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## MORPHOMETRICAL AND HISTOLOGICAL CHANGES IN DOMESTIC DUCK SALT GLANDS IN RESPONSE TO SALINATED WATER

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### ABSTRACT

The salt glands of freshwater and saltwater drinking domestic ducks were investigated morphometrically and histologically. In salt drinking ducks, the gland was dark red in color, larger in size and heavier in weight than the fresh water duck, in which the gland was red in color, smaller in size and lighter in weight. However, the principal cells were hypertrophied and the basal cells were highly proliferated. The connective tissue was thinner and the capillaries were congested. This study suggested that the marked changes in size, weight and color of salt gland during saltwater adaptation were due to a hypertrophy of the principle cells, increase in basal cell number and the congestion of blood vessels.

**Keywords:** Domestic duck, salt gland, hypertrophied, principle cells.

### Introduction

The salt glands (nasal gland) are very important adaptive tool in many animals species which live in osine, salty lacks and aired land such as water birds, snakes and crocodile (Sabat, 2000; El-Gohary, 2009; Babonis *et al.*, 2011; Cramp *et al.*, 2008). The salt gland have been reported in many different avian orders (Spheniciformes, Precelariiformes, Charadriiformes, Pelecaniformes, Anseriformes, Phoenicopteriformes) (Jarrar, 2009; El-Gohary *et al.*, 2013; Maysoon, 2012; Reshag *et al.*, 2016). Since the avian kidneys are not sufficient in excreting salt and electrolytes to control body fluid homeostasis (Holmes and Phillips, 1985). The marine birds used post-renal transport mechanisms to eliminates the excess NaCl and conserve water, in which salt glands had the major role (Lavery and Skadhauge, 2008). This mechanisms enable many birds species to switch between freshwater and salty water. The salt gland is a paired crescent-shaped gland, located under the skin up each eye. The salt gland consisted longitudinal lobules contains many secretary branched tubules opined in central canals, each secretary tubule lining with cuboidal epithelium (Hussein *et al.*, 2006; Al-Mansour, 2009; Waleed, 2016). The duct system of gland end with one main duct, in turn, form secondary and primary ducts running toward the nasal cavity and discharging the hypertonic fluid through the nose (Butler *et al.*, 1991). The avian salt gland developed and function after hatching (Rubega and Oring, 2004; Woodin *et al.*, 2008; Kim *et al.*, 2020). The aim of study was designed to investigate the morph metrical and histological response of domesticate duck salt gland to high concentration of salt in drinking water for long period.

### Materials and Methods

Twenty adults healthy local domestic ducks (with no respect to sex) were pouch from the local markets of Basra provinces. The birds were grouped into two groups (control and salt stress). The control group late to drink fresh water, the salt stressed group watered on salt water contained 3 % NaCl for 45 days (The water was given in plastic pools and replenished fourth time daily). All birds were sacrificed by decapitation. Both left and right salt glands were quickly removed from each bird of group. salt glands sizes and weight were measured. then immersed in 10% neutral buffered formalin for 72 hours. The paraffin sections at (5-7)  $\mu\text{m}$  thick were prepared and stained with Hematoxylin and eosin stain (Bancroft and Marily, 2002). Statistical analysis of the present study was carried out using one ways ANOVA.

### Results

The salt glands of control group and salt water group ducks were crescent in shape with two pointed anterior and posterior extremities, the dorsal boarder was carved, the ventral boarder which face the eye ball was concaved, the gland was firmly attached with supra orbital depression in the frontal bone, The skin of the head directly covered the gland. The color of the gland in control ducks was red, in treated ducks the color of gland was dark red (Fig. 1, 2).

