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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 \geq 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 \geq 7, 5 minutes after birth [32]; (5) IgG \geq 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], guadrupedal position within 127.5 \pm 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 methodendpoint, 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 (y-GT) (Gamma GT SL, kinetic method-Szasz-Tris, code ASR01194, Assel Srl, Rome, Italy); (9) creatinephosphokinase (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

Table 1
Hematological parameters expressed by average \pm standard (SD) deviation in 16 Amiata donkey foals from birth to 2 months of life

Hematological parameters	ТО	T24	T48	T1w	T2w	T3w	T4w	T6w	T8w	ANOVA test
RBC (M/ìL)	9.9 ± 0.8^{b}	$8.9\pm1.9^{a,b}$	$7.4\pm1.7^{a,b}$	7.4 ± 1.3^{a}	$\textbf{6.7} \pm \textbf{1.4}^{a}$	7.1 ± 1.2^{a}	7.7 ± 1.0^{a}	6.7 ± 1.5^a	7.1 ± 1.0^{a}	P < .05
WBC (K/ìL)	5.9 ± 1.5^{a}	$7.3\pm0.9^{\rm a,b}$	$7.9 \pm 1.7^{a,b,c}$	$8.4\pm0.7^{\rm b,c}$	$8.4\pm1.2^{ m b,c}$	$8.7 \pm 1.0^{ m b,c}$	$8.3 \pm 1.4^{ m b,c}$	10.1 ± 0.8^{c}	10 ± 0.5^{c}	P < .05
Hgb (gr/L)	16.8 ± 1.1^{c}	$15.1 \pm 2.3^{b,c}$	$13.5\pm0.8^{a,b}$	$12.6\pm1.7^{\text{a,b}}$	11.2 ± 0.8^{a}	12.5 ± 1.2^{a}	11.4 ± 1.1^{a}	10.6 ± 0.7^a	10.9 ± 0.1^a	P < .05
Hct (%)	50.2 ± 2.6^{c}	$42.5 \pm 1.3^{b,c}$	$40.0\pm2.4^{b,c}$	$35.3\pm2.0^{a,b}$	$31.8 \pm 2.4^{a,b}$	$35.2\pm4.0^{a,b}$	29.3 ± 1.1^{a}	29.6 ± 2.7^a	30.5 ± 1.4^{a}	P < .05
MCV (Fl)	49.8 ± 2	50.7 ± 2.1	50.3 ± 2.9	49.7 ± 2.3	49.2 ± 2.7	46.3 ± 2.0	42.6 ± 1.5	$\textbf{42.8} \pm \textbf{1.9}$	40.8 ± 1.8	NS
MCH (Pg)	$17.1\pm0.9^{a,b}$	$17.2 \pm 1.1^{a,b}$	$17.1 \pm 1.1^{a,b}$	$17.0\pm0.9^{a,b}$	$17.6\pm1.7^{a,b}$	$16.6\pm0.9^{a,b}$	$16.2\pm0.7^{a,b}$	$15.9\pm0.8^{a,b}$	15.3 ± 0.7^a	P < .05
MCHC (gr/dL)	34.3 ± 2.0	33.9 ± 1.8	34.2 ± 1.3	34.2 ± 1.2	35.1 ± 0.7	35.8 ± 1.9	37.3 ± 1.5	36.5 ± 1.5	$\textbf{37.4} \pm \textbf{0.7}$	NS
PLT (K/ìL)	$\textbf{342.0} \pm \textbf{58.2}^{c}$	$285.6 \pm 43.9^{b,c}$	$259\pm27.8^{a,b,c}$	$254.7 \pm 32.5^{a,b,c}$	$263.3 \pm 53.9^{a,b,c}$	$322.7 \pm 59.0^{b,c}$	$267.7 \pm 51.7^{a,b,c}$	$222.2\pm41.5^{a,b}$	180.8 ± 20.3^a	P < .05

T0, birth; T24h, 24 hours of life; T48h, 48 hours; T1w, 1 week; T2w, 2 weeks; T3w, 3 weeks; T4w, 4 weeks; T6w, 6 weeks; T8w, 8 weeks of age.

