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Hematological and biochemical findings in pregnant, postfoaling, and lactating jennies

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ABSTRACT

The aims of this study were to (1) verify if significant changes occur in hematological and biochemical parameters in jennies during the last 2 months of pregnancy and the first 2 months of lactation, and (2) determine any differences with mares. Hematological and biochemical parameters were evaluated in jennies every 15 days during late pregnancy, parturition, and early lactation. The Kolmogorov–Smirnov test, analysis of variance for repeated measurements and Tukey's multiple comparison test as post hoc were applied. The significance level was set at $P < 0.05$. Statistical analysis showed differences related to time for Red Blood Cells (RBC) count and Hematocrit (HCT), White Blood Cells (WBC) count, platelet count (PLT), total proteins, blood urea, triglycerides and total cholesterol concentrations, aspartate aminotransferase, gamma-glutamyltransferase, creatine-phosphokinase activities, sodium (Na) and potassium (K). RBC and HCT were higher in late pregnancy than at foaling and during lactation. The relative anemia might be due to increased water ingestion because of fluid losses. The WBC count was higher at foaling than during late pregnancy and lactation. This could be related to the release of cortisol and catecholamine during delivery. The PLT trend showed lower values from delivery to the first 2 months of lactation compared to late gestation. Blood urea increased near parturition, and then remained constant during delivery and lactation, which might be due to the high energy demand at the beginning of lactation. Triglycerides and total cholesterol showed a decrease from delivery through the lactation period. Thus, jennies seem to have a similar metabolism of fats to ponies and draft horse mares, characterized by a greater fat content and mobilization than light breed horses. Aspartate aminotransferase activity decreased at parturition and early lactation, probably because of a predominance of anabolic over catabolic processes during pregnancy. Gamma-glutamyltransferase activity was lower at delivery and during lactation than at late gestation. This could be due to a physiological load on the liver in the perinatal period. Gamma-glutamyltransferase activity was always higher than in mares, but within the normal range for adult donkeys. Creatine-phosphokinase decreased near delivery, then was constant from parturition through the first 2 months of lactation. Na decreased during lactation, probably due to an increased renal retention mediated by aldosterone release during pregnancy. K showed the same trend as Na, and concentrations are in line with the species. The higher K during pregnancy may be due to reabsorption by the gut. Total proteins decreased more during the postpartum period and lactation than in the gestational period. In conclusion, our results showed significant changes in hematological and biochemical parameters in jennies during the last 2 months of pregnancy and the first 2 months of lactation and these changes are only partially comparable to mares.

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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. Donkey milk could possibly be used in children with intolerance to cow's milk [1,2] or in animal-assisted therapy [3]. The renewed interest in these animals is demonstrated by the number of studies on establishing the baseline data of both hematological and biochemical variables in the blood of adult [4–26] and in newborn donkeys [27,28].

Pregnancy and lactation are physiological periods that result in increased metabolic demands. Although homeostatic mechanisms keep substances in the blood at relatively constant levels, some changes in the concentrations of routine clinical chemistry analytes are likely to occur [29]. During pregnancy, an expansion in plasma volume and erythrocyte mass occurs as well as an increase in plasma protein synthesis [30]. In women, it is wel- known that during pregnancy reference intervals are different from the nonpregnant state [31–33].

The effects of pregnancy, parturition, and lactation on hematological parameters have only been studied in horses to a limited extent [34–44] in different breeds. Moreover, only one study [42] has reported a thorough investigation of the last months of pregnancy and first month of lactation, and no data are present in literature concerning hematological and biochemical values in jennies during the same period. Therefore, more detailed parameters in hematology and biochemistry during late pregnancy, parturition, and lactation in jennies could be useful for accurate diagnosis of diseases.

The aims of this study were to (1) verify if significant changes occur in hematological and biochemical parameters in jennies during the last 2 months of pregnancy and the first 2 months of lactation; and (2) determine any differences with respect to equine species.

2. Material and methods

2.1. Animals

The study was conducted on nine jennies belonging to the Amiata donkey breed for a total of 18 pregnancies and 16 foals born during a two-year study. Jennies were from 5 to 13 year old, weighed 300 to 350 kg, and were kept in collective paddocks at the Veterinary Teaching Hospital, Department of Veterinary Sciences, Pisa University. Approval for this study was obtained from the Ethical Committee on Animal Experimentation of the University of Pisa, and the protocol was sent to the Ministry of Health.

To provide National Research Council recommendations for energy, jennies were fed with meadow hay ad libitum along with commercial equine feed This feeding procedure began at 2 months prepartum and continued through postpartum and the first 2 months of lactation. Jennies were housed together during pregnancy in a paddock 10 × 20 m. Close to parturition, jennies were housed in 4 × 4 m boxes for the first 15 days of lactation and then returned to the paddock.

Jennies were included in this study according to the following criteria: (1) pregnancy length 353.4 ± 13.0 days [43]; (2) unassisted delivery; (3) treatment against gastrointestinal parasites and vaccinated against equine influenza, tetanus, and equine herpes virus-1, in accordance with the guidelines of the American Association of Equine Practitioners Infectious Disease Committee [44].

2.2. Sample collection and handling

Blood samples were obtained from the jugular vein. Each jenny was sampled every 15 days during late pregnancy, approximately 2 months before parturition, at parturition, and every 15 days during the first 2 months of lactation. Blood was collected in test tubes containing ethylenediaminetetraacetic acid (cod. 22056, FL Medical, Padua, Italy) and lithium-heparin test tubes (cod. 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), except for the sample collected at parturition.

2.3. Complete blood count

Ethylenediaminetetraacetic acid 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. The aim was 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) platelet count (PLT). Specimens containing clots or grossly hemolyzed were excluded. A quality control of the cell counter was performed each day.

2.4. Biochemical analysis

Heparinized samples were centrifuged at 3000 × *g* for 10 minutes as recommended by the manufacturer. Plasma was then frozen at −18 °C and analyzed within 15 days after collection. Clinical chemistry was performed with an autoanalyzer (Liasys; Analyzer Medical System-AMS, Rome, Italy; quality control normal level: ASR02010, and pathologic level: ASR02020, Assel Srl, Rome, Italy).

The parameters analyzed were (1) glucose concentration (Glucose SL, enzymatic colorimetric method, cod. ASR01202; Assel Srl, Rome, Italy); (2) creatinine (Creatinine, kinetic modified Jaffè method, cod. ASR01150; Assel Srl, Rome, Italy); (3) blood urea (Urea UV SL, kinetic enzymatic method, cod. ASR01143; Assel Srl, Rome, Italy); (4) triglycerides (Triglycerides-SL, enzymatic colorimetric method, cod. ASR01134; Assel Srl, Rome, Italy); (5) total cholesterol (Cholesterol liquid, trinder method-endpoint, cod. 7050; FAR, Verona, Italy); (6) total bilirubin (Total Bilirubin, colorimetric method without DMSO, cod. ASR01034/1; Assel Srl, Rome, Italy); (7) aspartate aminotransferase (AST), (AST SL, kinetic method UV IFCC- cod. ASR01220; Assel Srl, Rome, Italy); (8) gamma glutamyl transferase (GGT), (Gamma GT SL, kinetic method-Szasz-Tris,