### Morphometric results

The current study recorded clear increase in the size and weight of the salt gland in treated duck in compression with the same parameters of control group ducks. The mean weight of gland in treated birds was  $(0.33 \pm 0.11)$  gm. It was

heavier than the gland of control group birds which weighted ( $0.136\pm 0.02$ ) gm. The same difference was recorded about the size of gland, The size of gland in treated birds was bigger it was ( $0.324\pm 0.06$ ) cubic cm., The size of salt gland recorded in control group ducks was ( $0.144\pm 0.04$ ) cubic cm.

### Histological results

Histological investigation of salt gland in both ducks groups revealed that the main histological structures were the same. The gland was consisted of many longitudinal tubes packaged together by connective tissue capsule, In histological section each tubule appeared as honey combs or polygonal structures, each of them surrounded by thin connective tissue with blood capillaries. In side this structures numerous branched secretory tubules runs in radical direction from the centre to the margin. The centre of this structures were occupied by centre canal. The secretory tubules were branched with two ending the inner end opened directly in the centre canal, the other end was blind forming the peripheral region of the secretory tube (Fig. 3, 4). The lining cells of secretory tubules were simple epithelium of two types of cells arranged in one layer, the basal cells which located at the peripheral end of the secretory tubules .This cells were small and rounded in shape with dark irregular nucleus. The second type was the principle cells which were cuboidal in shape with rounded dark stained nucleus and eosophilic cytoplasm. The principle cells numerous, lining the majority of secretory tubules (Fig. 4,5). The histological study of the salt gland belong to ducks which drink salt water showed the flowing changes, The shape of each polygonal structures lost it regularly and changed to oval or rounded shape. The connective tissue which surrounded the tubes was thin (6) important cellular changes were noticed, the principle cells changed to round in shape and hypertrophied, there cytoplasm was light stained and the nucleus was large and faint (Fig.7,8). Another important changes occurred in the peripheral region, There was proliferation and increase in the peripheral or basal cells and the blood capillaries were congested (Fig. 8).

### Discussion

The gross or anatomical feature of the salt gland in both experiment group birds, which includes the crescent shape and supraorbital location were as the same as what was mention in all most all water birds such as (ducks, geese, common coot, moor hen, and flamingo) which was studied by (Maysoon, 2012; Hussein *et al.*, 2006; Al-Mansour, 2014; Reshag *et al.*, 2016). with some exception, the shape of salt gland in Kentish plovers and love birds was tubular in shape as mentioned by (Jarrar, 2009; Waleed, 2016). The changes in salt gland color from red color to dark red in duck which drink salt loaded water was noticed earlier by El-Gohary *et al.* (2013) they found that the color of salt stressed gland was dark red, while the gland color of fresh water ducks was pale red. The current study suggested that the cause of such alteration in gland color due to the shaft and increased of blood fallow toward the gland when the bird drink salty water. This suggestion depending on the study of Hossler and Olson (1990)(20) they found that there was Changes in blood flow to the salt gland of duck lived in osmotic loading (1% NaCl) food and water, this was adaptation strategy for hyper osmotic environment .

The present study revealed, that there were significant increased in the weight and size of gland in salt stressed

