Evaluation of Physiological Parameters in Response to Endurance Exercise of Zanskar Ponies Adapted to High Altitude of Ladakh Region

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ABSTRACT

Zanskar pony, a native horse breed of Ladakh mainly used for transportation in Trans-Himalayan region of India, is well adapted to high altitude hypobaric hypoxia environment. Due to extreme conditions of the Ladakh region, better endurance of these ponies under hypoxic and extreme cold conditions is of utmost concern for their recruitment in Indian Army. In the present study, 12 young trained Zanskar ponies were evaluated during endurance exercise at an altitude of 3292 m above mean sea level. The animals were subjected to carriage transport with 65-70 kg load or riding on a track of 5-6 km. Physiological parameters viz., pulse rate (PR), heart rate (HR), rectal temperature (RT), respiratory rate (RR) and oxygen saturation (SaO₂) were recorded in Zanskar ponies during pre-exercise (T_0) , post- exercise (T_1) and post recovery $(T_2 2 h \text{ post resting})$ stages. Results showed marked increase in PR, HR, RR and RT post exercise time points. The mean values of PR increased from 49.83±4.62 to 73.67±21.54 per min, HR from 48±13.60 to 75±15.82 beats/min, RR from 37.83±9.70 to 57.67±13.48 per min and RT from 99.62±0.34 101.04±0.53 °F from pre stress to post endurance stress. The mean SaO, level reduced significantly (88.58±6.75 at T_0 versus 64.00±18.70 at T_1 and 54.42±14.79 at T_2) post exercise. This indicated limited availability of arterial oxygen for tissues which could be vital factor for adverse change in some of physio-biochemical parameters. Though the trend of physiological response was similar for all the 12 animals, still variation at individual animal level was observed during endurance stress. In future, some of these physiological parameters along with biochemical and molecular parameters could be evaluated as potential biomarkers in selecting ponies with superior endurance trait specifically under hypoxic conditions.

Keywords: Zanskar pony; Endurance; Physiological response; Hypoxia; High altitude adaptation; Ladakh

1. INTRODUCTION

Ladakh, one of the world's highest inhabited regions, is located at the northern most Trans-Himalayan part of India. It is a cold-arid desert at over 3000 meters mean see level (MSL), having very harsh climate characterised by extreme temperature (-40 °C in winter and 35 °C in summer); low relative humidity, precipitation (80-300 mm) and oxygen level (nearly 60-70 percent of the oxygen concentration at sea level); high UV radiations and wind erosion. To have survival and adaptability at cold as well as hypobaric hypoxic conditions, the human and other species inhabiting such harsh environment have developed certain unique physiological adaptation including comparatively higher aerobic performance, low hypoxic pulmonary vasoconstrictor response, better levels of oxygen saturation, high hemoglobin (Hb) level, increased flow of oxygen carrying blood and higher oxygen diffusion from blood to tissues/cells¹⁻³. Positive selection and selective sweeps at certain candidate genes related to hypoxic pathway, Hb phenotypes and other blood characteristics is the probable genetic basis of high altitude adaptation in human and other species⁴⁻⁹. Through constant evolutionary pressure and natural selection, human and several other species get well adapted to cold hypobaric hypoxic conditions¹⁰⁻¹².

In difficult terrain of Ladakh region (high altitude, cold and arid conditions), where land resources are meager, animal wealth plays an important role in the life of the local people. Common livestock genetic resources in Ladakh area include cattle, sheep, goat, horse, donkey, double hump camel and yak. These livestock species are well adapted to hypoxia, low temperature, high ultra violet exposure and survival during scarce nutrition in lean period. The native animals have developed certain physical changes like small stature, large hair fur, and thick skin so as to combat with tough environment. Most of these animals have unique circulatory adaptability with enlarged lungs, heart and larger concentration of red blood cells to accommodate the hypobaric-hypoxic environment. Amongst the livestock species, ponies along with mules hold special

