<u>Original Article</u> Influence of Different Genders of Japanese Quail on the Functional State of Kidneys

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Abstract

Kidneys comprise the paired organ essentially responsible for excreting nitrogenous wastes, excessive water, inorganic salts, and toxic substances produced during the process of body metabolism. The maintenance of osmotic regulation and homeostatic fluid balance of the body is also performed by the kidneys. The current study investigated the histological features in quail birds of both genders at different age stages. A total of thirtysix adult male and female Japanese quail were allocated randomly into four different age groups of 30, 90, 180, and 270 days. They were fed high-protein food and water for two weeks, after which the kidneys of the quail were obtained and histological changes between males and females were evaluated. Data analysis was performed using SPSS-22. The results showed a 24% increase in Bowman's space in male quail compared with females after 30 days. Moreover, a 10% increase in Bowman's space was recorded at 270 days in male quail compared with females. The results showed a 12% increase in glomerular diameter in females compared with males. The data also showed a 12% increase in the diameter of Bowman's capsule in females compared to males. The outer diameter of the thin tubule in the loop of Henle in females increased by 4% compared to males. A 12% increase was noted in the outer diameter of the thickened tubule and the collecting tubes in male quail compared to the females at 30 days of age. Increases in the outer diameter of the proximal tubule of 6%, 16%, and 2% in female quail compared to males were recorded at 30, 180, and 270 days, respectively. Finally, the outer diameter of the distal tubule in males increased by 4% compared to females at 30 days and by 2% at 270 days. The current study described in detail the effect of a high-protein diet on the histology of different genders in quail kidney.

Keywords: histological, quail, high-protein, kidney

Influence des Différents Genres de Cailles Japonaises sur l'état Fonctionnel des Reins

Résumé: Les reins comprennent l'organe apparié essentiellement responsable de l'excrétion des déchets azotés, de l'excès d'eau, des sels inorganiques et des substances toxiques produites au cours du processus du métabolisme corporel. Le maintien de la régulation osmotique et de l'équilibre hydrique homéostatique du corps est également assuré par les reins. La présente étude a examiné les caractéristiques histologiques chez les cailles des deux sexes à différents stades d'âge. Au total, trente-six cailles japonaises mâles et femelles adultes ont été réparties au hasard dans quatre groupes d'âge différents de 30, 90, 180 et 270 jours. Ils ont été nourris avec de la nourriture et de l'eau riches en protéines pendant deux semaines, après quoi les reins de la caille ont été obtenus et les changements histologiques entre les mâles et les femelles ont été évalués. L'analyse des données a été

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effectuée à l'aide de SPSS-22. Les résultats ont montré une augmentation de 24% de l'espace de Bowman chez les cailles mâles par rapport aux femelles après 30 jours. De plus, une augmentation de 10% de l'espace de Bowman a été enregistrée à 270 jours chez les cailles mâles par rapport aux femelles. Les résultats ont montré une augmentation de 12% du diamètre glomérulaire chez les femelles par rapport aux mâles. Les données ont également montré une augmentation de 12% du diamètre de la capsule de Bowman chez les femmes par rapport aux hommes. Le diamètre extérieur du tubule mince de l'anse de Henlé chez les femelles a augmenté de 4% par rapport aux mâles. Une augmentation de 12% a été notée dans le diamètre extérieur du tubule épaissi et des tubes collecteurs chez les cailles mâles par rapport aux femelles à 30 jours d'âge. Des augmentations du diamètre extérieur du tubule proximal de 6%, 16% et 2% chez les cailles femelles par rapport aux mâles ont été enregistrées à 30, 180 et 270 jours, respectivement. Enfin, le diamètre extérieur du tubule distal chez les mâles a augmenté de 4% par rapport aux femelles à 30 jours, respectivement. Enfin, le diamètre extérieur du tubule distal chez les mâles a augmenté de 4% par rapport aux femelles à 30 jours, respectivement. Enfin, le diamètre extérieur du tubule distal chez les mâles a augmenté de 4% par rapport aux femelles à 30 jours, respectivement. Enfin, le diamètre extérieur du tubule distal chez les mâles a augmenté de 4% par rapport aux femelles à 30 jours, respectivement. Enfin, le diamètre extérieur du tubule distal chez les mâles a augmenté de 4% par rapport aux femelles à 30 jours, respectivement. Enfin, le diamètre extérieur du tubule distal chez les mâles a augmenté de 4% par rapport aux femelles à 30 jours et de 2% à 270 jours. La présente étude a décrit en détail l'effet d'un régime riche en protéines sur l'histologie des différents sexes dans le rein des cailles.

