

# INVESTIGATION ON PERFORMANCE OF DIFFERENT AGE AND GENDER USING BIOCHEMICAL PROFILE OF HORSES DURING 120 KM ENDURANCE RACE

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**ABSTRACT.** Exercise-induced changes of pro-oxidant generating substances and muscle fibre damaging enzymes are the possible causes of reduced performance in endurance horses. Therefore, this study aims to investigate on the performance of different age and gender using biochemical profile of endurance horses during 120 km endurance race. One hundred and eighty four Arabian endurance horses were physically examined and blood samples were collected post-race. After physical examination, the MD (F, n= 44; G, n = 86) and SC (F, n = 20; G, n = 34) were identified. T-test and pairwise correlation were used for the analysis. There were significant differences in CK, uric acid, lactate and age ( $P<0.0001$ ) respectively between the gender in the MD and SC. There were significant differences in AST ( $P<0.0029$ ) and GGT ( $P<0.0039$ ) respectively between the gender in the MD and SC. In conclusion age and gender significantly affect performance of endurance horses in relation to biochemical

profile during 120 km endurance race. Therefore, further studies are required to determine if age, gender and biochemical profile could be used to assess performance in endurance horses.

*Keywords:* age, gender, biochemical, endurance horses, performance

## INTRODUCTION

Assessment of biochemical, hematological and physical parameters of horses regarding age, gender and breeds (Lacerda *et al.*, 2006; Gurgoze and Icen, 2010; Fazio *et al.*, 2011) type of sporting events (Vincze *et al.*, 2010) and the myriad environmental factors (Serrano *et al.*, 2001) are crucial for the evaluation of health and fitness levels of horses (Gurgoze and Icen, 2010 and Fazio *et al.*, 2011).

According to Conley *et al.* (2000), 10 to 16 years old horses were still in their climax of performance. In Malaysia, up to 40% of the horse populations are between 11 to 15 years old and many of these horses

perform well in athletic competitions. In contrast, horses that were in their late teens were at the climax of their performance, competing in endurance rides, three-day eventing, jumping, dressage and other athletic competitions (Robert *et al.*, 2004).

Gamma glutamyl transferase (GGT) and uric acid were incriminated in the generation of free radicals and oxidative stress (Lee *et al.*, 2005; Yang *et al.*, 2007 and Sautin *et al.*, 2008). Oxidative stress was accountable for many metabolic disorders (Lee *et al.*, 2005 and Piccione *et al.*, 2007) causing cell damage or death (Piccione *et al.*, 2007 and Gondim *et al.*, 2009). GGT was responsible for exercise intolerance and poor performance (Whitfield, 2001; Lee *et al.*, 2005, and Yang *et al.*, 2007).

As a result of the low reactivity of uricase, uric acid increases exponentially when the metabolic pathway is blocked during exercises (Tullson *et al.*, 1995; Essén-Gustavsson *et al.*, 1999; Moriwaki *et al.*, 1999 and Castejon *et al.*, 2006). After strong muscular contractions hypoxanthine increases and uric acid concentration gets elevated in both, the contracting muscle and in the plasma (Norman *et al.*, 1987; Hellsten-Westing *et al.*, 1993 and Ji, 1999).

Horses with rhabdomyolysis reveal increased muscle enzymes, Creatine Kinase (CK), aspartate aminotransferase (AST) and Lactate (Hodgson *et al.*, 1994) leading to muscle damage (Piccione *et al.*, 2008). Formation of lactic acid can not be alienated from metabolic acidosis which could reveal pro-oxidant proclivity and lactic acidosis was connected with the generation of free

radical and lipid peroxidation (Bralet *et al.*, 1991 and Groussard *et al.*, 2000). In a study conducted by Anna *et al.* (2011) indicated an increased level of lactate in both stallions and mares after training.

In conclusion age and gender significantly affect performance of endurance horses in relation to biochemical profile during 120 km endurance race. Therefore, this study aims to investigate on the performance of different age and gender using biochemical profile of endurance horses during 120 km endurance race.

## MATERIALS AND METHODS

One hundred and eighty-four Arabian horses comprising of geldings and mares participated in an endurance competition of 120 km. Of this number, 86 geldings and 44 mares had metabolic syndrome and were eliminated from the race and 34 geldings and 20 mares completed the race successfully. The age and body weight of the horses ranged between  $12 \pm 0.29$  to  $9 \pm 0.31$  years and 350 to 450 kg respectively. Veterinary inspection was conducted after each loop of the races on all competing horses and physical parameters were recorded.

