ACTA SCIENTIARUM POLONORUM

Acta Sci. Pol. Zootechnica 22(2) 2023, 31–36

www.asp.zut.edu.pl pISSN 1644-0714 eISSN 2300-6145 DOI: 10.21005/asp.2023.22.2.03

ORIGINAL PAPER

Received: 24.11.2023
Accepted: 14.12.2023

EFFECTIVENESS OF BLOOD ELECTROLYTE HOMEOSTASIS REGULATION IN POLAR FOXES (*ALLOPEX LAGOPUS*, LINNAEUS 1758)

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ABSTRACT

The Arctic fox (*Alopex lagopus*, L. 1758) is maintained in captivity for breeding purposes due to the high quality of its winter fur. The success of breeding these animals depends primarily on their health, which determines high reproductive rates, low neonatal mortality and proper development of young animals. A prerequisite for maintaining good health is the maintenance of a constant internal environment, especially water-electrolyte and acid-base homeostasis. Sodium, potassium and chloride are the main electrolytes affecting volemia and osmolality of body fluids. In the current study, the results regarding the renal capacity of six-month-old foxes to regulate sodium, potassium, and chloride levels are presented, considering the sex of these animals. It has been shown that the concentration of sodium, potassium, and chloride in the blood serum of six-month-old polar foxes, as well as the osmotic pressure of the blood serum, were within the upper limits of physiological normal range. Higher concentrations of sodium and potassium were observed in males of these animals. Nonetheless, the results indicated efficient regulation of blood electrolyte homeostasis, both in females and males.

Key words: polar fox, plasma blood, natrium, kalium, chloride, osmotic pressure

INTRODUCTION

The Arctic fox, a member of the canine family, inhabits the polar tundra zone of the Northern Hemisphere's mainland, including Europe, Asia and North America. It is maintained in captivity for breeding purposes due to the high quality of its winter fur. The success of breeding these animals depends primarily on their health, which determines high reproductive rates, low neonatal mortality and proper development of young animals. A prerequisite for maintaining good health is the maintenance of a constant internal environment, especially water-electrolyte and acid-base homeostasis [Piotrowska et al. 2008, Szymeczko et al. 2009, Szymeczko et al. 2013, Seifter 2019, Skrzypczak 2021, Skrzypczak et al. 2021, Skrzypczak et al. 2022, Alamilla-Sanchez et al. 2023].

Sodium, potassium and chloride are the main electrolytes affecting volemia and osmolality of body fluids. By playing a decisive role in maintaining a constant volume and osmotic pressure of fluids, they influence the distribution of water in individual spaces and the acid-base balance of the body. The regulation of sodium, potassium, and chloride levels depends on renal function. The kidneys regulate chloride urinary excretion through the processes of glomerular filtration, tubular resorption and secretion [Skrzypczak and Drzeżdżon 2001, Kaneko et al. 2008, Michałek et al. 2010, Dratwa-Chałupnik et al. 2011, Laszczyńska et al. 2012, Thrall et al. 2012, Mokhber Dezfouli et al. 2012, Seifter and Chang 2017, Anderson 2020, Yasmin and Samad 2020].

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In view of the lack of studies in the world literature concerning the renal function in Arctic foxes, a comprehensive study of kidney function in terms of the regulation of water-electrolyte balance was conducted (unpublished data). In the current study, the results regarding the renal capacity of six-month-old foxes to regulate sodium, potassium, and chloride levels are presented, considering the sex of these animals.

MATERIAL AND METHODS

The research was conducted on 20 six-month-old polar foxes (10 females and 10 males) during the period Nov. 3–17. The animals were bred under the same environmental conditions (on one farm). They were evenly matched in terms of age and weight. The research was conducted in a biological research laboratory equipped with an automatic system for maintaining constant environmental conditions. The animals were housed in individual metabolic cages adapted for the quantitative collection of urine and feces. The foxes were fed a standard farm diet that completely met their nutritional requirements during the breeding period [NRC 2019]. They were fed once daily at noon and had unrestricted access to water.

