



Original Research

Hematology and Clinical Chemistry in Amiata Donkey Foals from Birth to 2 Months of Age

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ABSTRACT

The aim of the present study was to fill the void in data related to hematological and biochemical parameters of donkey foals. Whole-blood and plasma samples were obtained from 16 Amiata donkey foals at birth, at 24 and 48 hours, and at 1, 2, 3, 4, 6, and 8 weeks of age. RBC, WBC, hemoglobin concentration (Hgb), mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, platelet count, glucose, creatinine, blood urea, triglycerides, total cholesterol, total bilirubin, aspartate aminotransferase, γ -glutamyl transferase, creatine-phosphokinase, alkaline phosphatase, total calcium, potassium, sodium, phosphorus, and albumin were measured. Similar to equine foals, values of RBC, Hgb, and Hct decreased significantly after the first 48 hours of life in Amiata donkey foals, reaching values similar to adult donkeys. No changes were found for mean corpuscular volume and mean corpuscular hemoglobin concentration during the study period. The WBC was low at birth when compared with subsequent measurements, but significantly increased in the subsequent surveys. Platelet count was lower in the first week, with a secondary peak 2 weeks later, and then a decline again up to the eighth week. In agreement with equine foals, electrolyte concentrations, triglycerides, and total cholesterol did not show significant differences. Creatinine, total bilirubin, and blood urea showed a trend similar to RBC, Hgb, and Hct. For the first time, data of hematological and biochemical parameters in Amiata donkey foals are provided.

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1. Introduction

Donkeys (*Equus asinus*) have been close companions to humans for millennia and have been used as working animals all over the world. Nowadays, the new possible uses of donkeys could be production of milk for children who are intolerant to cow's milk [1,2] or in animal-assisted therapy and activity [3].

The renewed interest for these animals has been demonstrated by studies carried out to establish baseline

data of both hematological and biochemical variables in the blood of adult donkeys [4-21].

In horses, age-related changes in hematological parameters preclude the use of adult normal values in the evaluation of foals. To the best of our knowledge, no data regarding hematological and biochemical values in Amiata donkey foals have been provided to date. Thus, the need for a reference range of values for specific ages in Amiata donkey foals becomes evident. Therefore, the aim of this study was to follow the changes of hematological and biochemical parameters in Amiata donkey foals from birth up to the second month of life to verify age-related changes, as reported in equine foals [22-29], to clarify whether dedicated reference ranges are required in younger animals.

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2. Material and Methods

Sixteen Amiata donkey foals (nine fillies and seven colts) belonging to the Regional Study Centre, Tuscany, Italy, were included in the study. Amiata donkey is one of the Italian breeds prevalently present on Amiata mountains in the center of Italy. In 1990, the Biodiversity Committee of European Parliament included the Amiata donkey in the list of endangered breeds (NL 215/90). All the donkey foals were born at the Veterinary Teaching Hospital “Mario Modenato,” Faculty of Veterinary Medicine, Pisa (Italy), between 2004 and 2010. Approval to conduct this study was obtained from the Ethics Committee on Animal Experimentation of the University of Pisa (DL 116/92). Foals and their mares underwent similar management conditions. Donkey foals have been included in this study on the basis of the following criteria: (1) pregnancy length ≥ 372 days [30]; (2) unassisted delivery; (3) jennies treated against gastrointestinal parasites and vaccinated against equine influenza, tetanus, and equine herpes virus-1, according to guidelines of the American Association of Equine Practitioners Infectious Disease Committee [31]; (4) Apgar score ≥ 7 , 5 minutes after birth [32]; (5) IgG ≥ 800 mg/dL at 24 hours of age (Snap Foal IgG test Kit, Idexx, Westbrook, ME) [33]; righting reflex present immediately after foaling, suck reflex within 10 minutes, sternal recumbency within 5 minutes [34], quadrupedal position within 127.5 ± 70 minutes, and nursing the mare within 200 ± 67.4 minutes after birth [35]. Foals were physically examined before each blood collection, and they appeared to be clinically healthy during the study period.

Blood samples were obtained from the jugular vein from each foal at (1) birth (T0) before colostrum consumption; (2) 24 hours of life; (3) 48 hours of life; (4) 1 week; (5) 2 weeks; (6) 3 weeks; (7) 4 weeks; (8) 6 weeks; and (9) 8 weeks of age. Blood was collected into test tubes containing potassium salt (2 molecules of) of ethylenediaminetetraacetic acid [K2-EDTA] (code 22056, FL Medical, Padua, Italy) and in lithium-heparin test tubes (code 22304, FL Medical, Padua, Italy). To avoid alterations related to diurnal variations, blood samples were collected at the same time each day (8:00–9:00 AM).

