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THE LABILITY OF SOME HAEMATOLOGICAL PARAMETERS IN CHICKENS AND DUCKS

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OPSOMMING: VARIEERBAARHEID IN SEKERE HEMATOLOGIESE PARAMETERS VAN HOENDERS EN EENDE

In hierdie studie is die invloed van beperking van lewensruimte (aanhouding in hokke) op die hematologie van hoenders en eende (soos vergelyk met dié van vry-lopende voëls) ondersoek. Statisties beduidende verskille is waargeneem in verskeie hematologiese parameters en die studie het bewys dat gevolgtrekkings wat gemaak kan word betreffende inter- en intraspesie-verskille afhang van hoe die voëls aangehou was en in hoe 'n mate spanning hulle beïnvloed. Dit word voorgestel dat projekbeplanning vir studies wat die hematologie van voëls ondersoek, hierdie faktore in aanmerking moet neem.

SUMMARY:

The effects of restricting chickens and ducks to cages on their haematology (as compared to that of free-roaming animals) were investigated in this study. Significant differences were observed in several haematological parameters and the study showed that conclusions that could be drawn concerning inter- and intraspecies differences depended on how the birds were housed and how susceptible they were to stress. It is suggested that experimental design concerning bird haematology should take these factors into account.

Introduction

A review of the literature shows that a substantial body of results has accumulated concerning the haematology of birds (Sturkie, 1976). Aspects which have recevied attention vary from physical properties of blood to processes involved in coagulation. Species differences are evident in the haemoglobin concentrations, packed cell volumes, erythrocyte counts, etc., in addition to male/female differences (Sturkie, 1976). Over and above this, it is known that age, hormones, hypoxia, capture stress, and other factors may influence haematological parameters in some species (Bond & Gilbert, 1958; Henschel & Louw, 1978). Factors which have not yet been investigated in depth are the effects of laboratory acclimatization on the haematology of birds. In this regard the restriction of movement, acclimatization times, changes of feeding patterns and other factors may be important, possibly resulting in altered haematological results being obtained which may explain some of the differences in reported values for the same species of bird (Sturkie, 1976). An associated factor is the effects of handling on the results obtained seeing that birds are known to easily become excited by these procedures. As an initial part of a broad study to establish baseline values for certain haematological parameters of local birds we investigated the effects of restricting chickens and ducks to cages on their haematology. The animals were subsequently allowed to roam freely and then re-investigated.

Procedure

Animals

Ducks, *Anas platyrhynchos domesticus* (White Peking), were obtained from a local breeder at the age of six days. Chickens, *Gallus domesticus* (White Leghorn), were obtained from a different breeder at the age of one day. Initially, both groups of birds were kept in a closed room but were subsequently put into wire cages with concrete floors (containing shelters) at the age of three months. The cages measured 3 x 4 x 3 metres. Floorspace was thus 12 m^2 and eight to 15 animals were housed per cage (about 1.5 m^2/chicken and 0.8 m^2/duck). Water was supplied and all animals were fed on mixed chicken feed and Broiler Finisher Pellets (Epol). In addition, "greens" were supplied at least once a week. The animals were frequently handled so as to reduce handling stress as far as possible and they became quite tame. At the age of seven months the females of both species began to lay eggs and experiments were begun at the age of nine months. The animals used for haematological investigations were thus mature birds of both sexes.

After the haematological investigation of caged birds, the animals were transported to a farm in the same geographical region and allowed to roam freely for two months during which time they were also handled. The same food as previously given was available, but the

animals were now also free to scrub. After two months the birds were transported back to the laboratory and haematological investigations were done within two hours of blood sampling. By designing the experiment in the above fashion, haematological parameters of the same birds could be compared before and after they were allowed to roam freely. The investigations were done during the same season (summer), the age of the birds was known and they were accustomed to handling. The results obtained would thus show whether the procedures used here influenced haematological parameters in these animals.

Haematological investigations

Blood was obtained from fasted animals (overnight) from the basilic vein and heparin (4 mg/ml) used as anticoagulant (Hattingh & Smith, 1976). Blood pH, pO_2 and pCO_2 were measured with a Radiometer BMS3, the packed cell volume with a Christ centrifuge, plasma chloride concentration with a Buchler-Cotlove Chloridometer (Model 4-2008), plasma protein concentration according to the method of Lowry, Rosebrough, Far and Randall (1951) using bovine serum albumin as standard, total plasma lipids using a Merck test kit and plasma sodium and potassium concentrations using a Gallenkamp flame photometer. Blood glucose concentration was measured using a Biochemica test kit (GOD-Period method) and osmottic fragility of red cells according to the method of Ezell, Sulya and Dogden (1969) in buffered saline solutions. Haemoglobin concentrations, red blood cells counts and red blood cell dimensions were determined as described previously (Hattingh, 1972). All data were subjected to the Students' t-test and values of P equal to or less than 0.01 were considered significant. Results are reported as means ± S.D.