The experiment was preceded by a three-day adaptation period of the animals. The tests were conducted over three days, separated by a one-day break. They were always initiated at the same time of day (10:00 a.m.).

Blood for testing was collected from the arm vein, anticoagulated with heparin (Heparin, Biochemie, Austria), immediately centrifuged (3000 rpm for 10 minutes), and the obtained plasma was frozen at –25°C until analysis.

Plasma and urine concentrations of sodium and potassium were analyzed using atomic emission spectrometry (AAnalyst 400 – PerkinElmer, λNa = 589 nm and λK = 766.5 nm); chloride concentrations were determined using the conductometric method (Chlorimeter 50 cl Trydent Med. s.c.); osmotic pressure was measured using the cryoscopic method with an automatic semi-micro osmometer (KNAUER).

Means and standard deviations were calculated. To assess the significance of the differences in the measured indicators at a level of significance of $P \leq 0.05$, the results underwent statistical calculations using a one-way ANOVA with repeated measures and Duncan's range test (Statistica 6.0).

The experimental protocol was approved by the Local Commission of Ethics for the Care and Use of Laboratory Animals (Resolution No. 2/21012010).

RESULTS

The results are presented in Tables 1–4. Plasma sodium concentrations in the tested foxes were high, ranging from 150.13 to 155.28 mmol · l^{-1}, with a mean value of 152.84 mmol · l^{-1}. The mean blood sodium concentration in females was lower (151.64 mmol · l^{-1}) than in males (154.04 mmol · l^{-1}). The difference in the values of this indicator between the sexes was statistically significant ($P \leq 0.05$) (Table 1).

Table 1. Plasma sodium concentrations (P_{Na}) in female and male polar foxes, mmol · l^{-1}

Days of testing		P_{Na}, mmol · l^{-1}		
		♀♂ n = 20	♀ n = 10	♂ n = 10
1	$\bar{x}$	151.45	150.59	152.31
	SD	1.39	3.04	3.15
2	$\bar{x}$	152.33	150.13	154.53
	SD	1.56	2.77	1.88
3	$\bar{x}$	154.74	154.20	155.28
	SD	2.29	4.66	3.95
Three-day average	$\bar{x}$	152.84	151.64*	154.04*
	SD	2.23	3.93	3.27

Rows 1–3 show the mean values for successive days of the study; mean values marked with * are significantly different at $P \leq 0.05$; ♀ female; ♂ male.

The mean plasma potassium concentration in polar foxes was 4.69 mmol · l^{-1} (4.58–4.95 mmol · l^{-1}). Plasma concentration of this electrolyte in males was higher (4.76 mmol · l^{-1}) than in females (4.62 mmol · l^{-1}). These differences were statistically significant ($P \leq 0.05$) (Table 2). In the female group, less individual variation was observed in the concentration of this electrolyte.

Table 2. Plasma potassium concentrations (P_K) in female and male polar foxes, mmol · l^{-1}

Days of testing		P_K, mmol · l^{-1}		
		♀♂ n = 20	♀ n = 10	♂ n = 10
1	$\bar{x}$	4.72	4.59	4.85
	SD	0.24	0.28	0.33
2	$\bar{x}$	4.69	4.58	4.80
	SD	0.14	0.27	0.25
3	$\bar{x}$	4.66	4.68	4.64
	SD	0.17	0.23	0.22
Three-day average	$\bar{x}$	4.69	4.62*	4.76*
	SD	0.18	0.26	0.27

Rows 1–3 show the mean values for successive days of the study; mean values marked with * are significantly different at $P \leq 0.05$; ♀ female; ♂ male.

The mean chloride concentration was 108.2 mmol · l^{-1} and ranged from 106.73 to 110.35 mmol · l^{-1}. No sta-

tistically significant differences were observed between the mean plasma chloride concentrations of female polar foxes 108.52 mmol · l^{-1} (106.73–110.35) and males 107.86 mmol · l^{-1} (107.53–108.17) (Table 3).