K2-EDTA samples were analyzed with a cell counter (Hecovet C 01030360/ITA, and CAL-SEAC 71010810 multiparametric hematology calibrator, SEAC-RADIM Co, Florence, Italy) at least 5 minutes after the collection, so as to allow a full contact and interactions between blood cells and anticoagulant, to determine (1) erythrocyte count (RBC), (2) leukocyte count (WBC), (3) hemoglobin concentration (Hgb), (4) mean corpuscular volume (MCV), (5) mean corpuscular hemoglobin (MCH), (6) mean corpuscular hemoglobin concentration (MCHC), and (7) PLT.

Heparinized samples were centrifuged at 3,000 g for 10 minutes, as recommended by the manufacturer, and then plasma was frozen at -18°C and analyzed in a single batch. Clinical chemistry was performed with an autoanalyzer (Liasys, Analyzer Medical System-AMS, Rome, Italy; quality control normal level: ASR02010, pathologic level: ASR02020, Assel Srl, Rome, Italy). The parameters analysed were (1) glucose concentration (Glucose SL, enzymatic colorimetric method, code ASR01202, Assel Srl, Rome, Italy); (2) creatinine (Creatinine, kinetic modified Jaffè method,

code ASR01150, Assel Srl, Rome, Italy); (3) blood urea (Urea UV SL, kinetic enzymatic method, code ASR01143, Assel Srl, Rome, Italy); (4) triglycerides (Triglycerides-SL, enzymatic colorimetric method, code ASR01134, Assel Srl, Rome, Italy); (5) total cholesterol (Cholesterol liquid, trinder method-endpoint, code 7050, FAR, Verona, Italy); (6) total bilirubin (Total Bilirubin, colorimetric method without DMSO, code ASR01034/1, Assel Srl, Rome, Italy); (7) aspartate aminotransferase (AST) (AST SL, kinetic method UV -IFCC- code ASR01220, Assel Srl, Rome, Italy); (8) gamma-glutamyl-transferase (γ -GT) (Gamma GT SL, kinetic method-Szasz-Tris, code ASR01194, Assel Srl, Rome, Italy); (9) creatine-phosphokinase (CK) (CK NAC SL, kinetic method UV, code ASR01074, Assel Srl, Rome, Italy); (10) alkaline phosphatase (ALP) (Alkaline Phosphatase SL-DGKC- kinetic method, code ASR01162, Assel Srl, Rome, Italy); (11) total calcium (Calcium OCPC, colorimetric method, code ASR01050, Assel Srl, Rome, Italy); (12) potassium (pHox Plus L, Stat Profile, Nova Biochemical, Milan, Italy); (13) sodium (pHox Plus L, Stat Profile, Nova Biochemical, Milan, Italy); (14) phosphorus (P) (Phosphorus UV, direct method with molybdate, code 90009800, SEAC-RADIM, Florence, Italy); and (15) albumin (Albumin BCG, bromocresol green method, code 90009781, SEAC-RADIM, Florence, Italy). The same operator always performed the biochemistry profile, according to standard methods.

2.1. Statistical Analysis

Average and standard deviation were calculated for each hematological and biochemical parameter at all sampling times. Kurtosis and Skewness coefficients were calculated to verify data distribution. Data distribution was normal; thus, analysis of variance (ANOVA) for repeated measurements and Bonferroni test as post hoc were performed for each hematological and biochemical parameter at all sampling times. Significance level was set at $P < .05$.

3. Results

The results for hematological and biochemical parameters, expressed as average \pm standard deviation, of Amiata donkey foals are reported in Tables 1 and 2, respectively.

Regarding complete blood count, ANOVA and Bonferroni test showed statistical differences for RBC, WBC, Hgb, Hct, MCH, PLT, whereas no differences were shown for MCV and MCHC.

Regarding biochemical parameters, ANOVA and Bonferroni test showed statistical differences related to sampling time for plasma glucose, creatinine, urea and triglycerides plasma concentrations, and for AST, CK, γ GT, and ALP activities. No statistical differences were obtained for total cholesterol, total bilirubin, and albumin plasma concentrations.

No statistical differences related to time were obtained for electrolytes.

4. Discussion and Conclusions

To our knowledge, this is the first report on hematological and biochemical parameters in Amiata donkey foals during the first 2 months of life. These data could be useful

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