

## **Histomorphological Comparative Study of the Adrenal Glands in Local Guinea Fowl (*Numida Meleagris*) and Muscovy duck (*Cairina Moschata Domestica*)**

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### **Abstract:**

The thorough concept of histomorphology of adrenal glands might incompletely be advanced via a comparative histochemical study on the avian class's different representatives. Thus, this study was aimed to examine this particular portion of the histological features of the adrenal glands in the local Guinea fowl *Numida meleagris* and Muscovy duck *Cairina moschata domestica*. Ten adult birds, each of Guinea fowl and Muscovy duck were used in this study. Anatomically, in both birds, the adrenal gland showed a small paired structure lying in the celomic cavity placed on each side alongside the cranial ends of the kidneys. The left gland was triangular whilst the right gland was elongated, and the color varies from yellowish to reddish-yellow. Histologically, the gland was surrounded by a connective tissue capsule sending thin trabeculae to enter the parenchymal glandular tissues. The glandular interstitial tissue was rich in blood vessels, and sinusoids. The parenchyma showed cortical tissues intermingled with the medullary cells. The cortical tissue was separated into the peripheral/subcapsular zone and inner/central zone. The subcapsular zone was located close to the capsular area and organized in the mesh of arch-like cords, while the central zone showed straight and curved cords with columnar cells. These zones were intermingled with the basophilic medullary cells that are termed chromaffin. The obtained data in the current study is assumed to contribute to understanding the histomorphological structures of the local Guinea fowl and Muscovy duck adrenal glands.

**Keywords:** peripheral zone, central zone, adrenal, Guinea fowl, Muscovy duck.

### **INTRODUCTION**

Guinea fowl are abundant among the oldest of the gallinaceous birds (Christopher, 2005). The Muscovy duck is a large duck native to Mexico (Ricardo de Matos, 2008). The avian adrenal glands are paired yellowish/orange organs, with a pear/triangle shape placed close to the superior ends of the cranial kidney lobe and surrounded by fatty tissue (Sadoon, 2018, Colcimen and Cakmak 2020). Adrenal glands have a vital function such as metabolism, maintains homeostasis and/or stress response (Fathima *et al.*, 2012; Humayun., *et al.*, 2012; Schinner *et al.*, 2012; Peng *et al.*, 2005; Qureshi *et al.*, 2020; Himmelstein *et al.*, 2021). These functions are affected by devoid the birds from the gland and can lead to death after a period (Gulmez *et al.*, 2004). The intermingling of avian parenchymal adrenal glands are made it difficult to be divided into the three distinct zones (z.glomerulosa, z. fasciculata, and

z. reticularis) as in mammalian counterparts (Bancroft *et al.*, 2013.). This study was intended to explore the histomorphology of adrenal glands through comparative histochemical studies. Thus, this study was aimed to examine this particular portion of the histological features of the adrenal glands in the local Guinea fowl and Muscovy duck.

## MATERIALS AND METHODS

### Sample collection

The study was achieved under the rigorous commitment to the guidelines of concern and use of animals in the study, teaching, and testing (Olfert *et al.*, 1993), following ethical approval by The College of Veterinary Medicine, University of Diyala. Ten mature birds each local Guinea fowl *Numida meleagris* and Muscovy duck *Cairina moschata domestica* were used in this study. The body weight was recorded then the birds were euthanized with an overdose of a mixture of 25mg and 5mg/kg ketamine and xylazine respectively (Hess, 2005). All morpho-biometric records were performed promptly after body dissection. The birds in each group were weighed and then the position, shape, color, weight, length, width, and boundaries of each adrenal gland were recorded. For the histological study, the specimens were instantly collected from dissected birds and then fixed into the 10% neutral buffered formalin (NBF) for 24 hours. Then proceed with routine histological technique steps (Randall *et al.*, 2002).

### Statistical data analysis of the adrenal gland:

Data were calculated by Statistical Package (SPSS, IBM Corp.). T-test was used to find the difference within each group while ANOVA single factor test used to find the difference between groups for each parameter under investigation. was used for The correlation between body weight and parameters of adrenal glands was performed using Pearson's correlation coefficient. P-value of  $\leq 0.05$  and  $\leq 0.01$  (two-tailed) was set to be statistically significant.