At the end of the endurance race the horses were classified as successfully completed (SC) the race or metabolic disordered (MD). Thus, the criteria for assessing a horse as (SC) depends on the horse's capability to uphold normal gastrointestinal, respiratory, cardiac, or musculoskeletal status and with a heart

rate of equal to or below 64 beats  $\text{min}^{-1}$  and with an exceptional hydration status after a maximum of 20 min of recovery period. MD endurance horses are those that could not attend to the above mentioned norms and were afterward eliminated from the endurance race (Khaled and Ahmad, 2008).

The ambient temperature and relative humidity were recorded at an interval of 30 minutes from the beginning of the race to the end. The mean temperature ( $^{\circ}\text{C}$ ) and humidity (%) were  $29.96 \pm 1.1^{\circ}\text{C}$  and  $71.73 \pm 4.05\%$  respectively during the period of the endurance race. The geographical scenery was good and favorable, water points were also provided at designated places along the track. The ambient temperature and humidity were measured using Casella<sup>®</sup> thermo-hygrometer.

Blood samples were obtained from all the horses via the jugular venipuncture into heparinized vacutainer tubes for biochemical analysis. The blood sample collection was performed immediately after 20 min of the recovery period and analyzed immediately in the laboratory, which is located within the premises of

the event. Uric acid, lactate, AST, CK and GGT were determined with chemistry analyzer (Hitachi 920<sup>®</sup>) using standard diagnostic kits (Roche<sup>®</sup>). The data were analyzed using pairwise correlation and T-test. The statistical software package JMP<sup>®</sup> 9. NC: SAS Institute Inc. was used for the analysis. Analyses were considered as significant at  $P < 0.05$ .

## RESULTS AND DISCUSSION

The post MD and SC blood lactate, uric acid, GGT, CK, AST, age and gender of endurance horses after covering a distance of 120 km were presented in Table 1 and 2 respectively.

Table 1 showed the age, gender, uric acid and lactate concentrations of MD and SC endurance horses. There were significant differences between gender in terms of age, uric acid and lactate concentration ( $P < 0.0001$ ) respectively in the performance groups (MD and SC).

Table 2 showed the age, gender, CK, AST and GGT concentrations of MD and SC endurance horses. There were significant differences between gender

**Table 1.** Performance of different age and gender using biochemical profile in endurance horses during 120 km race

Gender	MD			SC		
	Age (yrs)	Lactate ( $\text{mmol L}^{-1}$ )	Uric Acid ( $\mu\text{mol L}^{-1}$ )	Age (yrs)	Lactate ( $\text{mmol L}^{-1}$ )	Uric Acid ( $\mu\text{mol L}^{-1}$ )
G	$12^a \pm 0.29$	$4.36^a \pm 0.25$	$56.26^a \pm 7.38$	$10^b \pm 0.34$	$2.04^b \pm 0.25$	$19.86^b \pm 6.32$
F	$12^a \pm 0.29$	$4.19^a \pm 0.17$	$50.79^a \pm 5.09$	$9^b \pm 0.31$	$1.88^b \pm 0.18$	$17.80^b \pm 4.90$

Values are expressed as mean  $\pm$  S.E <sup>a, b</sup>, within each row; means with different superscripts are significantly different at  $p < 0.05$ .

MD = Metabolic disordered endurance horses; SC = Endurance horses that have successfully completed the race. G = Gelding; F = Mare.

**Table 2.** Performance of different age and gender using biochemical profile in endurance horses during 120 km race

Gender	MD				SC			
	Age (yrs)	GGT (U/L)	AST (U L <sup>-1</sup> )	CK (U L <sup>-1</sup> )	Age (yrs)	GGT (U/L)	AST (U L <sup>-1</sup> )	CK (U L <sup>-1</sup> )
G	12 <sup>a</sup> ± 0.29	20 <sup>a</sup> ± 0.9	591.9 <sup>a</sup> ± 53.5	1903 <sup>a</sup> ± 208	10 <sup>b</sup> ± 0.34	18 <sup>b</sup> ± 1.0	461.9 <sup>b</sup> ± 36.9	461 <sup>b</sup> ± 83
F	12 <sup>a</sup> ± 0.29	20 <sup>a</sup> ± 0.7	808.9 <sup>a</sup> ± 72.8	2108 <sup>a</sup> ± 153	9 <sup>b</sup> ± 0.31	17 <sup>b</sup> ± 1.0	502.9 <sup>b</sup> ± 33.9	575 <sup>b</sup> ± 72