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significance as these are the main pack animals for the local people and the troops in this region. Zanskar, a recognised pony breed of the India, is distributed mainly in Zanskar valley lying between 3500 m to 7000 m MSL altitudes in Ladakh region. These ponies are well adapted to hypobaric-hypoxia conditions at high altitude. Over the years these ponies are known for their sure footedness, ruggedness, ability to withstand extreme cold climates, work tirelessly, and carry loads on hilly tracks. These ponies are better suited than mules for snow-bound high-altitude areas and can carry 50-60 kg load. The Zanskar ponies are well-built, with medium height (120-140 cm), predominant eyes, heavy and long tail, and uniform gait. The body hairs are fine, long and glossy. These ponies are highly docile, resistant to diseases and can survive on local feed and fodder, in case of emergencies, can be deployed immediately without any acclimatisation¹³. Further, these ponies require least management practices and have less feed consumption.

Although Zanskar ponies are very important for the region, the population of the Zanskar ponies is showing decline in recent few years and is in the category of endangered breed. The current population of Zanskar ponies is only nine thousands in Ladakh (National livestock census, 2012). The Zanskar ponies seem to be evolved and adapted over the years under harsh conditions of Ladakh and are resource for unique genes or gene combinations. These ponies have developed functional and structural adaptations/ acclimatisation like development of muscle mass, a high proportion of muscle fibers, high densities of mitochondria, ability to increase oxygen-carrying capacity of blood, buffering and transport capacity for CO2, intramuscular storage of energy substrates (in particular glycogen), lactate depletion capacity of the liver and muscles, and efficient use of evaporation for thermoregulation^{14,15}. Loss of Zanskar ponies could result in loss of these gene(s) forever. The deployment of these ponies as pack animals for the army logistic support in Trans Himalayan region will help in revival of this breed in the region. It will also lead to significant saving to the state since movement of pack animals from plains is a costly affair and not advisable because they not only require special feed and management but need time to get acclimatised to harsh conditions of Ladakh.

Hence, selection of Zanskar ponies with efficient <u>Z</u> load carrying capacity and high endurance would be useful for meeting the demands of army and local people, conservation of important livestock resource with adaptation to cold hypoxic conditions, survival at minimum feed and fodder, and low management cost. Therefore, present study was undertaken to investigate the physiological responses to endurance stress and load carrying ability of Zanskar ponies so as to understand their performance traits at higher altitude.

2. MATERIAL AND METHODS

2.1 Location and Animals

Under the study, 12 Zanskar ponies maintained at Zanskar Ponies Breeding Unit of Siachen Vets, Remount Veterinary Services (RVS), Partapur (Leh) at an altitude of 3292 m above MSL were subjected to endurance exercise. Parental lines of

these ponies were taken over from DIHAR Zanskar research section, Leh for further breeding through continuous selection. Permission for conducting the experiment was obtained from RVC wing of Indian Military. A total of 12 ponies -5 males and 7 females based on their age, similar body size, physical fitness and temperament were selected for the experimentation (Table 1). All of the ponies, except twoYS-032 and ZP-065 were young, about 3 years of age. The selected animals were examined for their sound health, physical fitness and any injury. All of the ponies were moderately built and docile. However, male ponies were slightly larger in body size compared to females. The typical Zanskar pony animals are depicted in Fig. 1. These ponies were under the training with medium regimen for load carriage in high altitude driven hypoxic conditions for about 6 months. The experiment was conducted at noon in the month of August 2015, characterised by comfortable ambient temperature in day time of 30-32 °C and moderate air velocity. The ambient temperature and humidity were also recorded at an interval of 30 mins during endurance test using digital hygro-thermometer.