Mots-clés: histologique, caille, riche en protéines, rein

1. Introduction

A quail is a bird of the order of Galliformes, which are considered rather primitive birds, and most of the species in this order are medium-sized birds. Their body form and behavioral characteristics are similar to those of domestic chickens. In birds, the urinary system consists of elongated, paired kidneys and muscular ureters which drain each kidney and open into the urodeum of the cloaca (1). Birds have no renal pelvis or urinary bladder (1, 2). Undoubtedly, the kidneys play numerous vital roles in birds. One primary role of the kidney is the elimination of metabolic wastes as well as excess water (3). Avian kidneys also aid the liver in detoxification. Differing renal anatomies and physiologies lead to different renal diseases in birds compared to mammals.

The avian renal system is quite unique among vertebrate kidneys (4). In both mammals and birds, the kidneys are principal organsin concert with maintaining the unchanging nature of the internal environment by providing a balance between glomerular filtration, renal tubular secretion volume, osmolality, ionic content, and pH of body fluids. The kidney plays an important role in maintaining homeostasis. The urinary system of birds consists of large paired kidneys lying symmetrically one on either side of the vertebral column, drained by ureters that open into the urodeum of the cloacae; no urinary bladder is present in birds (5). The surface of the kidney is covered by a large number of small rough structures with shallow depressions between them. Each of these structures is formed from the cortical kidney. Eight or ten of these cortical units form a group that drains urine into a single medullary cone. This region of kidney tissue is known as lobules and consists of a few elongated units of cortical tissue together with the single medullary cone into which their urine flows. Medullary lobules are essentially cones surrounded by a connective tissue sheath open to the cortex at the wide end and attached to a major urethral branch at the other (6). The avian kidney has two types of nephron: the cortical type which is reptilian in form, devoid of a nephron Henle loop, and confined to the cortical region of the lobule; and the medullary type which has a nephron loop that penetrates the conical medullary region of the lobule similar to that in mammals (7, 8). One of the most unique characteristics of avian kidneys is the presence of 2 types of nephrons, one with and one without a loop of Henle (1, 9). Most avian nephrons are loopless or cortical (10, 11), and thus the capacity of the avian kidney to concentrate urine is limited and less than that of mammals (4). In mammals, all nephrons contain a loop of Henle; some of them are longer than others. The ability to conserve ions and water may be correlated with the structure of nephrons. The surface

of the kidney is covered by a large number of small rough structures with shallow depressions between them. Each of these structures are a unit of the cortical kidney. The avian glomerulus is similar to its mammalian counterpart, but it is smaller and has a simpler system of capillary loops arranged around the core of mesangial cells. The distal convoluted tubule (DCT) is characteristically different from the proximal convoluted tubule (PCT) in that the cells of the lining epithelium possess no brush border, and the epithelial cells are approximately cuboidal in shape (6).

The aim of the present study was to describe the histological characteristics of the Japanese quail kidney and to determine whether this characterization is sex dependent.

2. Material and Methods

Japanese quail (Coturnix japonica) were raised in the Bird Research Unit of the Agrarian and Technological Institute, People's Friendship Russian University, Moscow, Russia. The experiments were approved by the Animal Experiments Committee of the Agrarian and Technological Institute in December 2019. Thirtysix apparently healthy 30-, 60-, 180-, and 270-day-old male and female Japanese quail were used for the current experiment and classified into three groups according to their age. The experiments were carried out in February 2020.

The poultry was fed with complete industrial feed: PK-2 (0-30 days), DK-51 (30-44 days), and PK-1P (> 44 days). Part of the recipe comprised: wheat, corn, soybean meal, sunflower meal, gluten corn feed, vegetable oil, protein concentrate, lysine, methionine, threonine. table salt. monocalcium phosphate, limestone flour, baking soda, and complex feed additive for poultry (vitamins + trace elements). Quail food helps maintain the birds' health at the proper level. Normally, this is achieved by maintaining a certain level of protein in the diet. Up to 30 days of life, the chick needs feed mixtures in which 26% of proteins are present. For the next 2 weeks of life, young animals need to be fed with a lower percentage of proteins. They already have 17% protein. Adults need food that contains 21% protein.

The composition of the ration provided to the birds is summarized in the research material. The quail birds were given a highly enriched protein diet (10% over normal) at all stages of rearing. The protein level in the diet was regulated by changing the amount of soybean meal, lysine, methionine, and threonine in the diet. The amount of protein in the diet varied according to chick age: up to 30 days - 28% of proteins are present; 31-44 days - 19%; over 44 days - 24%. Birds at three different age stages (30-, 90-, 180-, and 270-day) were selected. The weight of the live birds was measured, and then the birds were slaughtered and the morphological changes on the kidneys resulting from the high-protein diet were studied. The experimental diets were corn-soybean meal based and formulated according to the nutritional requirements determined by Silva and Costa (12) (Table 1).