Table 3. Plasma chloride concentrations (P_{Cl}) in female and male polar foxes, mmol · l^{-1}

Days of testing		P_{Cl}, mmol · l^{-1}		
		♀♂ n = 20	♀ n = 10	♂ n = 10
1	$\bar{x}$	107.30	106.73	107.87
	SD	1.56	1.88	2.49
2	$\bar{x}$	108.32	108.47	108.17
	SD	1.99	2.80	1.96
3	$\bar{x}$	108.94	110.35	107.53
	SD	1.79	2.20	3.12
Three-day average	$\bar{x}$	108.19	108.52	107.86
	SD	1.86	2.70	2.49

Rows 1–3 show the mean values for successive days of the study; mean values marked with * are significantly different at $P \leq 0.05$; ♀ female; ♂ male.

Plasma osmotic pressure in male and female polar foxes was similar and showed no statistically significant differences. The mean value of plasma osmotic pressure in females was 304.03 ±2.55 mmol · l^{-1}, and in males it was 304.18 ±3.45 mmol · l^{-1} (Table 4).

Table 4. Blood osmotic pressure ($P_{osm.}$) in female and male polar foxes, mmol · l^{-1}

Days of testing		$P_{osm.}$, mmol · l^{-1}		
		♀♂ n = 20	♀ n = 10	♂ n = 10
1	$\bar{x}$	304.90	303.10	306.70
	SD	2.44	2.38	3.38
2	$\bar{x}$	302.88	304.05	301.70
	SD	1.80	2.87	1.60
3	$\bar{x}$	304.55	304.95	304.15
	SD	1.66	2.28	3.22
Three-day average	$\bar{x}$	304.11	304.03	304.18
	SD	2.13	2.55	3.45

Rows 1–3 show the mean values for successive days of the study; mean values marked with * are significantly different at $P \leq 0.05$; ♀ female; ♂ male.

DISCUSSION

The kidneys play a crucial role in maintaining water and electrolyte balance in the body, participating not only in the elimination of unnecessary metabolic products but also in the regulation of isovolemia, isoosmia, isoionia, and isohydria. Sodium is the most important cation in the extracellular fluid, determining the maintenance of osmotic pressure and constant volume of this water space. Sodium balance depends not only on renal function but also on the activity of humoral regulators, especially the RAA system, as well as vasopressin, ANP, PGE prostaglandin and PGI2 prostacyclin [Kaneko et al. 2008, Michałek et al. 2010, Thrall et al. 2012, Anderson 2020, Skrzypczak et al. 2021].

Potassium is the main intracellular cation. Only 1–2% of the total amount of potassium found in the body is present in body fluids, while 98–99% is located in the intracellular space, with the majority in skeletal muscle cells. Therefore, the concentration of potassium in the extracellular fluid depends not only on intake, absorption in the gastrointestinal tract, renal blood flow, filtration in the glomeruli, resorption and secretion processes in the renal tubules, but also on the translocation between the extracellular and intracellular spaces. Internal balance is rapidly regulated by insulin and catecholamines through the stimulation of Na+/K+-ATPase. Concentration of this electrolyte in the blood of most animals falls within the range of 3.5–5.5 mmol · l^{-1} [Dratwa-Chałupnik et al. 2011, Scalf 2014, Seifter and Chang 2017, Anderson 2020].

The chloride ion is the main anion in the extracellular fluid. Chlorides constitute approximately two-thirds of the anions in the extracellular fluid, and together with sodium and potassium, they regulate the water balance, osmotic pressure, and acid-base balance of bodily fluids. Chlorine can pass from plasma to blood cells and vice versa while transporting carbon dioxide through the blood, making it easily exchangeable. The concentration of chloride ions is closely linked to the level of sodium ions. The decrease or increase in their concentration is proportional in mmol · l^{-1}. It is accepted that in most animals, the concentration of chlorides falls within the range of 90 to 120 mmol · l^{-1} [Piotrowska et al. 2008, Korhonen and Huuki 2014,Seifter and Chang 2017].