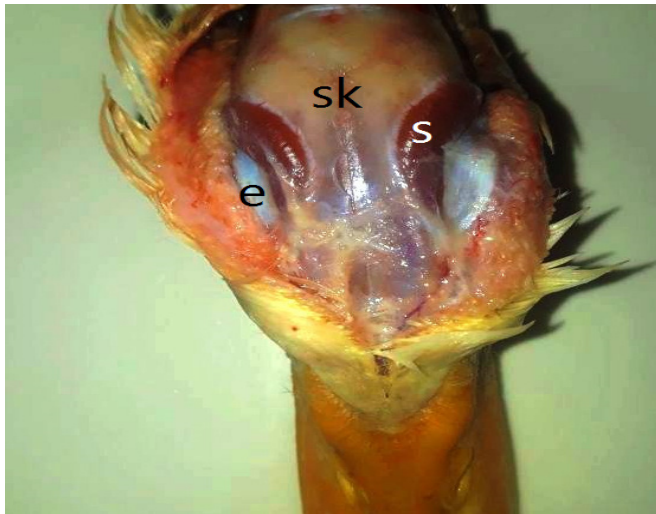
ducks. The weights of the salt glands of the salt-stressed group were obviously heavier and larger than those of the control ones. The same results are obtained from the study of Bokenes (1998) who found that the size and weight of salt gland in wild ducks which drink salty water was 50% larger than the salt glad of duck that drink fresh water. The same changes in weight and size of salt gland were reported in gull and white ibis was reported by (Burger and Gochfeld, 1984; Johnston and Bildstein, 1990) they found that there was marked hyper atrophy in gland in the birds which lives on salt water. In addition, the present findings are parallel with the observations of El-Gohary *et al.* (2013) they mentioned that the glands domestic ducks drink 1% NaCl solution for two weeks were heavier than the glands of fresh water ducks. The current study results was in agreement with results of Bennett and Hughes (2003) they noticed that when ducks switch from fresh water to saline habitat their salt glands of hypertrophy. Further support for the present morph metrical results comes from the study of Rubega and Oring (2004) and Woodin *et al.* (2008) they recorded that the mean mass and size of the redheads duck and other waterfowl salt glands were significantly varied depending on the salinity of water. The changes in the size of salt gland in ducks group, which drink salt water in this study proved the results of McArthur and Gorman (2009) they noticed marked decrease in the size of female eider ducks which do not drink salt water.

In the current work, in all examined groups, each gland histological specking consists of many branching tubules arranged in rows and separated by vascularized thin connective tissue. The parenchyma of each consisted of branched secretory tubules radiate out from a central canal, dialect connective tissue blood capillaries form the gland stroma. The secretory tubules are closely packed and arranged in parallel pattern. The secretory tubules are lining with a single layer of principal cuboidal cells forming a narrow lumen , the peripheral blind end of the tubules are lined round peripheral cells. This histological observed of the salt glands are in agreement with the results of many workers whom studied the histological structures of salt gland in many water birds species such as Hussein *et al.* (2006), Jarrar (2009), Mohammad (2012), Reshag *et al.* (2016) and Waleed (2016). They showed that the salt gland in different avian species is consists of several longitudinal lobules composed of secretory tubules radiating from central canals. The secretory tubules were lined by principle and basal cells.

In case of ducks which drink the salt water the salt gland responded to the salt stress .The histological response to tolerate the effect of high salt concentration were cellular proliferation and hypertrophy. This changes proved the results of Martin and Philpot (1973); Burgerand Gochfeld (1984). Muller and Hildebrandt (2003) and Reshag *et al.* (2016) found that the histological changes in the salt glands as response to salinity were blood vessels congestion hyperplasia, proliferation and differentiation in different subsets of cells. induces due to adaptive strategy of water birds. The results of this work disagree with results of (El-Gohary *et al.*, 2013) They mentioned that, The secretory tubules of the salt stressed ducks gland were numerous branched 4-6 times than in control duck salt gland, forming different arrangement pattern of tubules. The current study suggested that all this changes lead to increased the capacity of the gland to eliminate the excess salt from the birds body and control the fluid homeostasis, This agree with the finding