Results

Chickens

The results obtained with chickens are shown in Table 1. It is clear that significant differences existed between eight haematological parameters of males and females of the caged birds and between three parameters in the case of free-roaming birds. When the results of the two groups of chickens are compared, significant differences are found between the pO_2, Na^+ and K^+ values of male blood and between the erythrocyte numbers, plasma protein concentratins and Cl^- values of female blood.

Table 1

Haematological parameters of caged and free-roaming chickens

Parameter	Caged Birds		Free-roaming Birds	
	Males	Females	Males	Females
pH	7.42 ± 0.04	*7.35 ± 0.03	7.42 ± 0.05	7.38 ± 0.04
pO_2 (mmHg)	49.3 ± 7.5	43.4 ± 10.8	66.4 ± 9.7	52.7 ± 10.6
pCO_2 (mmHg)	36.8 ± 5.7	41.1 ± 6.0	36.2 ± 6.0	42.1 ± 6.1
Packed cell volume (%)	49.6 ± 3.0	*40.4 ± 4.6	47.9 ± 3.5	*35.7 ± 3.0
Haemoglobin (g%)	16.01 ± 1.15	*11.89 ± 1.41	14.04 ± 1.05	*10.34 ± 1.27
Erythrocyte count (x 10^6 cells/mm^3)	2.70 ± 0.32	*2.15 ± 0.21	2.62 ± 0.24	2.33 ± 0.26
Blood glucose (mg%)	122.5 ± 14.0	127.4 ± 11.8	112.3 ± 0.1	137.0 ± 19.8
Plasma protein (mg/ml)	46.5 ± 5.7	*62.3 ± 10.4	49.8 ± 5.1	48.2 ± 5.1
Plasma lipids (mg%)	320.0 ± 56.0	*1 911.0 ± 955.0	345.0 ± 83.0	*1 280.0 ± 237.0
Na^+ (mEg/l)	174.0 ± 4.0	168.0 ± 7.0	160.0 ± 6.0	155.0 ± 6.0
K^+ (mEg/l)	3.9 ± 0.7	3.3 ± 0.5	3.0 ± 0.4	3.4 ± 0.7
Cl^- (mEq/l)	125.0 ± 2.0	*118.0 ± 5.0	118.0 ± 6.0	129.0 ± 3.0
Mean cell fragility (% Na Cl)	0.36 ± 0.02	*0.32 ± 0.02	0.36 ± 0.02	0.33 ± 0.01
Erythrocyte length (μ)	10.6 ± 0.3	10.8 ± 0.4	11.2 ± 0.4	11.7 ± 0.6
breadth (μ)	6.7 ± 0.1	6.9 ± 0.1	7.3 ± 0.3	7.2 ± 0.1
N	8	8	8	8

Means ± S.D.

* Indicates a significant difference between the values of males and females within a group.

Table 2

Haematological parameters of caged and free-roaming ducks

Parameter	Caged Birds		Free-roaming Birds	
	Males	Females	Males	Females
pH	7.43 ± 0.08	7.44 ± 0.08	7.44 ± 0.04	7.41 ± 0.02
pO_2 (mmHg)	51.8 ± 9.2	60.4 ± 6.5	58.4 ± 6.8	56.6 ± 5.1
pCO_2 (mmHg)	23.5 ± 4.5	26.9 ± 2.5	26.0 ± 1.5	*31.1 ± 3.0
Packed cell volume (%)	44.9 ± 3.4	40.4 ± 2.4	43.0 ± 3.2	40.7 ± 3.5
Haemoglobin (g%)	14.08 ± 1.48	13.15 ± 0.99	13.23 ± 1.24	12.82 ± 1.40
Erythrocyte count (x 10^6 cells/mm^3)	2.64 ± 0.24	2.05 ± 0.26	2.54 ± 0.19	2.39 ± 0.29
Blood glucose (mg%)	149.2 ± 34.1	110.8 ± 17.9	96.0 ± 11.6	95.5 ± 3.7
Plasma protein (mg/ml)	41.6 ± 4.3	*61.8 ± 9.1	44.4 ± 4.8	54.1 ± 7.3
Plasma lipids (mg%)	722.0 ± 118.0	1 204.0 ± 427.0	923.0 ± 158.0	1 167.0 ± 473.0
Na^+ (mEg/l)	168.0 ± 4.0	163.0 ± 2.0	151.0 ± 7.0	155.0 ± 7.0
K^+ (mEg/l)	3.9 ± 0.3	1.7 ± 0.3	1.8 ± 0.3	2.1 ± 0.1
Cl^- (mEq/l)	119.0 ± 3.0	114.0 ± 3.0	111.0 ± 4.0	113.0 ± 2.0
Mean cell fragility (% Na Cl)	0.35 ± 0.01	0.34 ± 0.01	0.34 ± 0.01	0.36 ± 0.02
Erythrocyte length (μ)	12.3 ± 0.06	11.5 ± 0.6	12.1 ± 0.7	11.7 ± 0.4
breadth (μ)	6.8 ± 0.4	6.4 ± 0.2	7.3 ± 0.5	7.0 ± 0.2
N	7	7	7	7

Means ± S.D.

