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## Histochemical study of adrenal gland of one humped camel (*Camelus dromedarius*)

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### Abstract

The investigation was carried out on adrenal glands of 6 recently died adult camels. The adrenals were situated at the some distance from cranial aspect of the kidney embedded in perirenal fat. The adrenals were divided in stroma and parenchyma. Stroma consisted of capsule and trabeculae. Parenchyma composed of the cortex and medulla. Intense PAS positive reaction was seen in the capsule, trabeculae and zona glomerulosa. All zones of adrenal gland showed negative reaction for glycogen in PAS with saliva stain. Mucosubstances showed strong reaction in capsule, trabeculae, zona glomerulosa and outer medullary area.

**Keywords:** Adrenal gland, PAS, one humped camel

### Introduction

The adrenal gland is one of the most important organ because it plays a significant role in the body activities and essential for the maintenance of whole life. It regulates many physiological functions both in foetal and postnatal life (Hill, 2007) [3]. The adrenal gland is an indispensable organ that takes part in the formation of hypothalamic pituitary adrenal axis (HPA) termed as body's stress system. Moreover the HPA, mainly controls the levels of cortisol and other important stress related hormones (Hu and Funder, 2006; Pippal and Fuller, 2008) [4, 10].

The adrenal gland is closely associated with metabolic activity related to farm animal productivity. Mineralocorticoids, produced in the zona glomerulosa (ZG), contribute to the homeostasis of electrolytes and the volume of circulating fluid, mainly by the stimulation of resorption of water, Na<sup>+</sup> and K<sup>+</sup> excretion. Glucocorticoids synthesized in the zona fasciculata (ZF) regulate primarily the metabolism of carbohydrates but also the metabolism of proteins and lipids. Their activities consist of glucose sparing, increased glucose production, decreased lipogenesis and support lipolysis. They also suppress both the inflammatory and immune reactivity, especially during a stress reaction, thus reducing resistance of animals to bacterial, viral and fungal infections.

The anatomical structures of adrenal gland is even though studied in few countries but their studies are important because it plays a pivotal role in adaptation of camels located at different geographical locations and in different extreme environments. Present study is aimed to study the histo-chemical details of adrenal gland of dromedary camels of Rajasthan.

### Materials and Methods

The adrenal glands were collected from 6 recently died adult camels from VCC, CVAS, Bikaner, Rajasthan. The recommendations of the ethical committee were followed for present research and investigation of the organs were carried out in the Department of Veterinary Anatomy, CVAS, Bikaner. For light microscopic studies, the samples were fixed in 10% formalin for routine staining and in bouin's fluid for special staining for 48 to 72 hours. The tissues were proceeded by Alcohol-xylene method using cedar wood oil (Luna, 1968) [7], paraffin blocks were prepared, numbered and stored at 4°C in refrigerator. Sections of 5-6 µm thickness were obtained then placed on albumenized slides and kept overnight in hot air oven at 36°C and finally stained for general histomorphological and histochemical observations.

## Results and Discussion

### Mc Manus's reaction for glycogen

Intense PAS positive reaction was observed in the capsule showing the presence of glycogen (Fig 6). The findings of present study were in close harmony with Roy and Saigal (1987)<sup>[11]</sup> in sheep, Olukole *et al.* (2016)<sup>[9]</sup> in rat, Kour *et al.* (2017)<sup>[6]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in Iraqi goat. The findings of present study were in partial harmony with Nicander (1957)<sup>[8]</sup> in dog who observed the Small amounts of glycogen were found in the capsule.

The trabeculae were also positive for the glycogen showed intense PAS positive reaction (Fig. 6 and 7). The findings were in agreement with the reports of Abubakar (2015)<sup>[1]</sup> in goat and Olukole *et al.* (2016)<sup>[9]</sup> in rat.

The zona glomerulosa showed strong PAS positive reaction (Fig. 6 and 7). The findings were in agreement with the reports of Idelmann (1978)<sup>[5]</sup> in ox, Olukole *et al.* (2016)<sup>[9]</sup> in rat, Kour *et al.* (2017)<sup>[6]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in Iraqi goat.

