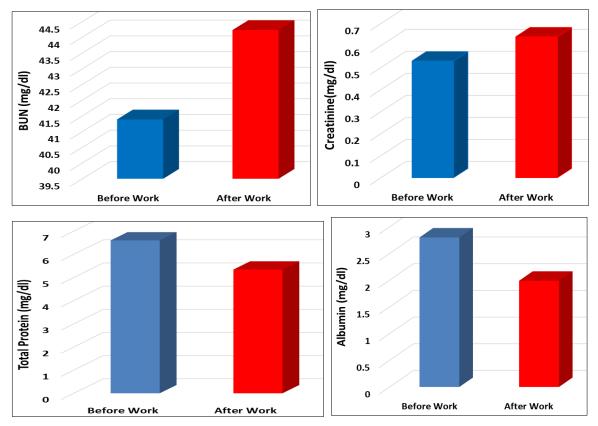


Fig 2: Mean Serum Protein metabolites during working with pack load in Grey Donkeys in Southern Agro Climatic Zone. Data between before and after work differ significantly (p<0.05) *After Work*Before Work



Fig 3: Mean Serum Liver Enzymes during working with pack load in Grey Donkeys in Southern Agro Climatic Zone. Data between before and after work differ significantly (p<0.05) *After Work *Before Work

Energy Metabolites

The mean (\pm SE) Glucose, Cholesterol and Triglycerides concentration before and after work is presented in Figure 3. The mean serum Glucose (mg/dl), Cholesterol (mg/dl) and Triglycerides (mg/dl) were significantly (p<0.05) increased after work in Grey donkeys at the Southern agro-climatic zone.

Moolchandani and Sareen (2015) ^[11] reported a significant decrease in serum glucose concentration which is in agreement with the present study this might by due to the increased demand for energy by muscles, during exercise the stress hormone cortisol increases and plays a vital role by enhancing glycolytic activities.

Further, Moolchandani and Sareen (2015)^[11] also observed a decrease in cholesterol and triglycerides serum concentration which is in agreement with our study, the decrease in

cholesterol and triglycerides might be due to increased adrenals and thyroid activities. Thyroid hormones are negatively correlated with cholesterol concentrations (Lehninger *et al.*, 2000)^[9]. Gupta *et al.* (2016)^[3] reported that cholesterol is an important metabolite required for the synthesis of steroids and its content was significantly low during the stress due to increased liver lipolysis under the influence of increased cortisol hormone.

Gupta *et al.* (2016) ^[3] reported a decrease in triglycerides concentration in blood during stress because Triglycerides are esters derived from glycerol and three fatty acids, that are used for energy and help in the homeostasis of blood glucose and adipose fat from the liver (Karami *et al.* 2014) ^[7]. So, negative energy balance during stressful conditions causes the mobilization of triglycerides to the liver which leads to a decrease in triglyceride concentration.

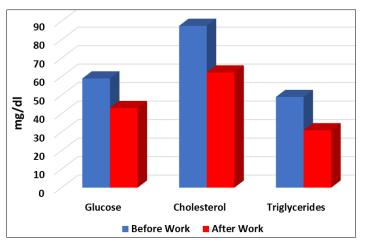


Fig 4: Mean Serum Energy Metabolites Enzymes during working with pack load in Grey Donkeys in Southern Agro Climatic Zone. Data between before and after work differ significantly (*p*<0.05) *After Work *Before Work

Mineral Profile

The mean (\pm SE) Calcium, Phosphorus, Sodium and Potassium concentration before and after work is presented in Figure 3. The mean serum Calcium (mg/dl), Phosphorus (mg/dl), Sodium (mmol/l) and Potassium (mmol/l) were significantly (p<0.05) increased after work in Grey donkeys at the Southern agro-climatic zone.

Gupta *et al.* (2016) ^[3] reported a decrease in calcium and phosphorus contents low calcium and phosphorus concentration in blood affects their body structure and feed availability. Further, increase in Na, K and Cl might be due to dehydration that occurred during work in Southern agroclimatic zone.

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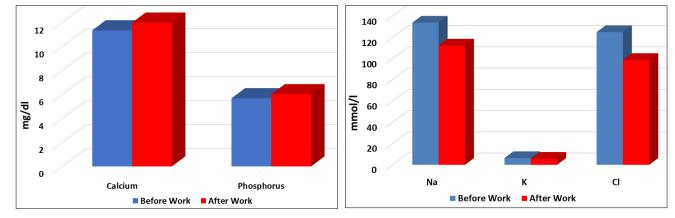


Fig 5: Mean Serum Mineral Profile during working with pack load in Grey Donkeys in Southern Agro Climatic Zone. Data between before and after work differ significantly (p<0.05) *After Work *Before Work

Conclusion

The present study established the reference values for haemato-biochemistry of Grey donkeys at the southern agroclimatic zone and effect of pack load on hematological and biochemical parameters has been recorded. This study reflects the adaptability of this breed in southern agro-climatic zone.

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