2.2 Exercise Regimen

Out of these 12 animals, 5 animals were given endurance exercise with 60 kg of loads, 6 animals were subjected for riding stress with a rider weighing about 70 kg. One pony-YS-

 Table 1.
 Details of Zanskar ponies included in the experiment at Siachen Vets of Remount Veterinary Services, Partapur

| Animal No. | Sex | Stage | Birth | Endurance Exercise |
|---------------|--------|------------------------|------------|-----------------------|
| YS-032 | Male | Geld (castrated adult) | 16.04.2010 | Without load |
| ZP-065 | Female | Mare (adult) | 25.05.2011 | Rider |
| ZP-078 | Male | Colt (Young) | 28.03.2012 | Rider |
| ZP-082 | Male | Colt (Young) | 19.04.2012 | Rider |
| ZP-083 | Female | Filly (Young) | 30.04.2012 | Load |
| ZP-086 | Female | Filly (Young) | 18.05.2012 | Rider |
| ZP-088 | Female | Filly (Young) | 28.06.2012 | Load |
| ZP- 089 | Female | Filly (Young) | 19.07.2012 | Load |
| ZP-090 | Male | Colt (Young) | 22.07.2012 | Load |
| ZP-091 | Female | Filly (Young) | 22.07.2012 | Load |
| ZP-092 | Female | Filly (Young) | 31.07.2012 | Rider |
| ZP-093 | Male | Colt (Young) | 25.08.2012 | Rider |



Figure 1. Depiction of facial features of typical Zanskar ponies.

032 was allowed to travel with no loading stress, as a control. Animals were allowed to travel in group of two at an interval of 10 min between the groups and were allowed to return back in same order and nearly at the same interval. None of the animal showed any discomfort during loading or riding and completed the trip without any interruption. All the animals were allowed to travel on a track of 5 km in hilly terrain with slightly uphill slope (track meant for endurance exercise), which took about 50 min to travel to and fro journey.

2.3 Physiological Parameters

Various physiological parameters pulse rate (PR), heart rate (HR), rectal temperature, (RT), respiratory rate (RR) and oxygen saturation (SaO₂) were recorded in Zanskar ponies just before initiating the exercise regimen (pre-endurance stress, T_o), immediately after completion of exercise (postendurance stress, T_1) and 2 h after exercise (post recovery period, T_{2}). Oxygen saturation was taken by holding the pulse oximeter (Masimo RAD-9 model, Massimo make) on the ear tip, whereas, pulse rate was recorded by palpating coccygeal artery at the base of the tail. Respiration rate was recorded by observing the movement of belly and thorax for one min. Heart rate was measured by placing a stethoscope against the chest of the horse under the elbow (olecranon) for a min and recording the value as beats/min. Rectal temperature was measured by inserting the tip of digital thermometer into the rectum until it gave the beep sound.

2.4 Data Analysis

Means and standard error were calculated for each of the parameter. Statistical difference for each parameter was determined by Analysis of variance 1 (ANOVA1) followed by Tukey's multiple comparison tests.

3. RESULTS AND DISCUSSION

The endurance exercise regimen carried out during the experimentation resulted in modulation of various physiological parameters in Zanskar ponies at different stages. Various physiological responses like pulse rate (PR), heart rate (HR), respiration rate (RR), rectal temperature (RT) and oxygen saturation (SaO₂) during pre and post stress of endurance test for individual animal were recorded. Except for SaO, there was marked increase in various physiological parameters from pre stress (T_0) to post stress (T_1) stages in all the 12 animals included in the study. As expected, changes in physiological parameters were least in control animal as it was allowed to travel without any load carriage. Interestingly, the physiological responses were not sex specific, as animals of both of the sexes showed similar trend and extreme values for the observed parameters specifically PR, HR and RR.

While accounting for all the animals together, there was significant (p<0.05) increase in mean values of various physiological parameters (PR, HR, RT and RR) except SaO₂ during post endurance exercise (Fig. 2). The mean value of PR during pre-stress state across 12 animals was 49.83 \pm 4.62 per min while

immediately post stress, the mean PR increased to 73.67±21.54, and returned to 52±6.34, close to the pre-stress range after 2 h of resting period (Fig. 2 (a)). The mean value of PR increased significantly (p<0.05) by ~1.5 times at T₁ as compared to T₀, indicating the effect of endurance stress in the animals.

