# Assessment of some liver enzyme activities in healthy Iraqi racing Horses .

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## Summary

The current study was conducted for the measurement of normal range and mean value of serum alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) in (125) clinically normal Iraqi racing horses (males and females, Arabian and Crossbred), aged 2 - 8 years; < 4, 4 and > 4 age groups.

The results revealed that the range and the mean value  $\pm$  SEM of these enzymes were as follows ; serum ALT 2 – 28 U/L and 11.07  $\pm$  0.62 U/L, serum AST 30 – 210 U/L and 78.00  $\pm$  3.62 U/L and serum ALP 63 – 204 IU/L and 96.84  $\pm$  2.52 IU/L respectively .However, significant difference (P < 0.05) in serum ALT was recorded between Arabian and Crossbred , while serum AST values showed a significant differences (P < 0.05) between males and females , as well as between less and more than four years age groups .On the other hand , there was no significant difference in serum ALP values between the studied groups .

In conclusion , this study recorded and established the normal range reference values and mean  $\pm$  SEM of serum liver enzymes ALT , AST and ALP in clinically healthy Iraqi racing horses .

Key words : Liver , enzymes , activities , racing horses .

#### الخلاصة

اجريت هذه الدراسة على (125) جواد سباق سوية سريرياً (ذكور واناث ، عربية ومضربة) تراوحت اعمارها بين 2 - 8 سنوات في مجاميع عمرية > 4 ، 4 و 4 < اشتملت الفحوصات على قياس ALP, AST, ALT في المصل لتحديد المديات الطبيعية ومتوسط القيم في خيول السباق السوية . اظهرت الدراسة الحالية ان المديات والمعدلات مع اللخطأ القياسي لهذه الانزيمات كما يلي U/L 28 - 2 ALT و U/L 3.62 ± 78.00 و U/L 210 - 30 AST ، U/L 0.62 ± 78.00 و U/L 210 - 30 ALP و 10/L 204 - 63 ALP على التوالي .

تظهر الدراسة وجود فرق معنوي عند مستوى (P < 0.05) في قيم ALT بين الخيول العربية والمضربة ، بينما تشير النتائج ان قيم AST لها فرق معنوي عند مستوى (P < 0.05) بين الذكور والاناث بالاضافة الى المجاميع الاكثر والاقل من اربعة سنوات عمراً. ومن جهة اخرى لوحظ عدم وجود فروق معنوية بين قيم ALT المسجلة للمجاميع المختلفة . والمحدلات بالاضافة الى المجاميع الاكثر والاقل من اربعة سنوات عمراً. ومن جهة اخرى لوحظ عدم وجود فروق معنوية بين قيم ALT المسجلة للمجاميع المختلفة . والمحدلات لي الدراسة قيم ALP المسجلة للمجاميع المختلفة . والمعدلات ± الخطأ القياسي لانزيمات الكبد AST , ALT و ALP في مصل خيول السباق العراقية السوية سريرياً .

# Introduction

Alanine aminotransferase, AST and ALP are closely related enzymes of clinical significance, particularly in the assessment of liver function [1] and widely used as markers for acute and chronic hepatocellular damage due to various causes [2]. ALT is liver specific enzyme presents in high concentration in the cytoplasm of hepatocytes, elevated with hepatocellular damage, necrosis , hepatocyte proliferation or hepatocellular degeneration, while AST found in many tissues including muscles (cardiac and skeletal) , cytoplasm and mitochondria of hepatocytes and is useful in evaluating muscle and liver damage [3]. The ALP presents in high concentration in the liver, spleen, bone, intestinal lining and kidneys . Serum ALP measurement is of great importance in hepatobiliary diseases, and bone diseases associated with increased osteoblastic activity [4]. However, several workers in different countries except in Iraq documented serum levels of these enzymes in horses, and clarified that the normal reference range of ALT, AST and ALP were effected by age, sex, and physiological conditions, among them [5] found that serum ALT in thoroughbred mares and standardbred were  $2.8 \pm 1.4$  U/L and  $4.3 \pm 3.6$ U/L respectively . In 2005 [6] found that serum ALT mean value was 9.92  $\pm$ 2.9 IU/L. The range reference value of serum ALT by [7] was 2.7 - 21 U/L, [8] and [9] was 3 - 23 U/L . Serum AST values for thoroughbred and standardbred were  $165 \pm 33.6$  U/L and  $217 \pm 140$  U/L respectively [5]. Other serum AST reference values were found as follows : 180 - 365 IU/L [10],  $269.81 \pm 74.4$ IU/L [6], 116 – 287 U/L [7], 220 – 600 U/L [8], 226 – 366 U/L [9], 90 – 400 U/L [11] and 346.6 - 399.1 U/L [12] . Moreover , serum ALP values documented by [5] were  $59 \pm 16.7$  U/L ,  $45.6 \pm 10.8$  U/L for thoroughbred and standardbred respectively ,  $422.7 \pm 110.37$  IU/L [6] . The range reference values for serum ALP were ; 68 – 200 IU/L [10] , , 70 – 227 U/L [7] , 100 – 4003 U/L [8] and 66 - 212 U/L [11]. This study was conducted to document the normal reference values of serum ALT, AST and ALP of healthy racing horses in Iraq.