Bonferroni test: within row, different superscripts (a, b, c) denote a significant difference (A \neq B \neq C: P < .05).

Table 2

Clinical chemistry parameters expressed by average \pm standard (SD) deviation in 16 Amiata donkey foals from birth to 2 months of life

Clinical chemistry parameters	T0	T24	T48	T1w	T2w	T3w	T4w	T6w	T8w	ANOVA test
Glucose (mg/dL)	$97.6 \pm 37.1^{a,b}$	$128.6 \pm 15.7^{a,b}$	$126.6 \pm 7.4^{a,b}$	$109.9 \pm 22.6^{a,b}$	$121.2 \pm 37.4^{a,b}$	86.4 ± 12.4^{a}	130.0 ± 19.4^{b}	$103.3 \pm 18.1^{a,b}$	$109.6 \pm 10.5^{a,b}$	P < .05
Creatinine (mg/dL)	$\textbf{2.4}\pm\textbf{0.7}^{a}$	1.3 ± 0.3^{b}	1.2 ± 0.3^{b}	$0.9\pm0.1^{\rm b}$	1.0 ± 0.1^{b}	$1.2\pm0.2^{\rm b}$	1.2 ± 0.3^{b}	1.3 ± 0.2^{b}	1.2 ± 0.3^{b}	P < .05
Urea (mg/dL)	$\textbf{38.4} \pm \textbf{14.1}^{a}$	$\textbf{32.9} \pm \textbf{9.0}^{a,b}$	$28.3\pm8.7^{a,b}$	$\textbf{23.4} \pm \textbf{6.3}^{a,b}$	19.6 ± 5.3^{b}	$25.0\pm1.7^{a,b}$	$24.6\pm10.6^{a,b}$	$27.1 \pm 7.5^{a,b}$	$\textbf{22.6} \pm \textbf{4.5}^{b}$	P < .05
Trygliceride (mg/dL)	34.8 ± 7.2	46.0 ± 22.0	58.0 ± 38.6	60.6 ± 30.4	81.6 ± 42.0	59.2 ± 12.7	61.0 ± 14.0	$\textbf{52.4} \pm \textbf{29.2}$	$\textbf{76.6} \pm \textbf{23.4}$	NS
Total cholesterol (mg/dL)	151.0 ± 29.2	161.7 ± 32.8	157.4 ± 39.6	141.4 ± 26.9	135.8 ± 43.4	141.8 ± 61.4	123.1 ± 41.0	118.3 ± 41.2	115.8 ± 13.9	NS
Total bilirubin (mg/dL)	$0.3\pm0.2^{a,b}$	$0.3\pm0.1^{a,b}$	$0.4\pm0.1^{a,b}$	0.5 ± 0.3^{b}	$0.2\pm0.1^{a,b}$	0.2 ± 0.1^a	0.1 ± 0.03^a	$\textbf{0.2}\pm\textbf{0.04}^{a}$	$0.2\pm0.07^{a,b}$	P < .05
AST (U/L)	$\textbf{60.3} \pm \textbf{14.6}^{a}$	$107.9 \pm 26.3^{a,b}$	$125.1 \pm 57.7^{a,b}$	$109.7 \pm 43.4^{ m a,b}$	$89.3\pm21.8^{a,b}$	$\textbf{88.6} \pm \textbf{16.2}^{a}$	$114.0 \pm 31.9^{a,b}$	$137.1 \pm 66.6^{a,b}$	165.3 ± 38.3^{b}	P < .05
γGT (U/L)	$20.5\pm7.9^{a,b}$	35.5 ± 17.0^{b}	$29.4 \pm 11.3^{a,b}$	$\textbf{23.2} \pm \textbf{11.4}^{a,b}$	$29.6\pm6.7^{a,b}$	$25.9\pm10.2^{a,b}$	14.3 ± 10.0^a	13.8 ± 5.7^a	12.5 ± 6.0^a	P < .05
CK (U/L)	$\textbf{80.8} \pm \textbf{41.1}^{c}$	$\textbf{20.2} \pm \textbf{16.4}^{a}$	17.4 ± 13.5^{a}	$57.4\pm24.0^{\text{a,b,c}}$	$\textbf{22.9} \pm \textbf{15.3}^{a,b}$	$40.7\pm25.5^{a,b,c}$	$73.0 \pm 36.3^{b,c}$	$61.7\pm21.8^{a,b,c}$	$44.6\pm8.6^{a,b,c}$	P < .05
ALP (U/L)	879.7 ± 658.3^{c}	1041.6 ± 408.1^{c}	$656.0 \pm 233.4^{a,b,c}$	$378.6 \pm 152.9^{a,b}$	326.7 ± 111.1^{a}	${\bf 387.8} \pm {\bf 40.4}^{a,b}$	${\bf 372.9} \pm {\bf 49.0}^{a,b}$	$407.0 \pm 132.5^{a,b}$	$415.5 \pm 57.1^{a,b}$	P < .05
Total Ca ⁺ (mg/dL)	10.7 ± 1.6	10.8 ± 0.9	10.9 ± 1.1	11.3 ± 2.3	10.9 ± 1.8	11.6 ± 1.3	12.0 ± 2.1	11.2 ± 1.3	11.0 ± 1.4	NS
K ⁺ (mmol/L)	4.5 ± 0.6	$\textbf{4.3} \pm \textbf{0.8}$	4.5 ± 0.6	$\textbf{4.2}\pm\textbf{0.5}$	$\textbf{4.4} \pm \textbf{0.3}$	$\textbf{4.5} \pm \textbf{0.2}$	$\textbf{4.5} \pm \textbf{0.3}$	$\textbf{5.0} \pm \textbf{0.5}$	$\textbf{4.6} \pm \textbf{0.1}$	NS
Na ⁺ (mmol/L)	132.5 ± 11.6	136.6 ± 3.7	135.0 ± 3.8	133.9 ± 4.1	133.3 ± 2.3	134.0 ± 3.6	132.5 ± 4.7	133.3 ± 1.5	132.3 ± 1.4	NS
P (mg/dL)	$\textbf{4.7} \pm \textbf{1.4}$	4.9 ± 1.0	5.6 ± 1.7	$\textbf{7.4} \pm \textbf{2.4}$	5.0 ± 2.3	$\textbf{6.5} \pm \textbf{1.2}$	$\textbf{6.4} \pm \textbf{1.7}$	$\textbf{6.6} \pm \textbf{1.2}$	6.1 ± 1.1	NS
Albumin (g/L)	$\textbf{3.2}\pm\textbf{0.6}$	$\textbf{3.2}\pm\textbf{0.6}$	$\textbf{3.2}\pm\textbf{0.8}$	$\textbf{3.1} \pm \textbf{0.7}$	$\textbf{3.0} \pm \textbf{0.9}$	$\textbf{3.2}\pm\textbf{0.2}$	$\textbf{2.8} \pm \textbf{0.6}$	3.5 ± 0.7	$\textbf{3.0} \pm \textbf{0.8}$	NS