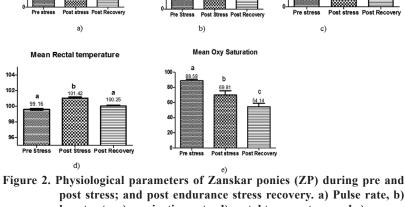
Similar trend was observed for HR values with significant (p < 0.05) increase in mean values from 48 ± 13.60 beats/ min in pre stress stage to 75±15.82 beats/min in post stress stage and returned to 49.67±5.51 beats/min in post recovery stage (Fig. 2 (b)). The mean values of HR also increased by ~ 1.5 times at T_1 as compared to T_0 . RR mean value increased from 37.83 \pm 9.70 per min at T₀ to 57.67 \pm 13.48 per min at T₁ and 49.33±5.21 per min at T₂ (Fig. 2c). The RT values also increased significantly (p<0.05) during post endurance stress with values of 99.62 \pm 0.34 at T₀, 101.04 \pm 0.53 at T₁ and 100.03 \pm 0.43 at T₂ time points (Fig. 2d). On the other hand, the mean SaO₂ level reduced significantly (p<0.05) after post endurance stress. Its value decreased from 88.58 ± 6.75 at T_o, to 69.81 ± 18.70 at T₁ and further to 54.14 ± 14.79 at T₂ time points (Fig. 2(e)). The reason for increase in physiological parameters post stress could be explained by the fact that during exercise there is increase in body core temperature resulting in enhanced blood flow towards periphery including skeletal muscle, so as to dissipate heat through skin to meet the enhanced energy requirements. Hence, HR, RR and other physiological parameters get enhanced during post stress. In Zanskar ponies, the increase in HR, PR and RR post exercise may indicate the increased requirement of blood oxygen to the tissues under hypoxic conditions.

The resting values of RT and HR in Zanskar ponies were within the normal range of 99.5-101.5°F and 26-42 beats/min for equines as suggested by Evans and Rose¹⁶. However, the resting stage mean value of Zanskar ponies showed tangible differences with exotic horses reported in one of the previous study¹⁷. In Zanskar ponies, the mean value of HR rate at

Mean Respiration rate

Mean Heart rate

Mean Pulse Rate



post stress; and post endurance stress recovery. a) Pulse rate, b) heart rate, c) respiration rate, d) rectal temperature and e) oxygen saturation. Values are expressed as means± SE. Different letters indicate significant differences (P<0.05).

resting stage was 48 ± 13.60 beats/min whereas in exotic horses, the mean value was 37.3 ± 6.1 beats/min. Similarly, RR mean values in Zanskar ponies was 37.83 ± 9.70 per min, while in exotic thoroughbred horse the mean RR value was 42.9 ± 16.7 per min. Further, mean RT values of Zanskar ponies (99.62±0.34) at resting stage were also different from that of exotic high thoroughbred horse (101.0±1.3). This difference in basic physiological parameters between the two horse types could be due to their genetic differences or due to the distinct environments to which they are adapted.

The oxygen saturation level measured across T_0 , T_1 and T_2 stages is another important physiological parameter especially at higher altitude where atmospheric pressure remains low. The mean SaO2 level showed significant reduction post exercise (88.58 at \tilde{T}_0 versus 64.00 at T_1 and 54.42 at T_2), indicating limited availability of arterial oxygen. It is interesting to note that PR, HR and RT at post recovery stage were not significantly higher than pre-stress condition. However, RR was significantly higher than pre-stress condition. This phenomenon is coupled with significantly lowest SaO₂ level indicates that two hours period for recovery from stress induced condition is not sufficient. Since, oxygen saturation is one of the critical parameters indicating various changes in physiobiochemical markers in animals experiencing endurance stress, under hypoxic conditions. Therefore, it would be interesting to conduct further research by including more number of animals to evaluate the association of HR, RR, PR, RT and SaO2 with endurance fitness in ponies working at high altitude hypoxic conditions. Further, efforts are underway to generate biochemical, hematological and molecular data (identify candidate genes and associated pathways) in order to evaluate various parameters and understand the overall mechanism of endurance stress in Zanskar horses.

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