## RESULTS

**Grossly**, adrenal glands were indicated as small paired organs, positioned on each side of the median line in the abdominal cavity. They were placed opposite the caudal margin of the lungs and the cranial division of both kidneys, and anterior to the junction of the caudal vena cava. In the male, the right gland was observed triangular in shape and its ventral surface was detected close to the right testis. Whilst, the left gland was elongated oblong and adjacent to the frontal part of the left testis. In the female, the ovary fully covered the ventral surface of the left adrenal gland (Figure 1). Their color in guinea fowl showed variation between yellowish to reddish-yellow or creamy yellow (Figure 2). This variation in color and shape was also showed in the adrenal gland of the Muscovy duck. The color was a range from reddish-yellow to greyish-yellow. The right gland was roughly pyramidal with an apex pointing caudally, while the left one was elongated and flattened dorsoventrally (Figure 3).

As shown in Table (1), a significant difference was recorded in Guinea fowl regarding the length and width of the right and left adrenal glands ( $p = 0.05$ ). However, no significant difference was recorded for the thickness and weight. Table (2) showed a significant difference between Muscovy ducks regarding the length ( $p = 0.05$ ), however, no significant difference was recorded for the weight and thickness of the right and left adrenal glands. The same in Table (3), a significant difference was recorded between Guinea fowl regarding the

bodyweight ( $p = 0.000$ ). Finally, Table (4), as well showed a significant difference between animals regarding body weight, ( $p = 0.000$ ).

Table 1: Differences between adrenal glands in Guinea fowl

Parameters of adrenal glands	Right adrenal gland	Left adrenal gland	P-value
Length (mm)	6.67	7.15	0.05
	5.95	7.13	
	6.62	7.11	
	6.46	7.9	
	5.20	6.11	
P value	0.000	0.000	
Width(mm)	3.70	4.63	0.002
	3.53	4.44	
	3.66	4.53	
	3.49	3.93	
	3.55	3.9	
P value	0.000	0.000	
Thickness (mm)	3.51	3.37	0.641
	3.4	3.31	
	3.45	3.33	
	2.74	3.2	
	2.62	3	
P value	0.000	0.000	
Weight of gland( g)	0.27	0.29	0.294
	0.22	0.26	
	0.25	0.26	
	0.20	0.23	
	0.20	0.21	
P value	0.000	0.000	

Table 2: Differences between right and left adrenal gland of a Muscovy duck

Parameters of adrenal glands of Muscovy ducks	Right adrenal gland	Left adrenal gland	P-value
Length (mm)	8.91	9.72	0.027
	8.17	8.94	
	8.48	9.10	
	7.80	8.75	
	8.90	9.81	
P value	0.000	0.000	
Width (mm)	4.38	5.19	0.346603
	4.14	3.9	

	<b>4.2</b>	<b>4.8</b>	
	<b>4.1</b>	<b>3.7</b>	
	<b>4.3</b>	<b>5.1</b>	
<b>P value</b>	<b>0.000</b>	<b>0.000</b>	
<b>Thickness (mm)</b>	<b>3.38</b>	<b>3.34</b>	<b>0.395628</b>
	<b>3.12</b>	<b>3.08</b>	
	<b>3.26</b>	<b>3.20</b>	
	<b>3.1</b>	<b>2.94</b>	
	<b>3.31</b>	<b>3.22</b>	
<b>P value</b>	<b>0.000</b>	<b>0.000</b>	
<b>Weight of gland( g)</b>	<b>0.68</b>	<b>0.70</b>	<b>0.505623</b>
	<b>0.33</b>	<b>0.4</b>	
	<b>0.48</b>	<b>0.57</b>	
	<b>0.21</b>	<b>0.34</b>	
	<b>0.55</b>	<b>0.61</b>	
<b>P value</b>	<b>0.005</b>	<b>0.001</b>	

Table 3: Differences between experimental animals bodyweight (gram) in Guinea fowl

Parameter	Test Value = 0					
	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
<b>Body weight (gr)</b>	<b>36.019</b>	<b>4</b>	<b>0.000</b>	<b>41.24200</b>	<b>38.0629</b>	<b>44.4211</b>

Table 4: Differences between experimental animals body weight (g) of a Muscovy duck

Parameter	Test Value = 0					
	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
<b>Body weight (g)</b>	<b>15.736</b>	<b>4</b>	<b>.000</b>	<b>2.12000</b>	<b>1.7460</b>	<b>2.4940</b>

