

HAEMATO-BIOCHEMICAL OBSERVATIONS IN APPARENTLY HEALTHY EQUINE SPECIES

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ABSTRACT

The present study was carried out to establish the haematological and biochemical values in apparently healthy equine species kept under field conditions in Pakistan. For this purpose, 415 blood samples were collected from horses (n=242), mules (n=20) and donkeys (n=153) kept in periphery of Faisalabad city. A detailed anamnestic and clinical assessment in the form of a questionnaire was designed for each animal with specific emphasis on age, sex, body condition and lactation. Blood samples were evaluated for haematological and serum biochemical parameters. In general, a non significant difference was noted in all haemato-biochemical parameters in relation to different age groups, sex, body condition and lactation in apparently healthy horses and donkeys. In a comparison between horses, mules and donkeys, a significant difference was observed in PCV, Hb, TLC and fibrinogen. PCV and Hb were highest in the horses and mules, respectively. TLC and fibrinogen were highest in donkeys.

Key words: Haematology, serum biochemistry, horses, mules, donkeys.

INTRODUCTION

Laboratory aids are used extensively in the diagnosis of diseases, in preventive medicine, and as management tools. Laboratory based investigations are subjected to substantial variability arising from several sources, including differences between subjects (e.g., age, sex, and genetic variation), variations in sample collection and handling, and laboratory measurement errors. In interpreting laboratory test results of an individual, the clinician usually compares the observed values with reference values. Inappropriate reference values may increase the risk of either unnecessary additional investigations or failure to detect underlying disease (Soldberg, 1986; Kaneko *et al.*, 1997; Tsang *et al.*, 1998).

The haemato-biochemical values obtained abroad may not be fully applicable under local conditions because these are influenced by multiple factors including breed and environmental and management differences. Some variations also exist in results between laboratories using different reagents, methods, and instruments. Therefore, the purpose of this study was to determine reference haemato-biochemical values for the local equine population kept under field conditions of Pakistan to form a basis for clinical interpretation.

MATERIALS AND METHODS

Sample collection

This study comprised 415 clinically healthy equines including horses (n = 242), mules (n = 20) and donkeys (n = 153) of both sexes, selected randomly from periphery of Faisalabad city. Information about age, parity and body condition for each animal was recorded. Blood samples from each animal were collected with and without anticoagulant. Samples collected with anticoagulant (EDTA) were used for haematological studies. Samples collected without anticoagulant were used for serum extraction. Collected serum was stored at -20°C for its biochemical analysis.

Haemato-biochemical analysis

Blood samples collected with EDTA were analyzed for total erythrocyte counts, total leukocyte counts (TLC), differential leukocyte counts, haemoglobin (Hb), packed cell volume (PCV) and erythrocyte sedimentation rate. Serum total protein contents were determined by Goldberg refractrometer and fibrinogen (Fib) by Schalm method (Benjamin, 1978). For determination of alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) activities, commercially available kits were used following the instructions provided by the manufacturer (Diasis Diagnostik Sistemler Tic. Ve San. A. S. 28 Nisan Cad. Durak Sok. No. 4 34400 Okmeydani-Istanbul, Turkey).

Data analysis

Animals under observation were divided into various groups on the basis of species (horses, donkeys, mules), age (1-5, 6-10, 11-15 years), sex (male, female), lactation status (lactating, non-lactating) and body condition (good, fair, poor). One way analysis of variance (ANOVA) was applied on haematological and biochemical studies to know the differences between various groups. Duncan's multiple range test was applied for multiple means comparisons, where necessary.

RESULTS

A non significant difference was observed in all haemato-biochemical parameters between healthy horses, mules and donkeys except PCV ($P<0.050$), Hb ($P<0.001$), TLC ($P<0.012$) and Fib ($P<0.018$). PCV and Hb were highest in horses, followed by mules and donkeys, while TLC values were highest in donkeys, followed by mules and horses. Fibrinogen was highest in donkeys, followed by horses and mules. So far as the enzymic activity is concerned, the values of the investigated enzymes were generally lower in donkeys than in horses, though the difference was non-significant (Table 1).

A non significant difference was observed in all haemato-biochemical parameters in relation to different age groups, sex, body condition and lactation in horses and donkeys. The comparison on the basis of above mentioned groups in mules could not be made because all mules belonged to one age group (1-5 years) and sex (male).

A significant difference was observed in Hb concentration ($P<0.003$) and TLC ($P<0.002$) when lactating mares were compared with lactating donkeys. The Hb concentration was higher in lactating mares than that of lactating donkeys, while reverse was true for TLC (Table 2). In non-lactating mares and donkeys, all haemato-biochemical parameters showed non-significant difference, except fibrinogen which was significantly higher ($P<0.013$) in non-lactating donkeys than that of the mares (Table 2). So far as the enzymic activity is concerned, the values of the investigated enzymes were generally higher in non-lactating donkeys than in mares, though the difference was non-significant.