Quail were kept at a temperature of about 20°C and given a program of about 14 h light/day. The birds were housed with food and water. On the day of analysis at 9:00 am, bird body masses and histological kidney analyses were performed.

Morphometric analysis: After the birds reached 30, 90, 180, and 270days of age, the body mass of birds from each group was measured and the birds were sacrificed. The kidneys were excised and their masses were estimated.

The kidneys were dissected free of synsacral fossae. Tissue samples from the cranial, middle, and caudal divisions of each kidney were fixed in 10% neutral buffered formaldehyde for 24 h and processed to be embedded in paraffin in the routine manner. The transverse serial sections (5 μ m) were stained with H & E for general histological observation and a variety of techniques for types of fibers in the connective tissues: 1) Verhoeff's for elastic fibers, 2) Masson's trichrome for collagen fibers, and 3) Gomori's method for reticulum. To investigate the chemical character (pH)

of the secretion material, Alcian blue (AB) (pH 2.5) was used to determine acidic mucosubstances, and periodic acid-Schiff (PAS) reaction was employed to

determine neutral mucosubstances (13). Histological studies on stained sections were carried out by light microscopy.

Quality indicators of quail PK-2 ((complete fee)-30 days)	ed for	Quality indicators of complete feed for quail DK-51 (31-44 days)	Quality indicators of complete feed for quail PK-1P (> 44 days)		
Name	Unit rev.					
Metabolic energy of poultry	kcal / 100g	295	275	285		
Crude protein	%	28.0	19.0	24.0		
Crude fat	%	3.6	3.5	3.5		
Crude fiber	%	4.8	5.0	5.1		
Linoleic acid	%	1.8	1.8	1.8		
Lysine	%	1.15	0.85	1.05		
Methionine	%	0.55	0.45	0.50		
Methionine + cystine	%	0.75	0.70	0.72		
Tryptophan	%	0.2	0.2	0.2		
Threonine	%	0.65	0.45	0.55		
Ca	%	2.3	2.8	3.3		
Na	%	0.16	0.16	0.16		
Р	%	0.7	0.7	0.8		
P available	%	0.65	0.53	0.75		
Nacl	%	0.33	0.33	0.33		

Table 1. Composition of the diets provided to the birds

3. Results

The results of this study showed that when comparing the kidneys of male and female quail of different ages, significant differences were observed at the p<0.05level in the glomerular diameter, the diameter of Bowman's capsule, and Bowman's space. The data showed increases of 24% and 10% in Bowman's space in male quail at 30 and 270 days of age, respectively, compared with females. In 90- and 180-day-old birds, the data showed 4% and 6% increases, respectively, in Bowman's space in females compared with males. The results of the current study showed an increase in the diameter of the glomeruli in male quail by 24% compared with females at the age of 30 days and by 2% at the age of 90-270 days. Nevertheless, at the age of 180 days, an increase of 12% in glomerular diameter was found in females compared with males. The results further showed increases in the diameter of Bowman's capsule by 20%, 2%, and 8% in male quail compared with the females at 30, 90, and 270 days, respectively. However, at the age of 180 days, a 12% increase in the diameter of the Bowman's capsule was found in females compared with males.

	Age, days									
Donomotora	3	0		90	1	80	27	70		
Parameters	Μ	F	М	F	Μ	F	М	F		
	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$		
Bowman's space	0.24±0.083* 62%	0.15±0.07* 38%	0.13±0.01 52%	0.12±0.01 48%	0.09±0.00 47%	0.1±0.01 53%	0.11±0.01 55%	0.09±0.02 45%		
Diameter glomeruli	0.63±0.06 62%	0.38±0.01* 38%	0.49±0.03 51%	0.47±0.03* 49%	0.64±0.05 44%	0.81±0.04 56%	0.67±0.06 54%	0.65±0.06 46%		
Diameter Bowman's	$0.78 \pm 0.07 *$	0.53±0.03	0.64 ± 0.04	0.61 ± 0.04	0.64 ± 0.05	$0.81 \pm 0.04*$	0.67 ± 0.06	0.65 ± 0.06		
capsule	60%	40%	51%	49%	44%	56%	54%	46%		
Kidney capsule	0.26 ± 0.11	0.13±0.01	0.15 ± 0.02	0.22 ± 0.06	0.31 ± 0.02	0.40 ± 0.03	0.29 ± 0.01	0.44 ± 0.02		
thickness	67%	33%	41%	59%	0.44%	0.58%	40%	60%		