### Histological results:

The adrenal gland was covered by a thin capsule comprised of collagen and reticular fibers with fibroblasts and blood vessels. Short thin trabeculae originated from the capsule penetrate the parenchyma of the gland (Figure 4). The interstitial tissue was rich in blood vessels, and sinusoids (Figure 5). The adrenal parenchyma revealed an obvious external cortex and an internal medulla. The cortex was contained three zones: (z.glomerulosa, z. fasciculata, and z. reticularis), however, this study revealed there were cortical tissues intermixed with medullary cells, it was also showed that the cortex separated into two zones: peripheral/subcapsular zone (SCZ), and inner/central zone (CZ). The cells of the SCZ

exhibited staining tractably than the central ones (Figure 5). The cells of SCZ were found closely further down the capsule and ordered as it is a bulky dense arch-like/coiled cords normally anastomosing with one another and scattered with clusters of medullary cells enclosed by a basement membrane. Each cord encompassing cells which were vast, polyhedral to columnar of flipantly stained acidophilic and extremely vacuolar cytoplasm with a round/oval and peculiarly located nucleus reset on a basement membrane (Figure 5). However, central zone CZ indicated straight and arched threads each possessed paired rows of columnar cells with basally positioned nuclei and enfolding by medullary cells. They were lightly stained with a smaller amount of cytoplasmic vacuoles than the subcapsular cortical cells. Small groups of capillaries sinuses were noticed between the cortical cords (Figure 6). The additional form of cells in the CZ was basophilic/chromaffin cells organized in uneven small islets and/or even meshwork groups intermixed among the cortical tissues. They were shown in both SCZ and CZ with larger sizes nonetheless clusters of chromaffin cells in SCZ were less frequent than that of the CZ. These cells seemed small dark bluish groups of islets that concentrated near the sinusoids and placed in the middle of the acidophilic cells. They were polygonal in shape having a basophilic cytoplasm with dark rounded centrally positioned nuclei (Figure 6).

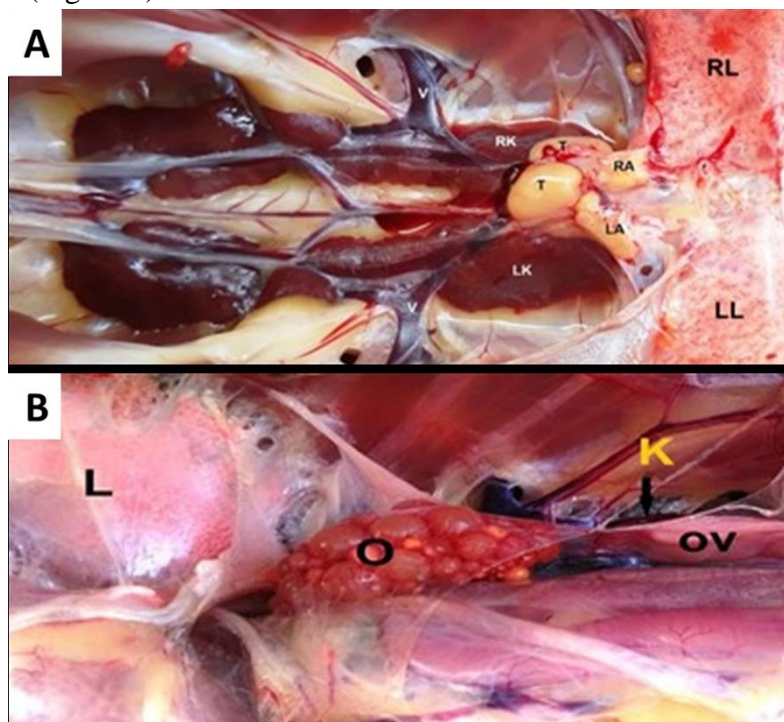


Figure 1: Photograph of the Guinea Fowl bird showed: (A): Male: RL, LL: right and left lungs, RA, LA: right and left adrenal gland, RK, LK: right and left kidneys, T: testis, V: bifurcation of the caudal vena cava. (B): Female showed: L: lung, O: Ovary: fully covered the adrenal glands; OV: Oviduct, K: Kidney.

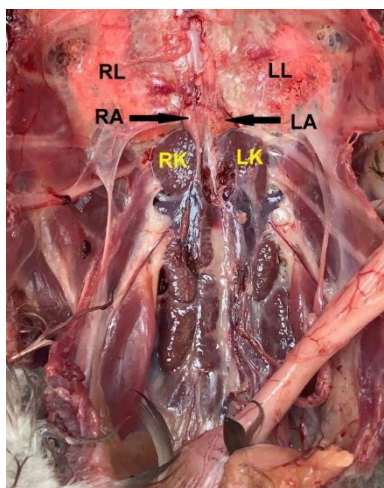


Figure 2: Photograph showed the anatomical position of the adrenal gland in Muscovy duck shows (RL, LL) right and left lungs, (RA) right adrenal gland, (LA) left adrenal gland, (RK, LK) right and left kidneys.