DISCUSSION

Haematology has been widely used to provide information about disease status, performance problems, and fitness in horses. Age, sex and breed as well as physical exercise may affect the results of haematologic values (Karazawa and Jamra, 1989; Kaneko *et al.*, 1997). In the present study, in general all haematological and serum biochemical parameters were consistent with previous reports (Duncan *et al.*, 1994; Eades and Bounous, 1997; Meyer and Harvey, 1998; Radostitis *et al.*, 2000; Al-Busadah and Homeida, 2005). The total number of leukocytes showed a significant difference ($P<0.012$) and was highest in the donkeys blood which is in accordance to results reported by Reece (1997) and Al-Busadah and Homeida (2005).

Table 1: Haemato-biochemical findings in apparently healthy equine species

Parameter	Horses	Mules	Donkeys	P value
Total erythrocyte counts ($\times 10^{12}/L$)	5.93 \pm 02.81	5.74 \pm 02.62	5.88 \pm 02.65	0.982
Packed cell volume (%)	37.75 \pm 4.57a	34.37 \pm 4.13b	32.42 \pm 4.38c	0.050
Haemoglobin (g/dL)	10.93 \pm 1.89b	10.10 \pm 0.73c	9.01 \pm 1.13a	0.001
Erythrocyte sedimentation rate (mm/hr)	105.13 \pm 24.59	99.63 \pm 23.13	104.83 \pm 15.66	0.822
Total leukocyte counts ($\times 10^9/L$)	7.18 \pm 2.87c	9.40 \pm 2.89a	9.75 \pm 3.29b	0.012
Neutrophils (%)	42.05 \pm 13.94	69.94 \pm 00.00	39.73 \pm 21.27	0.234
Lymphocytes (%)	45.73 \pm 15.74	27.98 \pm 00.00	43.47 \pm 19.32	0.579
Monocytes (%)	1.28 \pm 01.92	1.39 \pm 00.00	0.72 \pm 00.89	0.657
Eosinophils (%)	11.10 \pm 05.75	-	15.83 \pm 10.11	0.090
Basophils (%)	0.42 \pm 00.56	0.34 \pm 00.48	0.24 \pm 00.41	0.645
Total proteins (g/dL)	8.63 \pm 00.95	8.50 \pm 00.77	8.11 \pm 00.58	0.156
Fibrinogen (mg/dL)	630.40 \pm 208.60a	366.70 \pm 196.60b	692.30 \pm 203.80c	0.003
Alanine aminotransferase (U/L)	25.24 \pm 05.84	26.88 \pm 01.35	26.26 \pm 01.40	0.819
Aspartate aminotransferase (U/L)	229.67 \pm 46.81	220.14 \pm 42.00	223.30 \pm 32.78	0.861
Alkaline phosphatase (U/L)	490.94 \pm 82.39	419.38 \pm 57.48	485.46 \pm 98.74	0.288

Values bearing different letters in a row differed significantly.

Table 2: Haemato-biochemical findings in lactating and non-lactating mares and donkeys

Parameter	Mares	Donkeys	P value
Lactating			
Total erythrocyte counts (X 10 ¹² /L)	6.35 ± 03.06	5.62 ± 02.21	0.512
Packed cell volume (%)	37.88 ± 07.84	32.66 ± 06.42	0.060
Haemoglobin (g/dL)	11.01 ± 1.76a	9.04 ± 1.04b	0.003
Erythrocyte sedimentation rate (mm/hr)	101.21 ± 22.91	109.86 ± 13.45	0.343
Total leukocyte counts (X 10 ⁹ /L)	6.42 ± 1.89b	10.58 ± 2.81a	0.002
Neutrophils (%)	40.61 ± 12.67	40.63 ± 23.63	0.998
Lymphocytes (%)	47.15 ± 12.97	41.80 ± 21.04	0.457
Monocytes (%)	1.24 ± 02.20	0.43 ± 00.58	0.299
Eosinophils (%)	11.32 ± 14.06	16.94 ± 10.83	0.091
Basophils (%)	0.43 ± 00.63	0.19 ± 00.38	0.314
Total proteins (g/dL)	8.74 ± 00.97	8.11 ± 00.57	0.074
Fibrinogen (mg/dL)	633.30 ± 205.60	655.60 ± 188.20	0.755
Alanine aminotransferase (U/L)	26.51 ± 07.09	26.01 ± 07.28	0.845
Aspartate aminotransferase (U/L)	225.46 ± 57.05	217.84 ± 34.38	0.729
Alkaline phosphatase (U/L)	480.45 ± 65.93	508.94 ± 65.78	0.315
Non-lactating			
Total erythrocyte counts (X 10 ¹² /L)	5.26 ± 02.26	7.03 ± 05.31	0.631
Packed cell volume (%)	37.76 ± 10.32	30.50 ± 06.03	0.289
Haemoglobin (g/dL)	10.76 ± 02.21	8.80 ± 01.63	0.194
Erythrocyte sedimentation rate (mm/hr)	95.68 ± 21.42	99.75 ± 19.26	0.883
Total leukocyte counts (X 10 ⁹ /L)	8.49 ± 03.84	5.97 ± 03.28	0.486
Neutrophils (%)	42.98 ± 16.88	35.70 ± 14.23	0.255
Lymphocytes (%)	44.67 ± 21.02	50.95 ± 17.24	0.651
Monocytes (%)	1.28 ± 01.43	2.00 ± 01.16	0.824
Eosinophils (%)	10.92 ± 04.06	10.88 ± 06.83	0.091
Basophils (%)	0.45 ± 00.48	0.46 ± 00.65	0.962
Total proteins (g/dL)	8.41 ± 00.92	8.05 ± 00.73	0.688
Fibrinogen (mg/dL)	623.10 ± 224.20	775.00 ± 195.70	0.113
Alanine aminotransferase (U/L)	23.63 ± 03.07	27.25 ± 01.91	0.580
Aspartate aminotransferase (U/L)	238.37 ± 26.15	245.14 ± 14.22	0.334
Alkaline phosphatase (U/L)	480.45 ± 65.93	508.94 ± 65.78	0.315