Values are expressed as mean ± S.E.<sup>a, b</sup>, within each row; means with different superscripts are significantly different at  $p < 0.05$ . MD = Metabolic disordered endurance horses; SC = Endurance horses that have successfully completed the race. GGT = Glutamytransferase; AST = Aspartate aminotransferase; CK = Creatine kinase; G = Gelding; F = Mare.

in terms of age, CK concentrations ( $P < 0.0001$ ), AST ( $P < 0.0029$ ) and GGT ( $P < 0.0039$ ) respectively in the performance groups (MD and SC).

There were significant positive correlation among the geldings in respect of age and lactate ( $r = 0.3162$ ;  $P < 0.0004$ ), age and uric acid ( $r = 0.2936$ ;  $P < 0.0011$ ), age and CK ( $r = 0.2618$ ;  $P < 0.0037$ ) and there was a significant positive correlation among the mares in respect of age and CK ( $r = 0.2923$ ;  $P < 0.0238$ ).

In the present study all endurance horses within the age range of 12 years were eliminated (MD) from the race while, those within the age limit of 9 to 10 years (SC) performed spectacularly well in the endurance race of 120 km distance. This difference in performance could be due to age difference; and is different from the findings of Conley *et al.*, (2000) and Robert *et al.*, (2004) where they indicated that horses from the age of 10 and above were in their climax of performance.

There was a significant difference between the MD and SC performance

endurance horses within the gender and age categories, the mares had much lower concentrations of lactate and uric acid in both groups than the geldings. The age range of 12 years in the MD performance category had much higher concentrations of lactate and uric acid than those within the age limit of 9 and 10 years in the SC performance category. This could be due to the merged oxidative stress effect of lactate and uric acid (Lee *et al.*, 2005; Fazio *et al.*, 2007; Piccione *et al.*, 2007; Richard *et al.*, 2009 and Gondim *et al.*, 2009), and perhaps due to the devastating metabolic acidosis damaging effect of lactate on the muscle tissues leading to poor performance (Bralet *et al.*, 1991; Groussard *et al.*, 2000 and Hudson *et al.*, 2008). Lactate concentrations of  $4.19 \pm 0.17$  mmol/L to  $4.36 \pm 0.25$  mmol/L could perhaps indicate unfit threshold level in horses during equine endurance events and training protocols within the age range of 12 years in the MD performance category (Fielding *et al.*, 2009 and Lindner *et al.*, 2009) while a concentration of  $2.04 \pm 0.25$

to  $1.88 \pm 0.18$  could probably indicate the required aerobic performance level for fit horses within the age range of 9 to 10 years in the SC performance category.

In the present study there were significant differences between the MD and SC performance endurance horses within the gender and age categories, the geldings and mares had much lower concentrations of AST and CK within the age of 9 to 10 years in the SC performance category than those in the age group of 12 years in the MD performance category. This could perhaps indicate the occurrence of rhabdomyolysis which subsequently increases the muscle enzymes with resultant muscle damaging effect and reduced performance of the MD performance endurance horses (Hodgson *et al.*, 1994).

Furthermore, in the present study there was a significant difference between the MD and SC performance endurance horses within the gender and age categories, the geldings and mares had much lower concentrations of GGT within the age of 9 to 10 years in the SC performance category than those in the age group of 12 years in the MD performance category. This difference could probably emanate due to oxidative stress and exercise intolerance related with GGT during arduous endurance races (Whitfield, 2001; Lee *et al.*, 2004; Lee *et al.*, 2005, and Yang *et al.*, 2007). As an indicator of oxidative stress, GGT within its normal range has a dose response relationship with exercise intolerance and poor performance (Lee *et al.*, 2005, and Yang *et al.*, 2007).

## CONCLUSION

In conclusion age and gender significantly affect performance of endurance horses in relation to biochemical profile during 120 km endurance race. Further studies are desirable to determine if lactate, uric acid AST, CK and GGT could be used to assess performance during training and endurance events in relation to age and gender.

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