# **Materials and Methods**

Blood samples were drawn at different intervals during the peroid from February to May 2010, from Jugular venipunctures of (125) clinically healthy Iraq racing horses; males (39) and females (86), among these animals (34) were Arabian and (91) Crossbred; aged 2 - 8 years subdivided into < 4, 4 and > 4 age groups from equestrian club / Baghdad. The blood left to clot and then centrifuged for 5 - 10 minutes at 3000 rpm. The separated serum was used directly for the measurement of ALT and AST activity. While, 108 out of the total samples were used for the measurement of serum ALP; males (29) and females (79), Arabian (33) and Crossbred (75), under the same experimental subgroups. Serum activities of ALT and AST were measured by colorimetric method recommended by [13], and serum ALP activity was evaluated according to the colorimetric method of [14]. The SAS [15] program was used for statistical analysis. Significant differences between the means  $\pm$  SEM were

determined using least significant differences (LSD) , and significant level of (P < 0.05) was used in results evaluation .

#### Results

Serum activities of ALT , AST and ALP for horses independent of any subdivision are presented in table (1) . While the values of these enzymes according to sex , breed and age are presented in tables (2) and(3) . Serum ALT activity in healthy Iraqi racing horses was found to be  $11.07 \pm 0.62$  U/L , ranged from 2 - 28 U/L . It was  $11.22 \pm 1.17$  U/L in males and  $11.01 \pm 0.74$  U/L in females with no significant difference between them . The Serum ALT activity was significantly higher (P < 0.05) in Arabian horses compared with Crossbred ,  $13.08 \pm 1.26$  U/L and  $10.30 \pm 0.7$  U/L , respectively. There was no significant differences between serum ALT activity in horses aged < 4 years , 4 and > 4 ,  $11.05 \pm 1.04$  U/L ,  $12.46 \pm 1.18$  U/L and  $9.91 \pm 1.01$  U/L respectively .

On the other hand, the recoded mean value of serum AST activity was found to be  $78.00 \pm 3.62$  U/L , ranged from 30 - 210 U/L . The serum AST activity was significantly higher in males than in females (P < 0.05), it was found to be  $88.43 \pm 7.30$  U/L in males and  $73.26 \pm 4.03$  U/L in females . According to the breed subdivision, serum AST activity did not differ significantly between males and females , 86.85  $\pm$  7.04 U/L and 74.69  $\pm$  4.20 U/L respectively. The serum AST activity was also investigated according to age of horses and was found to be significantly higher in those aged < 4 years when compared to those > 4 years , 95.40  $\pm$  6.57 U/L and 63.60  $\pm$  4.72 U/L respectively (P < 0.05), while the 4 years group was  $79.17 \pm 6.93$ . Moreover, the serum activity of ALP was found to be 96.84  $\pm$  2.52 IU/L with the range of 63 – 204 IU/L . There were no significant differences in serum ALP activity between males and females  $101.44 \pm 5.24$  IU/L and  $95.15 \pm 2.86$  IU/L , Arabian and Crossbred ,  $97.80 \pm 6.98$  IU/L ,  $96.62 \pm 2.68$  IU/L and < 4 , 4 and > 4 years  $,96.57 \pm 4.58 \text{ IU/L}, 93.39 \pm 3.53 \text{ IU/L} \text{ and } 99.92 \pm 4.72 \text{ IU/L} \text{ respectively}$ .

