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Comparative Study on Haematological and some Biochemical Parameters of Rabbit, *Oryctolagus cuniculus* and Partridge, *Francolinus francolinus* from Iraqi Environment

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Abstract

The present study aimed to establish reference normal ranges of haematological and biochemical parameters of two vertebrates (rabbits and partridges). Twenty animals (10 rabbits, 10 partridges) were used in this study. Blood samples, collected from heart of rabbits, as well from the right jugular vein of partridge, and delivered to the laboratory. Some biochemical parameters (potassium, sodium, and uric acid) have been investigated, in addition blood samples were tested for: white blood cells (WBC), red blood cells (RBC), haemoglobin (HB), haematocrit (HCT), mean corpuscular hemoglobin (MCH), mean concentration of hemoglobin in the corpuscular volume (MCHC), mean corpuscular volume (MCV).

Results indicate that haematological and biochemical data are revealed statistically significant differences between rabbit and partridge in most parameter except sodium and potassium were showed no significantly different. The RBC and MCHC levels were significantly higher (P<0.05) in rabbit than in partridge. While, the results showed higher significant (P<0.05) in HB, HCT, MCV, and MCH concentrations were observed in partridge than in rabbit. White blood cells (lymphocytes, monocytes, and granulocytes) showed significant increase in partridge than in rabbit. Some biochemical values are showed non-significant difference in potassium and sodium value between rabbits and partridge. While, the uric acid showed significantly increase (P<0.05) in partridge than in rabbit.

Keywords: Haematology, Serum biochemical parameters, Rabbit, Partridge.

دراسة مقارنة لمعايير الدم وبعض الفحوصات الكيموحيوية للارنب الداجن *Oryctolagus cuniculus* والدراج العراقي الاسود *Francolinus francolinu* في البيئة العراقية

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الخلاصة

تهدف هذه الدراسة إلى وضع مؤشرات لبعض معايير الدم لأثنين من الفقاريات (الأرانب والدراج). استخدم في هذه الدراسة 20 حيوان (10 ارانب 10 دراج). تم جمع عينات الدم من قلب الأرانب، وكذلك من حبل الوريد الأيمن للدراج وتم نقلها للمختبر. تم اجراء بعض القياسات البيوكيميائية (البوتاسيوم، الصوديوم، وحمض اليوريك). بالاضافة لاختبار عينات الدم لخلايا الدم البيضاء (WBC)، وخلايا الدم الحمراء (RBC)، قياس تركيز الهيموجلوبين (HB)، و حجم كمية من كريات الدم الحمراء (HCT)، ومتوسط وزن هيموجلوبين كرية الدم الحمراء (MCH)، معدل نسبة تركيز الهيموجلوبين في كريات الدم الحمراء (MCHC)، ومتوسط حجم

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الكرية الحمراء (MCV). وتشير النتائج إلى وجود فروقات معنوية واضحة ذات دلالة احصاية للمعايير الدموية والكيموحيوية بين الأرانب الدراج في معظم الاختبارات عدا الصوديوم والبوتاسيوم اللذان لم يظهرا اختلافا كبيرا. وكانت مستويات RBC وMCHC أعلى بكثير (P <0.05) في الأرانب مما كانت عليه في الدراج. في حين اظهرت النتاج ارتفاع معنوي (P <0.05) في HGB, HCT, MCV, MCH, and MCH للدراج مما كان عليه في الأرانب. وأظهرت WBC زيادة معنوية كبيرة في الدراج مما هي عليه في الأرانب. ولم تظهر النتاج اختلافات معنوية ذات قيمة للبوتاسيوم والصوديوم بين الدراج والارنب. في حين، اظهر حمض اليوريك زيادة معنوية كبيرة (P <0.05) في الدراج مما كانت عليه في الأرانب.

الكلمات المفتاحية: الاختبارات الدموية، الفحوصات البيوكيمياوية، الارانب، طائر الدراج العراقي الاسود

Introduction

Rabbit (*Oryctolagus cuniculaus)* and Iraqi black Partridge (*Francolinus francolinus*) are two common vertebrate in Iraqi environment. Review of literature revealed that there are several previous studies investigative the haematological and biochemical parameter [1, 2].