T0, birth; T24h, 24 hours of life; T48h, 48 hours; T1w, 1 week; T2w, 2 weeks; T3w, 3 weeks; T4w, 4 weeks; T6w, 6 weeks; T8w, 8 weeks of age. Bonferroni test: within row, different superscripts (a, b, c) denote a significant difference ($A \neq B \neq C$: P < .05).

in clinical practice to assess not only the healthy donkey foals but also the ones suffering from perinatal diseases.

In Amiata donkey foals, RBC, Hgb, and Hct values decreased significantly after the first 48 hours of life, and then were constant during the entire study period, and were in agreement with those reported earlier in equine foals [22,24,33]. The reduction observed in those hematological parameters may be related to hemodilution, physiological destruction of erythrocytes by the spleen, and decreased erythropoietin production secondary to increased blood oxygenation by lungs after birth [22]. After the first week of life, RBC, Hgb, and Hct in Amiata donkey foals were similar to values reported in the Amiata adult ones [21], as well as in adult donkeys of different breeds [5,7,8,12,14,16,17]. No changes were found for MCV and MCHC during the study period. Some authors [22] reported a minimum MCV occurring at 4 months of life. In our study, donkey foals were investigated for the first 2 months of life when MCV, as in equine foals, remains constant and similar to the normal adult range, probably because of the concurrent presence of microcytes and macrocytes [22]. The stability of MCHC reflected the Hgb and Hct values.