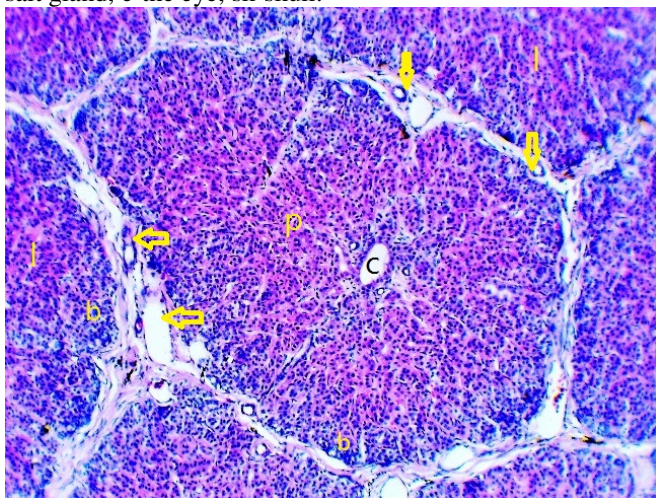
of (Marshall *et al.*, 1987); Barnes and Nudds, 1991) who reported that, the salt gland of water birds adapted to salt loaded water showed clear increases in size of the gland and the lining epithelium of the secretory tubules, the cells were modified to secreted salinated fluid and osmoregulation the body fluid salinity.



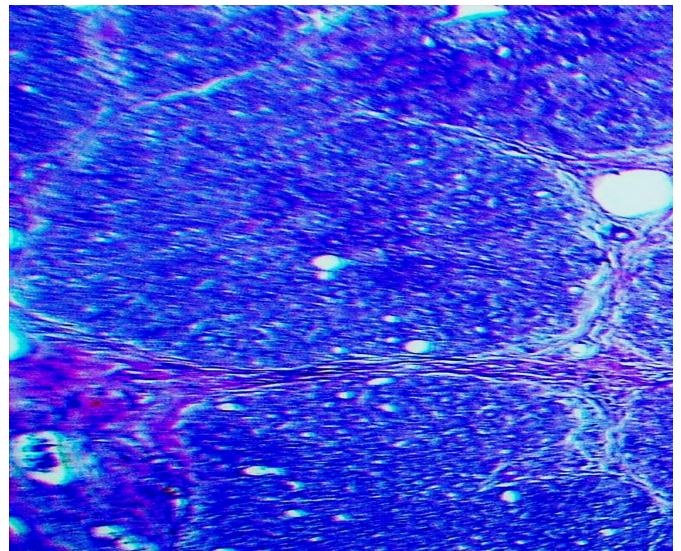
**Fig. 1 :** Macrograph of duck head (control group)shows: s- salt gland, e-the eye, sk-skull.



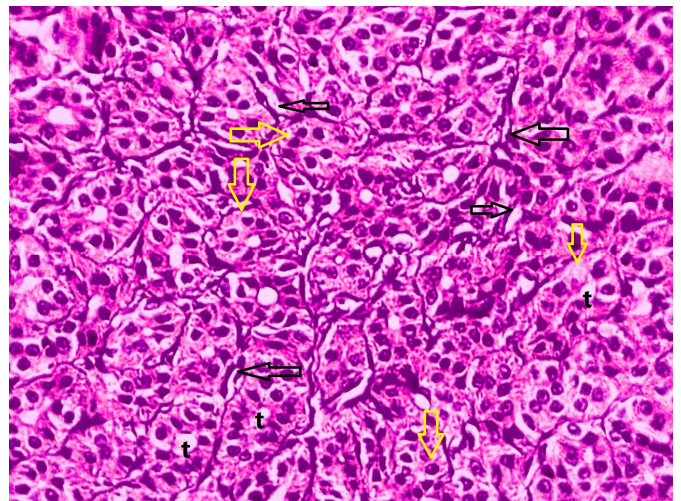
**Fig. 2 :** Macrograph of duck head (salt water group)shows: s- salt gland, e-the eye, sk-skull.