Table 2. Histological changes in Bowman's space, glomerular diameter, Bowman's capsule diameter and renal capsule thickness

In the course of the study, an increase of 12% in the outer diameter of the thin tubule in the loop of Henle was found in male quail compared to females at the age of 30 days and a 2% increase at the age of 90 days. In birds 180 and 270 days old, the data showed a 4% increase in the outer diameter of the thin tubule in the Henle loop in females compared with males. Females showed increases in the height of the epithelium lining of the thin tubules in Henle's loop of

2%, 18%, and 14% compared with males at 30, 90, and 180 days of age, respectively. At the age of 270 days, the males showed an increase of 4% in the height of the epithelium lining of the thin tubules compared with the females. Examining the inner diameter of the thin tubule in Henle's loop indicated increases in the inner diameter of males of 10%, 4%, 14%, and 22% at 30, 90, 180, and 270 days of age, respectively (Table 3).

					Age	davs			
Demonstern			30	9	0	18	60	2'	70
1 41 41	licters	М	F	М	F	Μ	F	Μ	F
		$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$
Henle's loop	outer diameter	0.48±0.03 56%	0.37±0.02* 44%	0.47±0.02 51%	0.46±0.02 49%	0.39±0.02* 48%	0.43±0.02 52%	0.46±0.01 48%	0.5±0.03 52%
of thin segment	lining epithelium height	0.24±0.10 49%	0.25±0.10 51%	0.11±0.01 41%	0.16±0.07 59%	0.09±0.01* 43	0.12±0.01 57%	0.12±0.01 52%	0.11±0.01 48%
	tube spangle diameter	0.23±0.02 55%	0.19±0.02 45%	0.22±0.03 52%	0.20±0.01 48%	$\begin{array}{c} 0.20 \pm 0.02 \\ 57\% \end{array}$	0.15±0.01 43%	0.30±0.03 61%	0.19±0.02 39%

Table 3. Histological changes; Henle's loop is thin-segment

Examination of the outer diameter of the thickened tubule in Henle's loop revealed an increase of 12% in the outer diameter of the thickened tubule in male quail compared with females at 30 days of age. At the age of 180 days, a 6% increase in the outer diameter of the thickened tubule was seen in females compared with males. In addition, the height of the epithelium lining of the thickened tubule in Henle's loop increased by 10% in males at the age of 30 days and by 12% at the age of 270 days compared with females. However, an increase of 16% was found in the height of the epithelium lining of the thickened tubules in female quail at 90 days of age and a 4% increase at the age of 180 days. In the outer diameter of the thick tubule in Henle's loop in male quail, increases of 12% at 30 days of age and 8% at 270 days of age were noted. At 180 days of age, however, the outer diameter of the thickened tubule in females had increased by 6% compared with males (Table 4).

Apparently, the discrepancy in the sequence of collecting tubules in female and male quail at different

age stages may be associated with functional nutritional needs, behavior, and the nature of the environment. In the course of the study, an increase of 12% in the outer diameter of collecting tubes in male quail was found at the age of 30 days. However, increases of 30%, 6%, and 14% were found in the outer diameter of the collecting ducts of female quail compared with males in 90-, 180-, and 270-day-old birds. The height of the epithelium lining in the collecting tubes of males showed increases of 20% and 6% compared with females in birds aged 30 and 270 days. However, female quail at the age of 90 days showed a 10% increase in the height of the epithelium lining of the collecting tubes compared with males. In addition, the inner diameter of the collecting ducts was studied, and the inner diameter of the collecting ducts in 30-day-old males was increased by 10% compared to females of the same age. However, increases in the internal diameter of the collecting ducts of 10% and 12% were noticed in female quail compared to males aged 90-180 days and 270 days of age, respectively (Table5).