The WBC count at birth in Amiata donkey foals was similar to equine foals [22,24-28], but lower compared with adult Amiata donkeys [21] and donkeys of other breeds [8,13,14,17]. WBC trend showed a lower count at birth, but a significant increase in the subsequent surveys as reported in equine foals [22]. WBC values in the foal at 1 week of life become similar to that in adults[8]. This might be due to cortisol surge immediately after birth [33] and then the increased bone marrow production related to environment exposure.

PLT at birth was slightly higher in Amiata donkey foals than in equine foals, but this value was similar at 1 week of life [22,24-28]. In our study, PLT declined in the first week, with a secondary peak 2 weeks later, and then declined again up to the eighth week. On the contrary, in equine foals, PLT counts were slightly higher from 2 weeks to 3 months of age. PLT counts in donkey foals become similar to adult Amiata donkey values [21] at 1 month of life, as well as adult donkeys of other breeds [8,14].

Plasma glucose concentration was higher at birth in donkeys than in equine foals [26]. Interestingly, after 24 hours up to 2 months of life, this value matches the two aforementioned species [23]. Plasma glucose concentration was higher than the adult donkey reference values [13,14] through the whole study period. There are no published data concerning blood glucose concentration in Amiata adult donkeys; however, in a group of 10 Amiata jennies, biochemical parameters were evaluated during a 2-month study period (first 2 month of lactation). In this group, blood glucose concentration was lower compared with Amiata donkey foals (Sgorbini et al., unpublished data).

Similar to equine foals, creatinine in Amiata donkey foals was high at birth [27], but declined rapidly to adult donkey reference values, both for Amiata breed and other breeds, at 24 hours of age [7,9,11,13,14,16,17,21]. An increased creatinine value after birth may reflect placental pathology from the mare rather than disease in the foal, as suggested in babies and in equine foals [33,36]. Interestingly, in equine foals, creatinine values decreased slower compared with Amiata donkey foals [23]. Blood urea concentration showed a similar trend as creatinine; thus, it declined within 24 hours of age, then remained constant during the first 2 months of life, and then similar to values reported both in adult Amiata donkeys and adult donkeys of other breeds [7,9,11,13,14,16,21]. As for creatinine, for blood urea also, the higher value after birth may reflect placental pathology and not foals' diseases, as suggested in babies and in equine foals [33,36].

Total bilirubin was higher at birth and remained constant up to the third week, and then decreased at adult ranges [9,14,16,21], similar to equine foals [23]. This could be caused by erythrocyte destruction during perinatal period.

AST was low at birth and increased at 24 hours of age, similar to equine foals [23,33]. Interestingly, AST activity in foals remained lower than in adult donkey of Amiata breed, as well as other breeds [9,11,16,21], during the entire study period, whereas in equine foals values are similar to adult horses at 1 week of life [23,33].

 γ -GT values were similar to those seen in equine foals [33] till up to 1 month of life, and then decreased at levels of adult donkey of different breeds [14]. On the other hand, if compared with Amiata adult donkey values, γ -GT values in Amiata donkey foals were lower for the entire study period [21].

CK activity resulted high at birth, and then declined and remained constant during the 2 months of the study. This enzyme is within the adult range in the equine foal [33], whereas in our study, it is constantly lower than in adult donkeys of Amiata breed and other breeds [9,13,14,16,21].

Similarly to equine foals, ALP activity in Amiata donkey foals was high during the first month of life. This enzyme is markedly elevated because of osteblastic activity and intestinal pinocytosis during the first 24 hours of life [23,33].

Electrolyte concentrations determined in this series showed no age-related differences during the entire study period, and values are similar in donkey and equine foals [33], as well as in adult donkeys [9,13,14,16,21].

No age-related differences were also found for triglycerides and total cholesterol. Triglyceride values were similar to equine foals [33] and adult donkeys [9,14], as well as Amiata jennies (Sgorbini et al, unpublished data), whereas total cholesterol was higher [9,14].