The experiment showed high plasma sodium concentrations in young polar foxes, both in females and males. Statistically significantly higher concentrations of this electrolyte were confirmed in males. The unpublished results of our research suggest that the statistically significantly higher plasma sodium concentration found in males was probably due to reduced renal clearance of this electrolyte. Other authors have also reported high sodium values in the blood of foxes. Piotrowska et al. [2008] observed sodium concentrations at the level of 158.00 mmol · l^{-1} in one-year-old female Arctic foxes. The authors demonstrated that a high concentration of this electrolyte in the blood also persisted in two-, three-year-old, and older vixens.

Benn et al. [1986], in a study involving 13 non-pregnant, non-feeding females and 17 male foxes (red and silver foxes), clinically healthy, aged 1 to 3 years, observed sodium concentrations of 156 mmol · l^{-1} (146–165), potassium of 4.8 mmol · l^{-1} (3.9–5.8), and chloride of 111 mmol · l^{-1} (106–122). They have provided these values as reference ranges for foxes. The latter authors also reported that blood potassium concentrations depended on sex, and were higher in females (5.2 mmol · l^{-1}) than in males (4.6 mmol · l^{-1}).

Korhonen and Huuki [2014] found that the serum sodium concentration in blue foxes was 145.7 mmol · l^{-1}, potassium was 4.6 mmol · l^{-1}, and chlorides amounted to 108.2 mmol · l^{-1}. Anderson [2020] determined that plasma sodium levels in dogs should be between 140–150 mmol · l^{-1}, while in cats, they should range from 146 to 157 mmol · l^{-1}.

In herbivorous animals, the concentration of electrolytes in the blood serum is lower, for example, in healthy adult cattle, sodium concentration ranges from 137 to 145 mmol · l^{-1}, potassium from 4.0 to 4.8 mmol · l^{-1}, and chloride from 99 to 106 mmol · l^{-1} (Mokhber Dezfouli et al. 2012). Herosimczyk et al. [2011] demonstrated that herbivorous animals already possess the ability to maintain water-electrolyte homeostasis during the neonatal period.

The plasma potassium concentration determined in the present experiment was high and statistically higher in males. Piotrowska et al. [2008] also observed a high level of this electrolyte in the serum of polar female foxes, with the highest level determined in the one-year-old vixen group (6.72 mmol · l^{-1}). Similarly, the potassium concentration was high in two- and three-year-old female foxes, within the range of 5.95 mmol · l^{-1}, and decreased slightly with the age of the animals.

Yuzhakov et al. [2021] observed sex-related differences in blood potassium concentrations in domesticated reindeer. The average concentration of this electrolyte in the whole blood of the reindeer population studied was 26.94 ±0.56 mmol · l^{-1}, with a variability of 24.61%. In females, it averaged 22.97 mmol · l^{-1}, while in males, it was 28.37 mmol · l^{-1}. The authors have demonstrated that the high potassium concentration in animals inhabiting the subarctic region is genetically determined. They found that in domesticated reindeer, the ratio of "high-potassium animals" was 88.02% and "low-potassium animals" constituted 11.98%.

Aguirre et al. [2000] studied 21 young, 5- to 7-week-old wild polar foxes (10 males and 11 females). They demonstrated that the concentration of sodium in the blood averaged 146 mmol · l^{-1} (133–159), potassium concentration was 5.4 mmol · l^{-1} (4.7–6.1), and chloride concentration was 104.3 mmol · l^{-1} (93–114).