Para	motors	3	60	90		1	180		270	
1 41 4	meters	М	F	М	F	Μ	F	Μ	F	
		$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$					
Henle's	outer diameter	0.59±0.02 56%	0.47±0.02 44%	0.50±0.02 50%	0.50±0.02 50%	0.47±0.02* 47%	0.53±0.02 53%	0.55 ±0.02 50%	0.54 ±0.02 50%	
loop of thick segment	lining epithelium height	0.12±0.01 55%	0.10±0.0 45%	0.10±0.01 42%	0.13±0.01 58%	$0.11 \pm 0.01 \\ 48\%$	0.12±0.00 52%	0.18±0.01* 56%	0.14±0.01* 44%	
	tube spangle diameter	0.32±0.03 56%	0.25±0.02 44%	0.26±0.02 50%	0.26±0.02 50%	0.25±0.02* 47%	0.28±0.01* 53%	0.30 ±0.03 54%	0.26±0.02 46%	

Table 4. Histological changes; Henle's loop thick segment

			Age, days									
Parameters		30		90		180		270				
		М	F	М	F	М	F	М	F			
		$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$			
	outer diameter	0.71±0.02* 56%	0.56±0.03* 44%	$\begin{array}{c} 0.65\pm0.03\\ 35\% \end{array}$	$\begin{array}{c} 1.19\pm0.03\\ 65\%\end{array}$	0.68±0.04 47%	0.76±0.05 53%	0.76±0.02 43%	$1.01\pm 0.08 \\ 57\%$			
Collecting tubules	lining epithelium height	$\begin{array}{c} 0.18\pm0.03\\ 60\% \end{array}$	0.12±0.01 40%	0.13±0.01* 45%	0.16±0.01* 55%	0.15±0.01 50%	0.15±0.02 50%	0.20±0.02 53%	0.18± 0.02 47%			
	tube spangle diameter	0.33±0.03* 55%	0.27±0.03* 45%	0.30±0.04 45%	0.36±0.04 55%	0.33±0.02 45%	0.36±0.03 55%	0.38±0.03 44%	0.49±0.05 56%			

Table 5. Histological changes in the collecting tubules

Data on the outer diameter of the collecting canal in quail showed an increase in males by 6% compared with females at the age of 30 days. However, the results showed an increase of 4% in the outer diameter of females compared with males at 90 days and of 18% at both 180 and 270 days of age. An increase of 2% was observed in the height of the epithelium lining of the collecting canal in males compared with females at the age of 90 days in the large bore diameter of the collecting canal in male quail, an increase of 6% was found compared to 30-day-old females. However, increases of 14%, 24%, and 20% were noted in the large inner diameter of female quail compared to males at 90, 180, and 270 days, respectively. In addition, the small-bore diameter of the collecting canal increased in males by 22% compared with females at 30 days and by 20% at 270 days. Nevertheless, a 2% increase was observed in the small inner diameter in females compared with males at 90 days and a 14% increase was seen at 180 days (Table 6).

The current results showed that the outer diameter of the proximal tubule in female quail increased by 6%, 16%, and 2% compared with males at 30, 180, and 270 days, respectively. An increase of 12% in the height of the epithelium lining of the proximal tubules was observed in female quail compared with males at the age of 30 days, and a 16% increase was seen at the age of 180 days. At the age of 270 days, the results showed a 4% increase in the height of the epithelium of the proximal tubule in males compared with females. In addition, the diameter of the inner tube of the proximal tubule increased in male quail by 22% compared with females at the age of 30 days and by 8% at the age of 90 days. However, at the age of 180 days, an increase in the diameter of the inner tube of the proximal tubule of 26% was seen in females compared with males (Table 7).

In the current study, the outer diameter of the distal tubule showed an increase in males of 4% compared to females at 30 days and of 2% at 270 days. Moreover, the results indicated the outer diameter of the distal tubule in females increased by 4% compared with males at 90 days and by 14% at 180 days. The results further revealed increases of 8%, 32%, and 22% in the height of the epithelium lining the distal tubules in females compared with males at the ages of 30, 90, and 180 days, respectively. In the inner diameter of the distal tubule, a 5% increase was found in females compared with males at 90 days and an increase of 8% was seen at 180 days. However, at 270 days of age, the results showed an increase of 8% in the inner diameter of the distal tubule in males compared with females (Table 8).

		Age, days									
Doror	notors	3	0		90	18	0	2'	70		
rarameters		М	F	Μ	F	Μ	F	Μ	F		
		$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	M ± m		
	outer diameter	1.25±0.07* 53%	1.09±0.05* 47%	1.11±0.04 48%	1.19±0.03 52%	0.95±0.06 41%	1.35±0.05 59%	1.47±0.10 41%	2.15±0.23 59%		
Collecting ducts	lining epithelium height	0.18±0.03 50%	0.18±0.08 50%	0.18±0.03 51%	0.17±0.01* 61%	0.11±0.01* 39%	0.17±0.02 61%	0.16±0.02 47%	0.18±0.02 53%		
	large tube spangle diameter	0.92±0.07* 53%	0.82±0.06* 47%	0.68 ±0.04 43%	$0.89 \pm 0.04 \\ 57\%$	0.6 ±0.06 38%	0.96±0.04 62%	1.10 ±0.06 39%	1.71±0.24 61%		
	small tube spangle diameter	0.43±0.06 61%	0.27±0.02 39%	0.27±0.02 49%	0.28±0.03 51%	0.23±0.02 43%	0.31±0.02 57%	0.25±0.03 60%	0.17±0.01 40%		