Zona fasciculata layer showed moderate PAS positive reaction for the presence of glucocorticoids (Fig. 7 and 8). The findings were in agreement with the reports of Olukole *et al.* (2016)<sup>[9]</sup> in rat, Kour *et al.* (2017)<sup>[6]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in Iraqi goat.

Zona reticularis showed weak PAS positive reaction (Fig 8 and 9). The findings of present study were in close harmony with Kour *et al.* (2017)<sup>[6]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in Iraqi goat. The findings of present study were in partial harmony with Nicander (1957)<sup>[8]</sup> in dog adrenals of adult dogs contained very little glycogen and Olukole *et al.* (2016)<sup>[9]</sup> in rat observed the adrenal capsule, the entire adrenal cortex and adrenal medulla were PAS-positive.

There was weak PAS positive reaction for glycogen in the inner and moderate positive reaction in outer medulla (Fig 9). The findings of present study were in close harmony Olukole *et al.* (2016)<sup>[9]</sup> in rat. The findings of present study were in partial harmony with Nicander (1957)<sup>[8]</sup> in dog, and Shehan *et al.* (2017)<sup>[12]</sup> in Iraqi goat.

### Mc Manus's reaction with saliva for glycogen

The capsule, trabeculae and zona glomerulosa showed PAS negative reaction for Mc Manus PAS with saliva stain (Fig. 6 and 7) also confirmed the observation of Roy and Saigal (1987)<sup>[11]</sup> in sheep, Kour *et al.* (2017)<sup>[6]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in Iraqi goat. The findings of present study were in partial harmony with Nicander (1957)<sup>[8]</sup> in dog observed the Small amounts of glycogen were found in the capsule.

After treatment with saliva, capsule, trabeculae and zona

glomerulosa showed negative reaction for carbohydrates in Mc Manus PAS with saliva stain because glycogen digested by human saliva.

Zona fasciculata, zona reticularis and medulla also showed PAS negative reaction for carbohydrates in Mc Manus PAS with saliva stain (Fig. 7, 8 and 9) also favoured the observation, Kour *et al.* (2017)<sup>[6]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in Iraqi goat. The findings of present study were in partial harmony with Nicander (1957)<sup>[8]</sup> in dog adrenals of adult dogs contained very little glycogen.

### PAS-Alcian Blue reaction for mucosubstances

The mucosubstances showed intense positive reaction in the capsule area in PAS-Alcian blue pH 1.0 (Fig. 1) as well as in pH 2.5 (Fig 3 and 4). In congruence with the observations of Hakeem *et al.* (1993)<sup>[2]</sup> in goat, Abubakar (2015)<sup>[1]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in the Iraqi goat.

Trabeculae were showed intense positive reaction for PAS-Alcian blue pH 1.0 as well as in pH 2.5.

There were presence of mucosubstances in the zona glomerulosa distinguished by moderate positive reaction for PAS Alcian blue pH 1.0 (Fig 1 and 2) and 2.5 (Fig 3 and 4) stain also favoured the observation of Abubakar (2015)<sup>[1]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in the Iraqi goat. The findings of present study were in partial harmony with Hakeem *et al.* (1993)<sup>[2]</sup> in goat Intense PAS positive reaction was in capsular fibres, entered in the zona glomerulosa and zona fasciculata.

Zona fasciculata area possessed mucosubstances in moderate amount (Fig. 3). There were little amount of mucosubstances in inner medullary area showing PAS Alcian blue reaction at pH 2.5. The findings of present study were in partial harmony with Hakeem *et al.* (1993)<sup>[2]</sup> in goat Intense PAS positive reaction was in capsular fibres, entered in the zona glomerulosa and zona fasciculata.

There were weak amount of mucosubstances in inner medullary area showing PAS Alcian blue reaction at pH 2.5 (Fig 5). The findings of present study were in close harmony with Hakeem *et al.* (1993)<sup>[2]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in the Iraqi goat.

Intense PAS Alcian blue reaction for mucosubstances was seen in outer medullary area at pH 2.5 (Fig 5). The findings of present study were in close harmony with Hakeem *et al.* (1993)<sup>[2]</sup> in goat and Shehan *et al.* (2017)<sup>[12]</sup> in the Iraqi goat.

The outer medulla and the inner medulla were clearly distinguished in the PAS Alcian blue stain in which it stained sharply (Fig 5).