Animal model systems are used extensively for experimental *In vivo* studies, when biochemical and haematological measurements are made; reference intervals for the animal species must be determined [3]. Measurement of haematological parameters provides valuable information in this regards and routinely used in human's and animal's medicines, but unfortunately due to lack of information, blood profile have not been widely used in avian medicine [4].

Although there have been an increasing number of studies in recent years devoted to determining comparative values of haematology and blood chemistry for wild bird species[5], reference values are available for only a very low percentage of them. It is fundamental in interpreting the results obtained for a given individual to know the normal ranges of variation of blood parameters and their variability with respect to such factors as age, gender, and captivity [6].

Haematological and blood chemistry analyses are among the methods which contribute to the detection of some changes in health status and can be useful aids for diagnosis of diseases in birds [2]. Haematological studies have been reported in a number of animal species including Turkeys *(Meleagris gallopavo*) [2], *Columba livia; Streptopelia decaocto; Streptopelia roseogrisea* [7], *hornbills (Aceros; Ceratogymna; Anthracoceros)* [5], several flamingo species [8].

Rabbits are widely used in haematological and biomedical research. Blood biochemistry is used to monitor progression of disease, Plasma or serum biochemical analyses provide information about internal organs, electrolytes [sodium (Na), chloride (Cl), potassium (K), calcium (Ca), phosphorus (P)], proteins (globulins, albumin) and nutritional or metabolic parameters (cholesterol, triglycerides, glucose) [9]. Haematololical analyses, which include red blood cell (RBC) counts, white blood cell (WBC) counts and differential WBC counts, provide information about the haematopoietic system and immunological response. These blood tests can serve as diagnostic adjuncts in the development of a presumptive or denitive diagnosis [10] with the establishment of reference values for blood parameters for a variety of marine bird species [11].

Rabbits are often used as live models in scientific research where changes in blood count occur, since they handle multiple blood sampling well, their surface veins are pronounced, and they are still and suitable for manipulation in delicate procedures [12]. Review of Iraqi literatures revealed that almost lack of studies about blood parameters of various domestic animals. This situation supports us to perform this study with hope to give more information about the haematological and biochemical study of the animals under investigation.

Material and method

Animal experiment:

The experiments were performed on 20 sexually mature and clinically healthy animals (10) rabbit *Oryctolagus cuniculus* and (10) partridge *Francolinus francolinus* collected from local market at Baghdad city. The animals were housed in open-air, under natural temperature and lighting conditions. They were supplied with water *ad Libitum* and fed twice daily.

On each blood collection date, 3.0 ml of blood was obtained from heart of rabbits and from right jugular (neck) vein of partridge after cleaning the vein puncture site with an alcohol swab. Blood was collected using a 3-ml syringe. Blood was immediately transferred into plain tube for serum separator

and EDTA tubes. Blood smears were prepared at the time of blood collection, air dried and stored at room temperature until stained with giemsa stain. Sodium, Potassium, Uric acid methods were done according to to Tietz [13].

Blood tests: The haematological tests included: haemoglobin (HB) measurements; packed cell volume (PCV); red blood cell (RBC), white blood cell (WBC) and mean cell haemoglobin (MCH); mean cell volume (MCV) and mean cell haemoglobin concentration (MCHC). The counts were recorded using an electronic particle counter (Industrial D Model, Coulter Electronics Ltd., Luton). The whole blood cells number as well the count of red blood cells (RBC), white blood cells (WBC) was counted in a chamber within an hour after blood sampling, taking into account their relation on blood smears, stained with Giemsa.

Statistical analysis: Data are expressed as the mean ± SE. The statistical significance was carried out using one- way analysis of variance test followed by Duncan's Multiple Range Test (SPSS statistical software package) [14]. A possibility of *P* value ($p \leq 0.05$) was considered as significant differences between means.