Table 6. Histological changes in the collecting ducts

Table 7. Histological changes; proximal convoluted tubules

			Age, days								
Daman	4	3	0	90		180		270			
Paran	heters	Μ	F	Μ	F	Μ	F	Μ	F		
		$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$	$M \pm m$		
Proximal convoluted	outer diameter lining epithelium height	0.46±0.02 47% 0.11±0.01 44%	0.51±0.05 53% 0.14±0.02* 56%	0.46±0.02* 50% 0.11±0.00 50%	0.46±0.02* 50% 0.11 ±0.01 50%	0.45±0.01 42% 0.10±0.00 42%	0.62±0.04 58% 0.14±0.00* 58%	0.41±0.02 49% 0.12±0.01 52%	0.43±0.03 51% 0.11±0.01 48%		
tubules	tube spangle diameter	0.28±0.03* 61%	0.18±0.02 39%	0.22±0.01 54%	0.19 ±0.01 46%	0.17±0.01 37%	0.29±0.02* 63%	0.19±0.02 50%	0.19±0.01 50%		

			Age, days								
D (30	90		18	80	2	70		
raran	leters	М	F	М	F	М	F	М	F		
		$M \pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$	$M\pm m$		
	outer diameter	0.46±0.01 52%	0.42±cc0.02 48%	0.41±0.01 48%	0.44±0.03 52%	0.40±0.02 43%	0.54±0.02 57%	0.38±0.03* 51%	0.37±0.02* 49%		
Convoluted tubules	lining epithelium height	0.16±0.07 46%	0.19±0.08 54%	0.11±0.01 34%	0.21±0.09 66%	0.09±0.00 39%	0.14±0.01 61%	0.10±0.01* 50%	0.10±0.01* 50%		
	tube spangle diameter	0.22±0.02 50%	0.22±0.03 50%	0.19±0.00 49%	0.18±0.01 51%	0.18±0.02 46%	0.21±0.01 54%	0.19±0.01 54%	0.16±0.01 46%		

Table 8. Histological changes; convoluted tubules

4. Discussion

4.1. Glomeruli (Glomeruli)

The results of the present study have shown that the glomeruli in quail, which are the subject of this study, can be divided into two types according to their regions of presence; cortical glomeruli, the first type, occupy a peripheral location of cortical tissue, while the second type is cerebral glomeruli that are located next to the cone. The pulp is also randomly distributed in the tissue of the cortex in the form of single, double, or triple groups. As a rule, glomeruli are small spherical structures consisting of a simple apparatus of capillary blood vessels, and their components differ in size, like the average outer diameter (Figure 1).



Figure 1. Cross section of a quail kidney showing the cortex (1), the medulla (2), the distribution of glomeruli in the tissue of the cortex which can be divided into cortical glomeruli (3) and glomeruli Medullary (4) (H&E spot).

The current study has further shown that the distribution of glomeruli into two types, cortical and cerebral, is consistent with the results of several studies on different birds (14, 15). The data showed that glomeruli are small spherical structures composed of a simple system of capillary blood vessels, which confirms what many other studies have reported on the glomeruli in birds, indicating that the glomeruli are bundles of capillary vessels that take a spherical or oval shape when viewed in cross section images (7).

Histological examination also showed that each glomerulus has one vascular pole and another

bladder pole, and cells with ellipsoidal nuclei at the vascular pole, called juxtaglomerular cells, which are in contact with a group of cells whose nuclei are located nearby, can be distinguished. The dense spot is called the macula densa. Histopathological examination also revealed that the glomeruli are surrounded by Bowman's capsule, which is characterized by two layers: an inner layer called the visceral layer and an outer one called the parietal layer. Its cells appear to be flattened epithelial cells, and there is a space between the two layers known as the capsule space, or Bowman's space (Figure 2).



Figure 2. Cross section of a quail kidney showing the histological structure of the renal corpuscle, showing the glomeruli (A), Bowman's space (1), visceral layer (2), urinary pole (3), parietal layer (4), vascular pole (5), macula densa (6), and juxtaglomerular cell (7) (H&E spot).