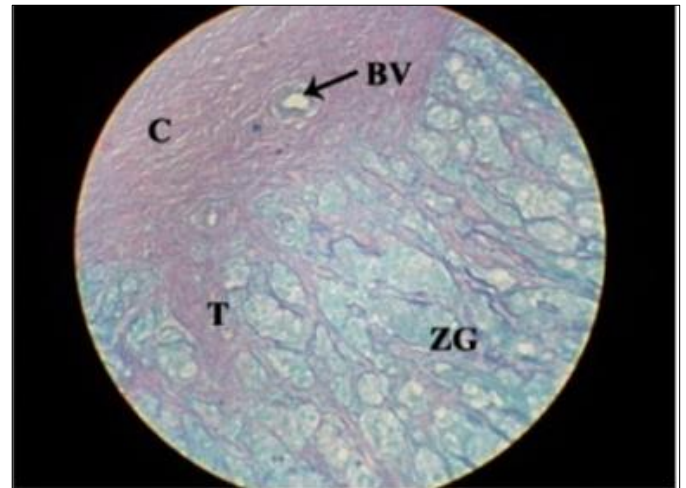
### PAS-Alcian Blue reaction for mucosubstances

1.	Mc Manus's method with saliva (PAS)	For glycogen	Singh and Sulochana, 1997 <sup>[13]</sup> .
2.	Mc Manus's method-PAS	For glycogen.	Singh and Sulochana, 1997 <sup>[13]</sup> .
3.	PAS- Alcian blue method pH 1.0	For Mucosubstances	Luna, 1968 <sup>[7]</sup> .
4.	PAS- Alcian blue method pH 2.5	For Mucosubstances	Luna, 1968 <sup>[7]</sup> .

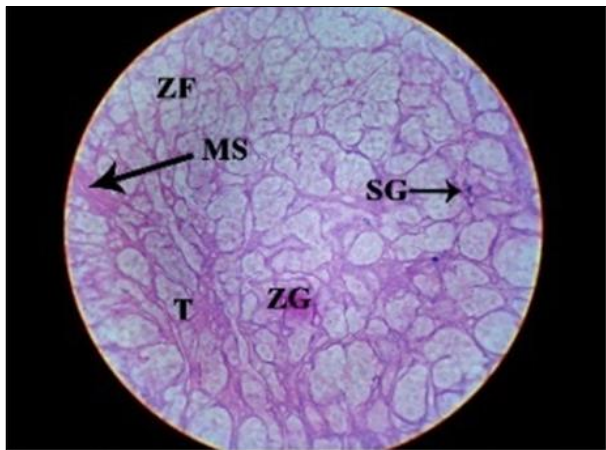




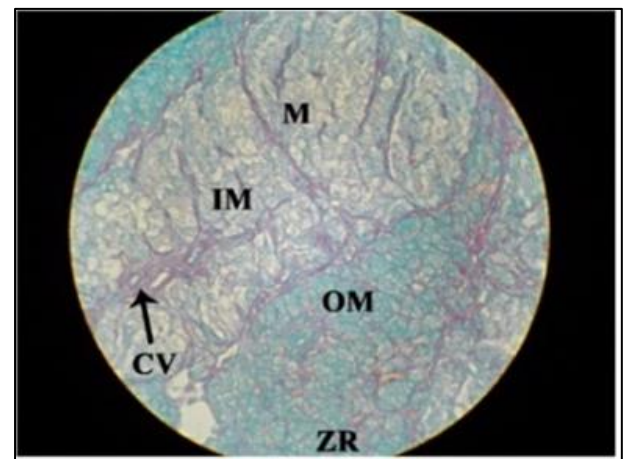
**Fig 1:** Photomicrograph of adrenal gland of camel showing the positive PAS Alcian blue reaction trabeculae and cortex zones. T- Trabeculae, ZG- Zona Glomerulosa, SG – Sulphate granules, MS- Mucosubstance (PAS Alcian blue method for mucosubstance PIT 1.0, 400X)