These work reported a reference database of hematological and biochemical values for clinically normal Amiata donkey foals at sequential stages of their life, and this database could be useful in clinical practice to assess foals suffering from diseases.

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References

 Carroccio A, Cavataio F, Montalto G. Intolerance to hydrolysed cow's milk proteins in infants: Clinical characteristic and dietary treatment. Clin Exp Allergy 2000;30:1597-603.

- [2] Muraro MA, Giampietro PG, Galli E. Soy formulas and nonbovine milk. Ann Allergy Asthma Immunol 2002;89(6 Suppl 1):97-101.
- [3] Borioni M, Marinaro P, Celestini S, Del Sole F, Magro R, Zoppi D, et al. Effect of equestrian therapy and onotherapy in physical and psychosocial performances of adults with intellectual disability: A preliminary study of evaluation tools based on the ICF classification. Disabil Rehabil 2012;34:279-97.
- [4] Wilson RT, Payne WJA. An introduction in animal husbandry in the tropic. London: United Kingdom; 1990. p. 581-603.
- [5] Zinkl JC, Mae D, Guzman P, Farver TB, Humble JA. Reference ranges and the influence of age and sex on haematological and serum biochemicals values in donkey (*Equus asinus*). Am J Vet Res 1990;51:408-13.
- [6] Pearson RA, Nengomasha E, Krecek RC. The challenges in using donkeys for work in Africa. In: Proceedings of the ATNESA Workshop "Meeting the challenges of animal traction". Karen, Ngong Hills, Kenya; 1995. p. 190-197.
- [7] French JM, Patrick VH. Reference values for physiological, haematological and biochemical parameters in domestic donkeys (*Equus asinus*). Equine Vet Educ 1995;7:33-5.
- [8] Folch P, Jordana J, Cuenca R. Reference ranges and the influence of age and sex on haematological values of the endangered Catalonian donkey. Vet J 1997;154:163-8.
- [9] Jordana J, Folch P, Cuenca R. Clinical biochemical parameters of the endangered Catalonian donkey breed: Normal values and the influence of sex, age, and management practices effect. Res Vet Sci 1998;64:7-10.
- [10] Aganga AA, Letso M, Aganga AO. Feeding donkeys. Livest Res Rural Dev 2000;12:1-3.
- [11] Mori E, Fernandes WR, Mirandola RM, Kubo G, Ferreira RR, Oliveira JV, et al. Reference values on serum biochemical parameters of Brazilian donkey (*Equus asinus*) breed. J Equine Vet Sci 2003;23: 358-64.
- [12] Mori E, Mirandola RM, Ferreira RR, Oliveira JV, Gacek F, Fernandes WR. Reference values on hematologic parameters of the Brazilian donkey (*Equus asinus*) breed. J Equine Vet Sci 2004;24:271-6.
- [13] Al-Busadah KA, Homeida AM. Some physical variables, biochemical and haematological parameters in Hassawi ass. Sci J King Faisal Univ 2005;6:145-52.
- [14] Caldin M, Furlanello T, Solano-Gallego L, De Lorenzi D, Carli E, Tasca S, et al. Reference ranges for hematology, biochemical profile and electrophoresis in a single herd of Ragusana donkeys from Sicily (Italy). Comp Clin Path 2005;14:5-12.
- [15] Lopez I, Estepa JC, Mendoza FJ, Rodriguez M, Aguilera-Tejero E. Serum concentrations of calcium, phosphorus, magnesium and calciotropic hormones in donkeys. J Am Vet Res 2006;67:1333-6.
- [16] Pitel PH, Moulin M. Approche des valeurs hematologiques et biochimiques chez deux races asinines. Pratique Veterinaire Equine 2006;38:19-25.
- [17] De Aluja AS, Mondragon RL, Castillo DA, Ochoa P. Hematological and biochemical reference values in the donkey (*Equus asinus*) in Mexico. In: Matthews NS, Taylor TS, eds. Veterinary Care of Donkeys. Ithaca, NY: International Veterinary Information Service. Available at: www.ivis.org.
- [18] Ahemed AE, Abdel-Hamid HY, Abdel-Rahim AA, Ismail MN. The influence of age and sex on some blood parameters in healthy donkey in south Valley Egypt. Bs Vet Med J; 2007:151-8.