In this study, nephrons located at different depths in the kidney are consistent with the findings of Ritchison (16) on honey badgers and Al-Ajeely and Mohammed (11) on pigeons. Previous studies on many bird species have reported two types of nephrons in the kidney, i.e. the cortical type and the medullary type (3, 10, 11), but Islam, Khan (17) and Abood (18) reported that chickens have three types of nephrons. The observations made in the current study are consistent with their results. Small cortical nephrons were located at the periphery of the cortical zone, just below the renal capsule. This finding reflects the results of Abood (18) for chickens, harriers, and mallards. The glomeruli of small and medium nephrons were located mainly in the cortex (cortical or mesonephric type), but in large nephrons they were located in the medulla (mammalian type or metanephric type). Some researchers (1, 3, 10, 11, 18) have reported that the renal corpuscles consist of glomeruli, mesangium, and bilayer glomerular capsule (Bowman's capsule). The current results are consistent with those of the mentioned studies. In quail, a dense patch representing a region of specialized tall epithelial cells was found at the vascular pole of the renal corpuscle, consistent with the findings of Islam, Khan (17) in Rhode Island chickens and white leghorns and those of Batah A L (3) in coots.

The change in glomerular diameter and Bowman's space may be due to functional requirements as well as to the nature of the environment surrounding the body, resulting in a mismatch in the size, quantity, and thickness of the tissue structure involved in the formation of various organs in the body (19).

4.2. Proximal Convoluted Tubule

Histological examination showed that the anterior part of the urinary tubule, which attaches to Bowman's capsule, is a proximal convoluted tubule (PCT), the lining of which consists of a single row of simple cuboidal epithelial tissue overlying the basement membrane. Round nuclei located in the center and dark in color, and the free surface of its cells contain long microvilli, which usually form the so-called brush border (Figure 3).

The results of the current study on the proximal convoluted tubule in quail were similar to those of several other studies which indicate that the lining of the tubule is composed of simple cuboidal epithelium tissue. The free surface of its cells has the edge of the hand (BB), and this is of great importance for increasing the absorption process (14, 20).

While some researchers have reported that proximal convoluted tubules are lined with a single layer of cuboidal cells (3, 10, 11, 18), others have noted that these tubules appear to be simply columnar (1). The Japanese quail had one layer of large cuboid cells. Recorded accumulations of apical microvilli of proximal convoluted tubules of buds in other avian species are consistent with the current results (1, 3, 10, 11, 18).

PAS-positive reactions of all epithelial cells of the proximal convoluted tubules in Japanese quail reflect the results of Al-Ajeely and Mohammed (11) in pigeons, but Nabipour, Alishahi (10) did not report PAS reactions in pigeons and owls. This positive reaction indicated the presence of sialic acid in mucosal components (21).

The lumen surface of the proximal convoluted cells of Japanese quail reacted positively to acidic mucopolysaccharides, whereas Nabipour, Alishahi (10) and Al-Ajeely and Mohammed (11) reported no AB reactions in pigeons, doves, and owls. Casotti and Braun (22) demonstrated that the presence of acidic mucopolysaccharides is involved in the prevention of tubal lumen obstruction.



Figure 3. Cross section of a quail kidney showing the area of the cortex (1) and its contents of parts of the renal unit, glomeruli (2), proximal convoluted tubules (3), distal convoluted tubules (4) and brush border (5) (H&E spot).

4.3. Distal Convoluted Tubule

Histological examination of transverse sections of the quail flock, which is the subject of this study, showed that the distal convoluted tubule is located within the cortical tissue and, apparently, is smaller in length and diameter than the proximal convoluted tubule. Furthermore, it is lined with cuboidal epithelial cells devoid of a brush edge, so it can be distinguished from the proximal convoluted tubule. The nuclei of its cells are spherical and occupy a central place in the cytoplasm (Figure 3).

The results of the current study regarding the distal convoluted tubule confirmed the findings of other researchers about the histological structure of the kidneys in different birds (14, 23). Distal convoluted tubules of the kidney in quail were consistent with previous studies (1, 3, 10, 11, 18). This is a large overlap of the histological structure associated with the nature of the distal convoluted tubule, associated with a similar functional structure in different vertebrates, which seems to be closely related to the histological structure plan, which is consistent with the functional specificity of each part of the kidney (14, 19, 24).

On the other hand, the results of the present study showed that there were significant differences (p < 0.05) between the mean outer diameter of the distal convoluted tubule in females and males, while there were no significant differences in the thickness of the inner lining of the distal convoluted tubule between female and male quail (Table 8).