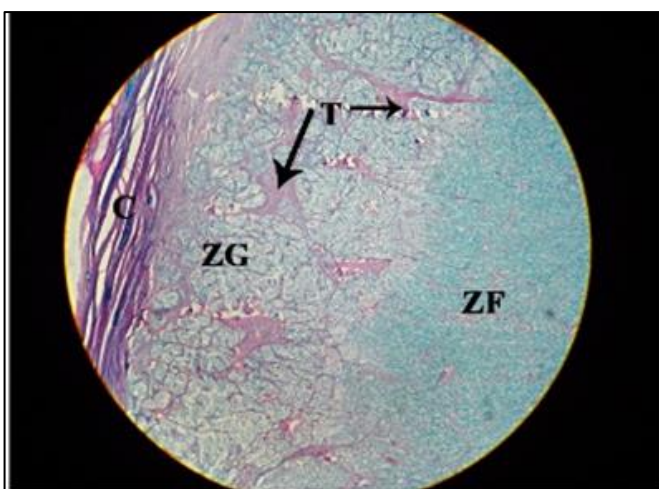
**Fig 4:** Photomicrograph of adrenal gland of camel showing the positive PAS Alcian blue reaction in capsule and zona glomerulosa, C- Capsule, BV- Blood vessels, ZG- Zona Glomerulosa, T- Trabeculae. (PAS Alcian blue method for mocosubstance pH 2.5, 400X)



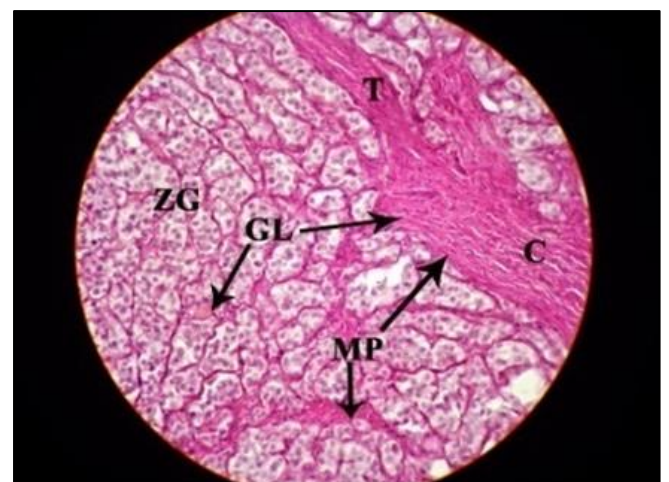
**Fig 2:** Photomicrograph of adrenal gland of camel showing the positive PAS Alcian blue reaction in cortex zones. T- Trabeculae, ZG- Zona Glomerulosa, SG- Sulphated Granules, MS- Mucosubstance, ZF- Zona Fascicular. (PAS Alcian blue method for mucosubstance pH 1, 400X)



**Fig 5:** Photomicrograph of adrenal gland of camel showing the positive PAS Alcian blue reaction in the cortex and medulla zones. ZR –Zona Reticularis, OM- Outer Medulla, IM- Inner Medulla, CV- Central Veins (PAS Alcian blue method for mucosubstance pH 2.5, 400X)

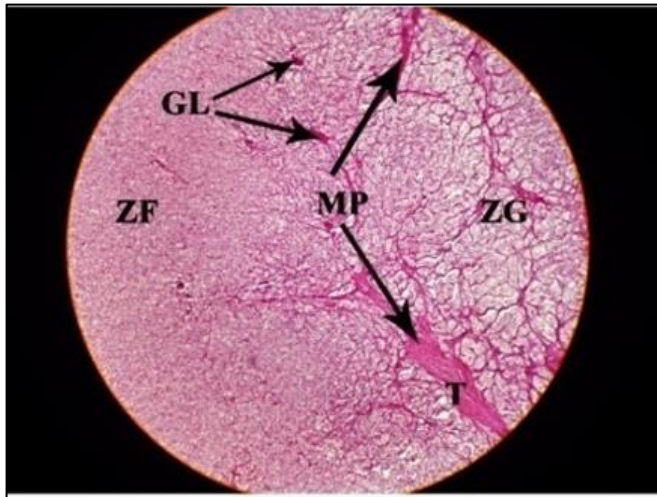


**Fig 3:** Photomicrograph of adrenal gland of camel showing the positive PAS Alcian blue reaction in cortex zones. C- Capsule, ZG- Zona Glmoerulosa, T- Trabeculae, ZF- Zona Fasciculata. (Pas Alcian blue method for mucosubstance pH 2.5, 100X)