- [19] Couroucè-Malblanc A, Fortier G, Moulin M, Valette JP, Petit L, Dumontier S, et al. Reference values on hematologic and biochemical parameters in French donkeys. In: Proceedings of the 10th International Congress of World Equine Veterinary Association (WEVA); Moscow, Russia; 2008. p. 458-60.
- [20] Lemma A, Moges M. Clinical, hematological and serum biochemical reference values of working donkeys (*Equus asinus*) owned by transport operators in Addis Ababa, Ethiopia. Livest Res Rural Dev 2009;21:1-8.
- [21] Orlandi M, Curadi MC, Leotta R, Berni P. Metabolic profle in the Amiatan donkey [in Italian]. Annali Facoltà Medicina Veterinaria di Pisa 1997;50:47-53.
- [22] Harvey JW, Asquith RL, McNulty PK, Kivipelto J, Bauer JE. Haematology of foals up to one year old. Equine Vet J 1984;16:347-53.
- [23] Bauer JE, Harvey JW, Asquith RL, McNulty K, Kivipelto J. Clinical chemistry reference values of foals during the first year of life. Equine Vet J 1984;16:361-3.
- [24] Waelchli RO, Lutz H, Hermann M, Ruegg C, Eggenberger E. Hematological reference values for foals in the first two months of life. Schweiz Arch Tierheilkd 1994;136:127-36.
- [25] Madigan JE. Hematology reference values, blood chemistry values and blood gases. Manual of Equine Neonatal Medicine. 2nd ed. Woodland, CA: Live Oak Publishing; 1991. p. 310-17.
- [26] Knottembelt DC, Holdstock N, Madigan JE. Appendix 2 (Reference values for hematology and biochemistry). Equine neonatology: Medicine and surgery. 1st ed. USA: Saunders Elsevier; 2004. 475-6.
- [27] McAuliffe SB, Slovis NM. Appendix 3 (Reference values for hematology) and Appendix 4 (Reference values for biochemistry). Color atlas of diseases and disorders of the foal. 1st ed. USA: Saunders Elsevier; 2008. 382-383.
- [28] Bernard WV, Reimer JM. Physical examination. In: Bernard WV, Barr BS, editors. Equine Pediatric Medicine. 1st ed. London, UK: Manson Publishing Ltd; 2012. p. 10-3.
- [29] Brommer H, Sloet van Oldruitenborgh-Oosterbaab MM, Kessels B. Haematology: Haematological and blood biochemical characteristics of Dutch wormblood foals managed under three different rearing conditions from birth to 5 months of age. Vet Q 2001;23: 92-5.
- [30] Pugh DG. Donkey reproduction. In: Proceedings of the Annual Convention of the American Association for Equine Practitioners 2002;48:113-14.
- [31] Adult Horse Vaccination Chart. Available at: http://www.aaep.org/ vaccination_guidelines.htm. Accessed April 10, 2010.
- [32] Vaala WE. Perinatology. Vet Clin North Am Eq Pract 1994;10:237-65.
- [33] Sgorbini M, Luchetti E, Ressel L, Poli A, Corazza M. Evaluation of two methodologies to determine immunoglobulins in serum of foals of Amiata donkey. In: Proceedings of the XIV SIVE/FEEVA Congress; Venice, Italy; 2008. p. 431-2.
- [34] Stoneham SJ. Assessing the newborn foal. In: Paradis MR, editor. Equine neonatal medicine: A case-based approach. 1st ed. Philadelphia, PA: Elsevier Inc.; 2006. p. 1-11.
- [35] Sgorbini M, Crisci A, Falconcini A, Rota A, Corazza M. Evaluation of semeiotic data, haematological and biochemical parameters in the amiata donkey newborn foals. Proceedings of the LXIII SISVET Congress; Udine, Italy; 2009;13:304-5.
- [36] Wilkins BH. Renal function in sick very low birthweight infants: 2. Urea and creatinine excretion. Arch Dis Child 1992;67:1146-53.