Crooks et al. [2000] studied an endangered population of the island fox (a species endemic to the Channel Islands) in the rainy and dry seasons. The latter authors did not observe differences in blood electrolyte levels in relation to season and sex. The sodium concentration in females was 146.82 mmol · l^{-1}, and in males, it was 146.5 mmol · l^{-1}. The potassium concentration was 4.15 and 4.12 mmol · l^{-1}, respectively, and the chloride levels were 114.64 and 114.17 mmol · l^{-1}, respectively. Inoue et al. [2012] also conducted extensive biochemical hematological studies on the endangered island fox (*Urocyon littoralis*) and did not observe differences in the concentration of sodium, potassium, and chlorides based on the age of the foxes. The sodium concentration in both young and old foxes ranged from 141 to 155 mmol · l^{-1}, potassium levels ranged from 3.7 to 5.5 mmol · l^{-1}, and chloride concentration ranged from 105 to 118 mmol · l^{-1}.

Piotrowska et al. [2008] demonstrated that in the blood of vixens, regardless of age (from 1 to 5 years), chloride concentration was high and ranged from 112.70 to 114.40 mmol · l^{-1}. These authors did not observe significant individual differences in sodium and chloride levels.

An indicator that characterizes the concentration of all osmotically active components of blood, apart from the main electrolytes – sodium, potassium, and chloride, is its molality. Plasma osmotic pressure in the present experiment was approximately 304 mmol l^{-1}, with very little individual variation. No differences were observed in the value of this indicator between the group of male and female foxes. The stabilized and sex-independent molality of the serum indicates efficient renal regulation of tubular resorption of the examined electrolytes and their excretion in the urine. In many laboratories worldwide (e.g., Antech Diagnostics, Phoenix Labs or West Los Angeles Animal Hospital), it is generally accepted that the osmotic pressure of the blood serum in carnivorous animals ranges from 270 to 320 mmol · l^{-1}. These values are considered as reference ranges.

CONCLUSIONS

The concentration of sodium, potassium, and chloride in the blood serum of six-month-old polar foxes, as well as the osmotic pressure of the blood serum, were within the upper limits of physiological normal range. Higher concentrations of sodium and potassium were observed in males of these animals. Nonetheless, the results indicated efficient regulation of blood electrolyte homeostasis, both in females and males.

ACKNOWLEDGEMENT

The research was supported with funds for statutory research of the West Pomeranian University of Technology in Szczecin.

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EFEKTYWNOŚĆ REGULACJI HOMEOSTAZY ELEKTROLITOWEJ KRWI LISÓW POLARNYCH (*ALOPEX LAGOPUS*, LINNAEUS, 1758)

STRESZCZENIE

Lis polarny (*Alopex lagopus*, L. 1758) ze względu na wysoką jakość skór zimowej okrywy włosowej jest utrzymywany w warunkach hodowlanych. Powodzenie hodowli tych zwierząt jest uzależnione przede wszystkim od ich zdrowia, które warunkuje wysokie wskaźniki rozrodu, niską śmiertelność neonatalną i prawidłowy rozwój młodych zwierząt. Warunkiem zdrowia jest utrzymanie stałości środowiska wewnętrznego, zwłaszcza homeostazy wodno-elektrolitowej i kwasowo-zasadowej. Głównymi elektrolitami wpływającymi na izowolemię i izoosmię płynów ustrojowych są sód, potas i chlorki. W niniejszej pracy przedstawiono wyniki dotyczące zdolności nerek sześciomiesięcznych lisów do regulacji natremii, kaliemii, chloremii i ciśnienia osmotycznego krwi, z jednoczesnym uwzględnieniem płci tych zwierząt. Stężenie sodu, potasu, chlorków w osoczu krwi młodych lisów polarnych oraz ciśnienie osmotyczne mieściły się w górnych granicach norm fizjologicznych. Obserwowano wyższą koncentrację sodu i potasu u samców tych zwierząt. Nie mniej wyniki wskazują na sprawną regulację homeostazy elektrolitowej krwi, zarówno u samic jak i samców.

Słowa kluczowe: lis polarny, osocze krwi, sód, potas, chlorki, ciśnienie osmotyczne krwi

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