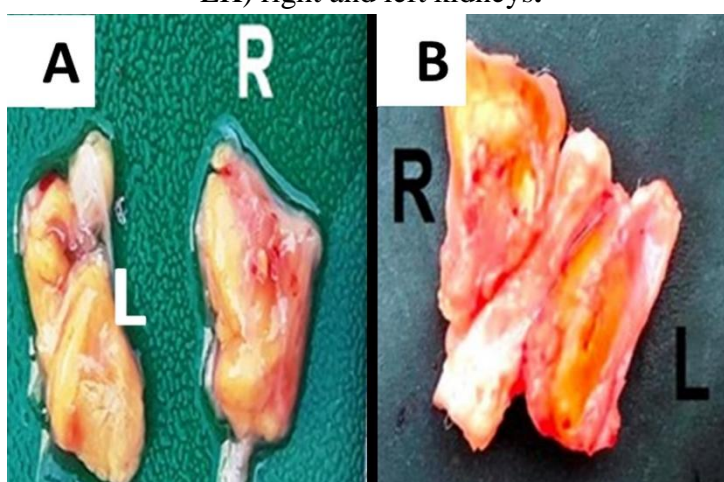


Figure 3: Photographs showed the anatomical dissection of the right and/or left adrenal glands A) Shows the adrenal glands in Guinea Fowl. B) Shows the adrenal glands in Muscovy duck.

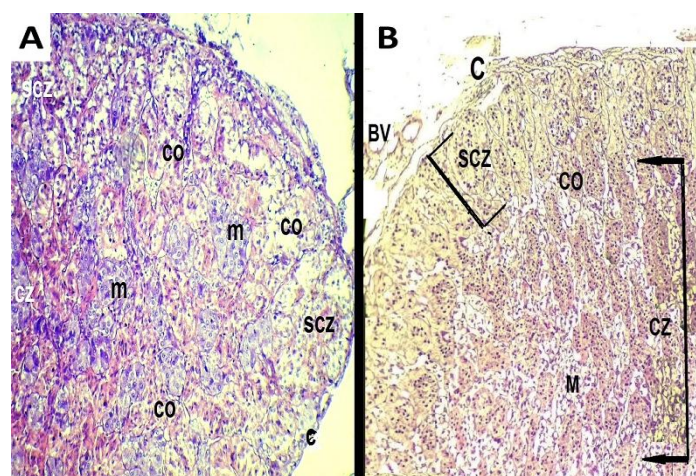


Figure 4: Photomicrographs illustrate the histological section of the adrenal gland A) Shows a cross-section of the adrenal gland in Guinea Fowl, while in (B) shows in Muscovy duck,

showed c- capsule SCZ: subcapsular zone CZ: central zone co: cortical tissue m: medullary tissue, BV: blood vessels. (H&E stain 100 X).

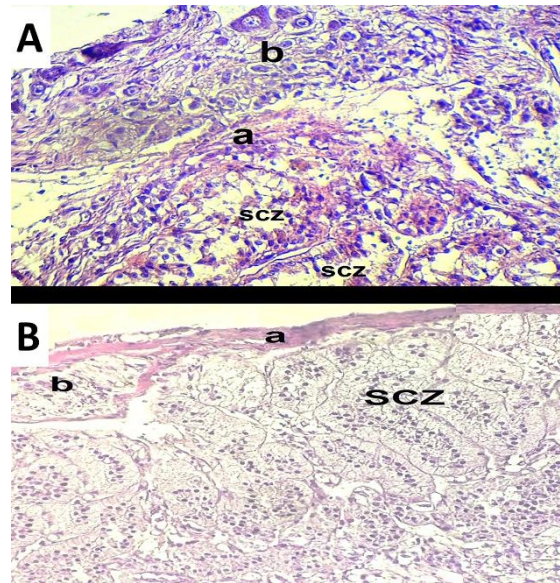


Figure 5: Photomicrographs show the histological section of the adrenal gland A) Shows a cross-section of the adrenal gland in Guinea Fowl, whereas (B) shows in Muscovy duck, capsule (a), autonomic ganglia (b), subcapsular zone (SCZ). (H&E stain A:200 X, B:400X).