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Enzymes	No. of horses	Range	$Mean \pm SEM$		
Serum ALT U/L	125	2 - 28	$11.07 \pm 0.62$		
Serum AST U/L	125	30 - 210	$\textbf{78.00} \pm \textbf{3.62}$		
Serum ALP IU/L	108	63 - 204	$96.84 \pm 2.52$		

Table (1) : Serum ALT , AST and ALP activities for Iraqi racing horses ; range and mean  $\pm$  SEM .

Table (2) : The serum mean ±SEM of ALT and AST activities according to sex	, breed
and age.	

No. of horses	Factors	ALT U/L	AST U/L
		Mean ± SEM	Mean ± SEM
39	Males	$11.22 \pm 1.17$	a 88.43 ± 7.30
86	Females	$11.01 \pm 0.74$	b 73.26 ± 4.03
34	Arabian	a 13.08 ± 1.26	$86.85 \pm 7.04$
91	Crossbred	b $10.30 \pm 0.70$	$74.69 \pm 4.20$
37	< 4 years	$11.05 \pm 1.04$	a 95.40 ± 6.57
40	4 years	$12.46 \pm 1.18$	ab 79.17 ± 6.93
48	> 4 years	$9.91 \pm 1.01$	b 63.60 ± 4.72

\* Different letters (a, b) mean significantly different at (P < 0.05).

No. of horses	Factors	ALP IU/L
		Mean ± SEM
29	Males	$101.44 \pm 5.24$
79	Females	$95.15 \pm 2.86$
33	Arabian	$97.80 \pm 6.98$
75	Crossbred	$96.62 \pm 2.68$
35	< 4 years	96.57 ± 4.58
33	4 years	$93.39 \pm 3.53$
40	>4 years	$99.92 \pm 4.72$

Table (3) : The serum mean ±SEM of ALP activity according to sex , breed and age .

#### Discussion

Values presented in the tables 1,2 and 3 were obtained from clinically normal racing horses in Baghdad – Iraq in order to provide reference range and means  $\pm$  SEM for serum ALT, AST and ALP which have been not previously measured. The results of the present study showed a significant elevation in ALT activity in Arabian horses compared to Crossbred, while there were no significant differences between males and females nor between different age groups ( table 2). Serum ALT activity was higher than that reported by [5] and [6], while the reference range reported by [7] was within the range recorded in this study. The higher ALT activity may be attributed to genetic factors or perhaps it refers to the type of feeding or addition of micronutrients [16].

The reference range values of ALT reported by [8] and [9] were similar, to a large extent, to that documented in this study. However, there were only four values out of 125 (3.2%) higher than the upper limit of range reference reported by them . On the other hand, serum AST results revealed a significant differences between males and females, as well as between age group more and less than four years old (table 2). The AST mean values  $\pm$  SEM of the four years old horses differ from both less and more than four, so a further study is required to affirm or negate these value differences. In this study the mean  $\pm$ SEM of serum AST is close to the result of [16]. The AST values reported by [5], [10], [6], [7], [8], [9] and [12] did not agree with the result of this study and were higher in range or mean values  $\pm$  SEM; the lowest value in range reference of AST in the above mentioned studies was 90 U/L, while 66.04% of the collected samples of the present study were less than 90 U/L. Moreover, the lowest mean values  $\pm$  SEM of serum AST activity reported by [5] and [6] was  $165 \pm 33.6$  U/L compared to few results of this work which were higher than 165 U/L . The difference which exist between this study and other workers is possibly due to the fact that the Iraqi racing horses have more frequent training, and this may lead to the clearance of AST from serum during exercise [17] . Serum ALP values documented in this study were higher than that reported by [5] and lower than those reported by [6] and [8]; the range values of serum ALP in 69.4 % of tested samples were lower than the lowest range reported by [8]. The serum ALP reported by [10], [7] and [11] were in agreement with the results of this study and were more or less close to them. The existed differences between values of this study as compared with results of other worker's may be attributed to one or the following factors; the absence of scientifically feeding programs, living in hot area, genetic factors and training or exercise [17] and [16].

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