Results

The mean haematological values obtained for the partridge and rabbit are presented in Table-1,-2 Specific variations between the classes were detected in all haematological values. The RBC and MCHC levels were significantly higher ($P<0.05$) in rabbit than in partridge. While, the significant higher ($P<0.05$) in HGB, HCT, MCV, MCH, and MCH concentrations were observed in partridge than in rabbit. WBC, lymphocyte, monocyte, and granulocyte showed significant increase in partridge than in rabbit. Some biochemical values are showed in Table-3, the results showed that there wrer non- significant in potassium and sodium values between two animals. While, the uric acid showed significantly increase ($P<0.05$) in partridge than in rabbit.

Table 1- Value of haematological parameters (RBC) in Partridge and Rabbit

Haematological parameters	Partridge		Rabbit	
	Mean± S.E	Range	Mean ± S.E	Range
RBC ($l0^6/mm^3$)	2.348 ± 0.205[a]	1.73–2.92	5.923± 0.156[b]	5.2-6.77
HGB (g/100 ml)	11.88 ± 0.918 [a]	8.8- 13.8	11.325± 0.355 [b]	10.1- 12.6
HCT (%)	37.3 ± 2.590 [a]	32.6%- 43.8%	34.737±0.662 [b]	32.8%- 37.2%
MCV (pg)	143.42 ± 3.023 [a]	137.7 – 148.7	59.212± 1.1609 [b]	54.4-65
MCH (%)	51.88 ± 0.355 [a]	50.1%– 4.9%	19.25±0.402 [b]	18.5%-19.25%
MCHC (μm^3)	32.58 ± 0.967 [a]	29.9 – 35.3	32.612±0.691[b]	29.9-34.7

Values with different letters are significant ±SE: Standard error

Table 2- The Leucocyte values (Mean ± SD) in Partridge and Rabbit

Haematological parameters	Partridge		Rabbit	
	Mean± S.E	Range	Mean± S.E	Range
WBC ($10^3/mm^3$)	97.88 ±1.107[a]	95.4-102	3.812±0.395 [b]	2.8-5.5
LYM ($10^3/mm^3$)	65.96 ±3.611 [a]	58.7-79.4	1.1875±0.0692 [b]	0.9- 1.4
MON ($10^3/mm^3$)	22.04 ±0.242 [a]	21.2 -22.5	0.362± 0.059 [b]	0.3-0.5
GRA ($10^3/mm^3$)	12.12 ± 2.038 [a]	7.5– 18.8	2.25±0.370 [b]	1.3- 3.6

Values with different letters are significant ±SE: Standard error

Table 3- Some biochemical parameters recorded from the partridge and rabbit

Test	Partridge		Rabbit	
	Mean± S.E	Range	Mean± S.E	Range
Potassium (mmol/l)	4.002 ± 0.551	6.878 - 3.705	40.63 ± 0.210	2.868-4.949
Sodium (mmol/l)	129.195 ± 0.975	124.024- 42.709	136.502 ± 3.470	124.024-148.379
Uric acid (mg/dl)	6.023 ± 0.582[a]	1.546 – 12.623	8.625 ± 0.357[b]	6.629 - 9.704

Values with different letters are significant ±SE: Standard error

Discussion:

The primary purpose of this study was to determine whether significant haematological differences were present in healthy rabbit in comparison with partridge. The lack of published reference intervals for haematological and biochemical constituents in the animals under investigation lead to reflects the preference of most investigators to establish their own reference intervals, Reference intervals may be set at the outset of some difficulties to discuss the results of the present study, making test animals their own controls. Alternatively, test and control groups can be monitored together; the animals would be housed under identical conditions and assays performed on test and control animals with the same analytical conditions, techniques, and instruments. However, the recent development of inter laboratory standardization now makes it possible to obtain comparable data between laboratories, thus avoiding the needless expense of determining in-house reference intervals [3].