Histological examination of the pulp area showed that it contains areas of a thin segment and another thick segment of Henle's loop, where areas of a thin segment appear to have a lining of simple cuboidal epithelial tissue, and its cells look like. They have a pyramidal shape with spherical nuclei with a central location, occupy a large area of cytoplasm and dark pigment, while the thick segment is also lined with cuboidal epithelial tissue, the cells of which seem to be thicker than in the thin segment, which has centrally located nuclei and receives more intense pigment compared to nucleated ones. The cytoplasm of the cells of the thick segment seems to be more pigmented than the cytoplasm of the thin segment; thus, it should be noted that the epithelial lining of the thin and thick segments of Henle's loop lacks the edge of the brush in the free surfaces of its cells (Figure 4).

The results of the current study support what others have found (7, 14), while they also show inconsistency with other studies that have dealt with the kidneys in vertebrates other than birds. This discrepancy may be due to variations in histological structure based on the functional specificity of the loop of Henle in different species (25).

Furthermore, the results of the present study have shown significant differences (p < 0.05) in the average diameters of the thin and thick Henle loop when comparing female and male quail (Tables 3,4). These differences may be associated with variations in the histological structure associated with functional specificity or with differences in diet and environment.



Figure 4. Cross section of a quail kidney showing the medulla oblongata (1) and its contents of the parts of the renal unit, thin segment (2) and thick segment (3) Henle loops, collecting tubule (4), collecting duct (5) (H&E spot).

4.4. Henle's Loop

4.5. Collecting Tubule

The results of the current study regarding the quail kidney, the subject of study, showed that the collecting ducts are larger than the proximal and distal tubules, and, apparently, occupy a large area. The lining of the cone of the pulp is cubic cells and can appear as columnar; it has spherical nuclei, which are located closer to the base of the cell than to its center and are dark in color, while the collecting ducts are elongated, appear larger in diameter than the collecting ducts, and have a lining consisting of a simple columnar epithelial tissue with oval or spherical nuclei, which occupies a basal position in the cell and is highlighted in bold (Figure 4).

The results of the current study confirm what researchers mentioned herein have reported, and this confirms the findings of other researchers regarding the kidney in birds (11, 14, 15).

In the current study, collecting ducts were lined with a single layer of cuboid or low table to thick cuboid. In contrast, Al-Ajeely and Mohammed (11) and Batah A L (3) reported only a simple cuboid lining in pigeons and coots. Thus, it can be concluded that the cellular activity of the collecting tubules in quail, chi in you, in quail

The current results also showed that there are significant differences (p < 0.05) in the average external diameter of the collecting ducts between female and male quail birds at different ages (Table 5).

4.6. Collecting Duct

The collecting ducts observed in the current study were lined with a simple columnar epithelium, which was similar to the data of Nabipour, Alishahi (10), but Abood (18) reported these tubules to be lined with simple cuboidal epithelium. Unlike other birds(1-3, 10, 11, 18), the characteristic border intersecting microvilli that protruded into the terminal network were observed on the luminal surfaces of the epithelial cells of the collecting canals of the kidneys in Japanese quail. This new data again indicates that quail's cell harvesting activity is higher than that of other birds studied. In the present study, epithelial histochemical reactions in the collecting ducts were similar to those in other poultry (10, 11). The collecting ducts reabsorb water and sodium from the renal tubules. In addition, they secrete mucus to prevent uric acid deposition; thus, they are responsible for keeping the tubule lumen open.

In conclusion, the present study is the first to report the histological and histochemical structures of the kidney in Japanese quail. The histological and histochemical properties of the kidney in Japanese quail were generally similar to those of chickens and some other species, except for the presence of three types of nephrons, a distinctive brush border and terminal web on the epithelial cells of the collecting ducts, the presence of all the connective tissue fibers in capsule and interlobular septa, the presence of the reticular and collagenous fibers in intertubular interstitial connective tissue and around renal corpuscles, the absence of smooth muscle fibers in the capsule, and the AB-reactions of the proximal convoluted cells. The various histological structures of the kidney showed no considerable differences among various divisions of the left and right kidneys. There were no significant effects of sex on the histology and histochemistry of the quail kidney.

Authors' Contribution

Study concept and design: Z. A. A. and N. I. K. Acquisition of data: A. N. and V. I. S. Analysis and interpretation of data: Y. A. V. and T. V. M. Drafting of the manuscript: A. A. N. and N. Y. S Critical revision of the manuscript for important intellectual content: E. V. K. Statistical analysis: R. R. G. Administrative, technical, and material support: E. A. K

Ethics

All the procedures and animal handling were approved by the Animal Ethics committee at the Peoples Friendship University of Russia (RUDN University), Russia, Moscow under the project number of 2021-1263764-3.

Conflict of Interest

The authors declare that they have no conflict of interest.

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