* Indicates a significant difference between the values of males and females within a group.

Ducks

In the case of duck blood few significant differences were found between male and female blood of the two groups (Table 2). In the case of caged birds, plasma protein values differed and in the case of free-roaming birds, pCO_2 values. When the two groups were compared, however, significant differences existed between blood glucose concentrations, Na^+ and K^+ values for male blood and between the pCO_2 values of female blood.

Species differences

By comparing the results obtained for males and females between species it was found that significant differences existed for caged birds in the case of total lipids, K^+ and Cl^- values (males) and K^+ values (females). In the case of free-roaming birds significant differences existed between total lipids and K^+ values in males and between haemoglobin concentrations, K^+, Cl^- and mean cell fragility values in females. It is therefore clear that in the above comparisons the conclusions that can be drawn differ depending on how the birds were housed and probably also on how ameniable they were to the experimental procedures of handling and transportation.

Discussion

In the literature haematological values for, amongst other birds, chickens and, to a lesser extent for ducks, have been summarized by Sturkie (1976), Freeman (1971), Jones and Johansen (1972) and others. In these articles, the range of particular parameters is very rarely stated and the usual trend has been to only list values without stating standard errors or deviations. In addition, no information concerning housing, feeding, handling, etc., is given but sexual and age differences are usually stressed. It is therefore difficult to compare the present results with those found in the literature in a meaningful way. However, certain observations need to be discussed, bearing in mind that the "normal" variation possibly occurring in the blood of the present study material is not known. (Due to the fact that caged and free-roaming animals had to be investigated in the same season and also that repeated blood withdrawals from animals with small blood volumes would have extended the observation period, this could not be done in the framework of the present protocol.) Packed cell volumes of hens used in the present study are all higher than those found in the literature (Freeman, 1971). In the case of caged birds these values are on average 23 per cent higher in males than in females. Jones and Johansen

(1972) indicate a figure of 16 per cent. Erythrocyte counts of chicken blood determined here are all lower than figures given in the literature (both males and females). The same applied to duck blood. Mean cell fragility values estimated in the present study are also lower than published values (Sturkie, 1976) whereas haemoglobin values for cocks and hens are all higher (Jones & Johansen, 1972). Blood pH of venous chicken blood is at the high range of values reported in the literature (Piiper & Scheid, 1973) and the same applies to plasma protein values (Frei & Perk, 1964). Plasma Cl^- values reported here are all above literature values and total lipid values usually below (Sturkie, 1976). If one now considers Table 1, it is clear that the parameters mentioned above all differ significantly (between males and females) in the blood of caged birds. In the literature the same trends are indicated although no statistics are presented. In the case of free-roaming chickens, however, these differences largely disappear remaining only in packed cell volumes, haemoglobin and total lipid values.

Male/females differences in duck blood on the other hand are not very pronounced and do not seem to be greatly influenced by the experimental procedure used here and due to the scanty nature of reported data few comparisons can be drawn. The above, taken together with the statistically significant differences observed within sexes in caged and free-roaming animals, indicate that from a comparative haematological point of view the male/female differences and species differences found here are nonsensical. The same applies to absolute values. These cannot be compared unless exact environmental and other conditions are stated. The above thus questions the validity of "normal or standard" values. In higher animals, including man, the variation in haematological values is well recognized and appreciated (Schalm, Jain & Carroll, 1975) but it would seem that lower animals show a greater variation or are less adaptable to these changes (Van Vuren & Hattingh, 1978). Exactly how susceptable birds are is not well documented and would merit further research. A recent study by Henschel and Louw (1978) has indeed indicated that certain free-flying bird species when caught exhibit a condition very similar to capture stress as experienced by wild ungulates. It is possible that the animals used in the present study were influenced adversely by transportation back to the laboratory after their two month free-roaming period. However, none of them were at any stage lethargic or unable to move as found by Henschel and Louw (1978) and the effects of transportation if present, were probably not very pronounced. The above are, however, important concepts to bear in mind when comparing haematological results of these animals.

Finally, the ducks used in this study were not influenced to the same extent as the chickens (considering male/female differences and differences within the sexes before and after roaming freely). One possible explanation for this is that chickens, especially hens, become excited very easily (Kawashiro & Scheid, 1975). Although the animals were tame and well accustomed to handling and blood sampling procedures, the chickens could be seen to react more adversely. In fish handling stress is known to influence haematological parameters and to cause variability in results (Van Vuren & Hattingh, 1978) and the same may apply here, although probably to a much lesser extent. Also, parasitic infestations, which are known to occur in birds and which may influence the haematology (Arnall & Keymer, 1975), could have been a contributing factor. The Epol pellets on which the birds were fed, contained Amprol (a coccidiostat) but it is possible that other infections may have been present which may have influenced one species more than the other. The above factors must be considered when studying bird blood and experimental design should as far as is possible take these into account.

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