Results of the present study revealed that the haematological parameters of partridge RBC, Hb, HCT, MCV, MCH, MCHC of rabbit were significantly different from the partridge, these results agree with smith and Hattingh [15] who found the avian blood differs in cells' characteristics from their mammalian counterpart. Several factors including physiological and environmental conditions, diet contents, water and feed restriction, fasting, age, affect the blood profiles of healthy birds. Although there is limited information concerning the normal blood profiles of different strains of avian at different ages.

In the present study, the sampled rabbits and partridge were randomly chosen. However, some of them were possibly suffering from distress and or sub-clinical, chronically infections. The width of the range between minimum and maximum values for several parameters could be due to inter-subject variability occurring in non-healthy animals. On the other hand, the standard error was generally narrow indicating that these differences in haematological and biochemical values could be caused by nutritional, environmental and hormonal factors.

Therefore, a specific analysis and comparison between different units has to be planned and performed. Further investigations are needed to better understand the usefulness of clinical chemistry and haematology for assessing the health of rabbits [16].

To the best of knowledge, there have been no studies of the haematology data in individuals of the partridge. Several small but significant differences were documented when haematologic values from rabbit and partridge were compared. Mean HGB, HCT, MCV, MCH and MCHC values were slightly lower in rabbit compared with partridge. The causes of these subtle differences is unknown, but differences in iron metabolism should be considered [17].

In general haematological parameters are affected by diurnal fluctuations or changes in daily physical and metabolic activities [18]. The mean haematological values RBC, HB of birds vary among species. Other factors, which affect the RBC counts, include breed, sex and the nutrition supplied to the bird, HCT, Hb concentration and RBC count had been reported to increase with age in chickens [19]. The red cell mass of birds is influenced by sex, environmental factors, age, hormonal influences and disease. In general, the total erythrocyte count increase with age and are higher in male than female birds [10].

The values for the RBC counts, haemoglobin concentrations, and haematocrit values were in accordance with those previously reported by Schmidt *et al*., [20] for pheasants, and by Pica *et al*., [21] for partridges, but all of these parameters were, however, lower than those observed by and Gibson and Lloyd [22] and Hauptmanova *et al*., [23] for pheasants and by Rico *et al*., [24] for adult partridges. In addition, although these parameters of the pheasants were within the range of most avian species such as the hen [25] chicken, turkey, quail, and ostrich [26], the same values were lower than those determined for the pigeon guillemot (*Cepphus columba*) [27]. The red blood cell counts according to Swenson [28] are influenced among other factors by nutrition, physical activities and volume and its reduction indicates anaemia.

Results of the present study showed that there were significantly different in WBC, LYM, MON, GRA values between rabbit and partridge, the leucocyte value in partridge is higher than the rabbit, In Ologhobo *et al*. [29] observed that an increase in white blood cells count above normal is an indication of the presence of exogenous substances and foreign bodies in the body. In this study, there was no case of such abnormal rise in values of white blood cells.

Lymphocytes are important in forming barriers against local disease conditions and may be involved in antibody formation [30]. In the same way, thrombocytes are important in the coagulation

process. Many authors have reported a large variation mainly in leukocytes and thrombocytes parameters due to age and nutritional condition of animals [31]. Aletor and Egberongbe [32] reported that red blood cell counts and packed cell volume (PCV) are mostly affected by dietary treatment. Under normal conditions the composition of blood is reasonably constant for any particular species with changes falling with fairly narrow limits [33].

Uric acid and urea are the main products of the nitrogen metabolism of birds, and uric acid is also the major nitrogenous waste product of birds, the level of uric acid in the blood of pheasant hens by species and diet [34]. The partridge had uric acid levels that were similar to those of other groups of birds in captivity, such as hornbills, pigeons, ducks, or cranes, but lower than those of raptors [35]. As also is the case with other groups of birds [34], the uric acid concentration was greater than the urea concentration of partridge.

In an analysis of the influence of diet on different biochemical parameters in birds [35], it was found that when the diet was rich in animal protein, the uric acid and urea concentrations were greater than when the diet was fruit or grains [5]. Previous studies have found similar relationships between metabolites related to fat and protein metabolism and haematocrit with the nutritional status or body condition of captive and free-living birds [36].

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