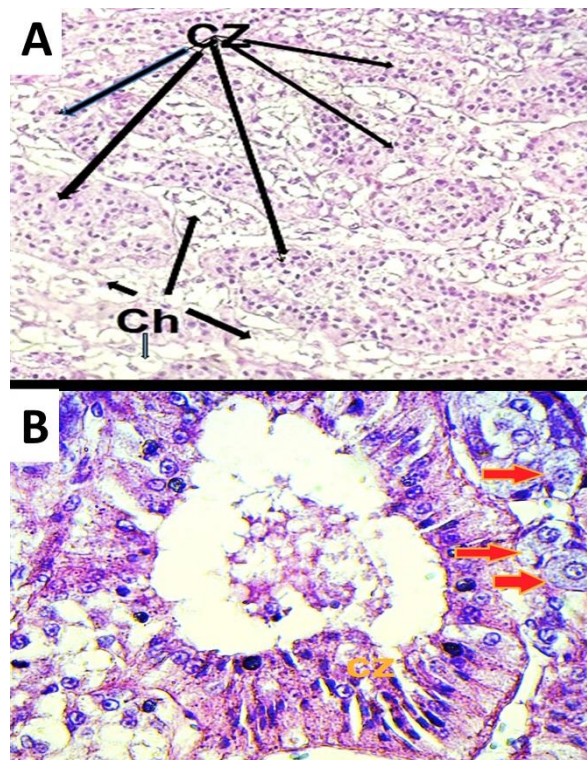


Figure 6: Photomicrographs show the histological section of the adrenal gland A) Shows a cross-section in Guinea Fowl, whereas (B) in Muscovy duck, the inner cortical cells (CZ) or (central zone), medullary or (chromaffin) (ch) represented in red arrows cells (H&E stain 400 X).

## DISCUSSION

**Anatomical Discussion:** The adrenal gland in both Guinea fowl and Muscovy Duck is parallel to those described by Humayun *et al.*, (2012) in Chicken and by Fathima *et al.* (2012) in duck. As it is in most birds, they are described as two separate distinct organs: right and left as in most birds, however in few birds such as whitehead sea-eagle, and crane, glands are combined into one organ. Whilst other birds such as woodpeckers and silver gulls shown an entirely united gland (Zhang, 1988). The differences between right and left glands in the Guinea fowl were analogous to that stated by Kober *et al.* (2012) in chicken; although the shape in Muscovy Duck is similar to that described by Fathima *et al.* (2012) in duck. They were showed differences in color and shape of the adrenal glands in Guinea fowl and that were similar to those in earlier studies by Peng *et al.* (2005) in domestic fowl and by Tang *et al.* (2009) in ostrich, whereas the variation in color in Muscovy duck is similar to that stated by Al-Jebori *et al.* (2016) in duck.

### **Histological Discussion:**

The adrenal gland in both birds was surrounded externally by a thin connective tissue capsule similar to what was shown previously by Humayun *et al.* (2012) in chicken and by Tang *et al.* (2009) in ostrich. These outcomes were in agreement with Salam, (1981) in laying fowls and broiler; and ( as well as with Suzan, (2012) in turkey. The adrenal parenchymal tissue of adult Guinea fowl was quite different from that of mammals; thus, this is in agreement with the outcomes in most birds; domestic fowl and chicken (Ghosh, 2001), Egyptian geese (El-Zoghby, 2015). The cortical tissues encompassed the peripheral and the inner/central zone, these outcomes were following those of Al-Jebori *et al.* (2016) in duck and Moawad *et al.* (2017) in chicken. Although, some studies done by Kober *et al.* (2012) and Sarkar *et al.* (2014) in chicken showed that adrenal cortical tissues were divided into three zones; subcapsular, peripheral, and central zones. The cells of the SCZ were flippantly stained than the central ones. The cells of SCZ were located directly further down the capsular tissue. These results were following those of (Gulmez *et al.*, 2004); (Wang *et al.*, 1999) in geese; (Moawad *et al.*, 2017) in chicken. While the inner cortical cells/central zone indicated straight and curved cords in longitudinal sections, similar outcomes were described by Humayun *et al.*, (2012) in chicken and Al-Jebori *et al.*, (2016) in duck. It was also indicated the second type of cells in the inner zone which was basophilic/chromaffin cells and was analogous to outcomes described by Basha *et al.*, (2009) in Japanese quail and Suzan, (2012) in turkey. These cells seemed in the form of small dark-bluish assemblies of islets, these findings were in agreement with El-Zoghby, (2015) in geese. In conclusion, the obtained data in the current study is assumed to contribute to understanding the histomorphological structures of the local Guinea fowl and Muscovy duck adrenal glands.

### **Conflict of interest**

The authors declare that they have no conflict of interest.

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