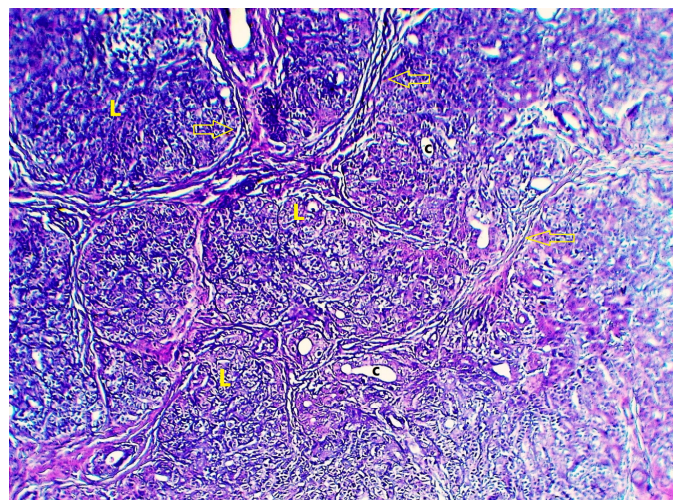
**Fig. 3 :** Transverse histological section salt gland(control) shows:, p- principle cells, b-basal cells c- central canal, connective tissue septa with blood vessels(arrows).(H&E stainX100).



**Fig. 4 :** Transverse histological section salt gland(salt water group) shows: changes in general structure of salt gland, polygonal lobule change to oval or rounded in shape ,dilated of central canal and decrease connective tissue with congestion blood vessels(arrows).(H&E stainX100).

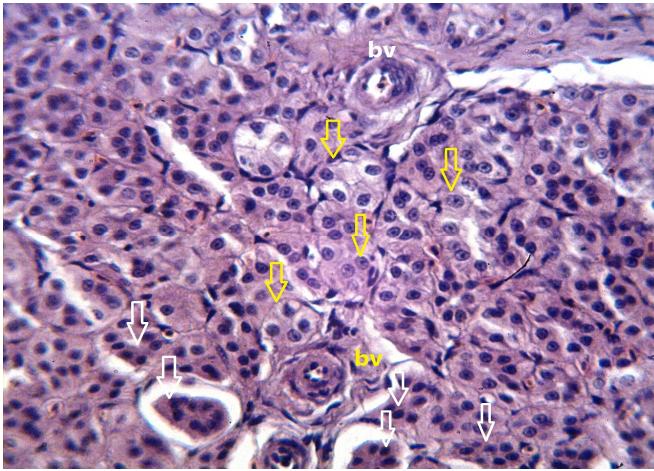


**Fig. 5 :** Transverse histological section salt gland(control)shows: t-secretary tubules, principle cells (yellow arrows), blood capillaries (black arrows).(H&E stainX400).

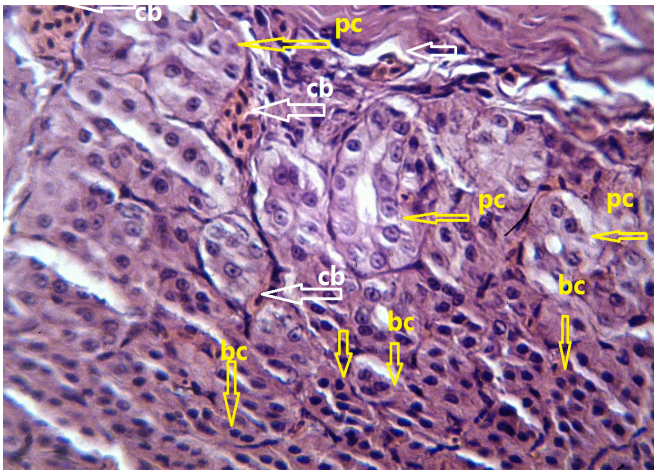


**Fig. 6 :** Transverse histological section salt gland(salt water group)shows: l- lobule, c- central canal, connective tissue septa (arrows). (H&E stainX40).





**Fig. 7 :** Transverse histological section salt gland (salt water group) shows: principle cells (yellow arrows), basal cells (white arrows), bv-blood vessels (white arrows). (H&E stain X400).



**Fig. 8 :** Transverse histological section salt gland (salt water group) shows: pc- principle cells, bc- basal cells, cb- congested blood vessels. (H&E stain X400).

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