Values bearing different letters in a row differed significantly.

In relation to different age groups, sex, body condition and parity, a non significant difference was observed in all haematolo-biochemical parameters in apparently healthy horses and donkeys in this study. According to Dinev and Khubenov (1986), normal values of haemoglobin and erythrocytes of young animals are lower than adults. With aging, the total count of leukocytes shows a lowering trend, while the percent of neutrophils rises. Serum total protein content is lower in young donkeys than older ones. However, Zinkl *et al.* (1990) reported that erythrocytes, lymphocytes and fibrinogen decreased with age. Sex and lactation had no effect on haemato-biochemical parameters in healthy animals (Dinev and Khubenov, 1986; Zinkle *et al.*, 1990; Harvey *et al.*, 1994).

In general, the alanine aminotransferase and alkaline phosphatase enzymes were higher than those reported by Meyer and Harvey (1998), Radostitis *et al.* (2000), Mohri *et al.* (2005) and Al-Busadah and Homeida (2005). An interaction between age groups was observed in horses for alkaline phosphatase activity, with a trend for decreased activity with age (Zinkl *et al.*, 1990).

REFERENCES

- AL-Busadah, K. A. and A. M. Homeida, 2005. Some physical variables, biochemical and haematological parameters in Hassawi ass. *Sci. J. King Faisal Univ.*, 6: 145-151.

- Benjamin, M. M., 1978. Outline of Veterinary Clinical Pathology. 3rd Ed., Iowa State Univ. Press, Ames, Iowa, USA.
- Dinev, D. and K. D. Khubenov, 1986. Normal values of the hematological, biochemical and enzymological indices of the donkey. *Vet. Med. Nauki.*, 23: 69-75.
- Duncan, J. R., K. W. Prasse and E. A. Mahaffey, 1994. Veterinary Laboratory Medicine: Clinical Pathology. 3rd Ed., Iowa State Univ. Press, Ames, Iowa, USA.
- Eades, S. C. and D. I. Bounous, 1997. Laboratory Profile of Equine Diseases. Mosby Year Book. Saint Louis, Missouri, USA.
- Harvey, J. W., R. L. Asquith, M. G. Patel, J. Kivipelto, C. L. Chen and E. A. Ott, 1994. Haematological findings in pregnant, postparturient and nursing mares. *Comp. Hema. Intern.*, 4: 25-29.
- Kaneko, J. J., J. W. Harvey and M. L. Bruss, 1997. Clinical Biochemistry of Domestic Animals. 5th Ed., Academic Press, San Diego, USA. pp: 485-516.
- Karazawa, E. H. and M. Jamra, 1989. Normal hematological parameters. *Rev. Saude. Publ.*, 23: 58-66.
- Meyer, D. J. and J. W. Harvey, 1998. Veterinary Laboratory Medicine: Interpretation and Diagnosis. 2nd Ed., Saunders, Philadelphia, USA.
- Mohri, M., K. Sardari and N. Farzaneh, 2005. Serum biochemistry of Iranian Turkmen (Akhhal-Teke) horses. *Comp. Clin. Pathol.*, 13: 128-131.
- Radostits, O. M., C. C. Gay, D. C. Blood and K. W. Hinchcliff, 2000. Veterinary Medicine, 9th Ed., ELBS Bailliere Tindall, Philadelphia, USA, pp: 870-871.
- Reece, W. O., 1997. Physiology of Domestic Animals. Williams and Wilkins, New York, USA.
- Soldberg, H. E., 1986. Approved recommendation on the theory of reference values, part 1. The concept of reference values. *Clin. Chem. Acta*, 165: 111-118.
- Tsang, C. W., R. Lazarus, W. Smith, P. Mitchell, J. Koutts and L. Burnett, 1998. Hematological indices in an older population sample: derivation of healthy reference values. *Clin. Chem.*, 44: 96-101.
- Zinkl, J. G., D. Mae, P. G. Merida, T. B. Faver and J. A. Humble, 1990. Reference ranges and influence of age and sex on hematologic and serum biochemical values in donkeys (*Equus asinus*). *Amer. J. Vet. Res.*, 51: 408-413.