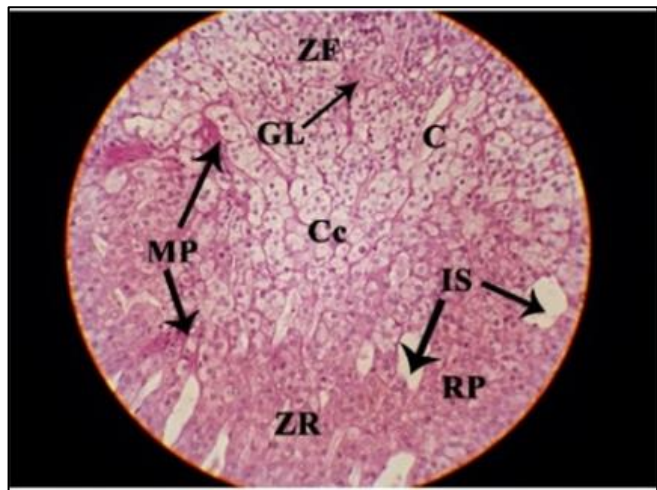


**Fig 6:** Photomicrograph of adrenal gland of camel showing positive PAS reaction in capsule zona glomerulosa. C- Capsule, ZG- Zona Glomerulosa MP- Mucopolysaccharides, GL- Glycogen, T- Trabeculae. (Mc Manus method PAS stain, 400X)

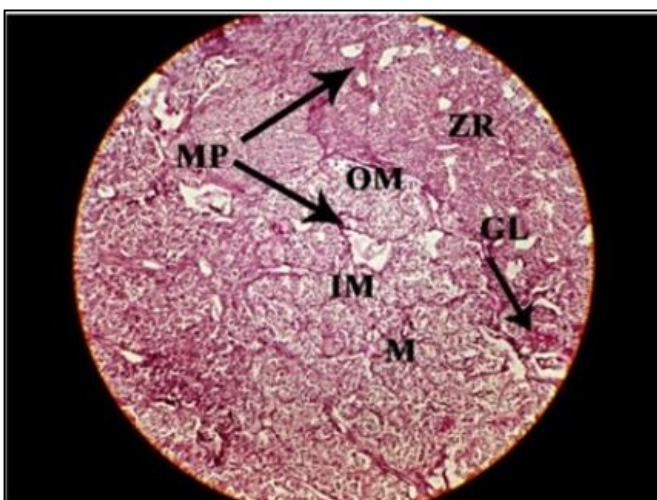




**Fig 7:** Photomicrograph of adrenal gland of camel showing positive PAS reaction in cortex zone. ZG- Zona Glomerulosa, ZF- Zona Fasciculata, MP- Mucopolysaccharides, GL- Glycogen. T- Trabeculae. (Mc Manus method PAS stain, 100X)



**Fig 8:** Photomicrograph of adrenal gland of camel showing positive PAS reaction in cortex zone. ZF- Zona Fasciculata, ZR- Zona Reticularis, MP- Mucopolysaccharides, GL- Glycogen, RP- Reticular pattern, IS- Irregular sinusoid, Cc-Columnar cells. (Mc Manus method PAS stain, 400X)



**Fig 9:** Photomicrograph of adrenal gland of camel showing positive PAS reaction in cortex and medulla zones. ZR- Zona Reticularis, M- Medulla, MP- Mucopolysaccharides, OM- Outer Medulla, IM- Inner Medulla, GL- Glycogen (Mc Manus method PAS stain 100X)

**Table 1:** Histochemical reactions exhibited by the different components of the adrenal glands of One Humped Camel

Histochemical reaction	Location in the adrenal gland	Intensity
PAS	Capsule	+++
	Trabeculae	+++
	Cortex	++
	Zona glomerulosa	+++
	Zona fasciculata	++
	Zona reticularis	+
	Medulla	+
	Inner medulla	+
	Outer medulla	++
Alcian blue pH 1.0	Capsule	+++
	Trabeculae	+++
	Cortex	++
	Zona glomerulosa	++
	Zona fasciculata	++
	Zona reticularis	+
	Medulla	++
	Inner medulla	+
	Outer medulla	+++
Alcian blue pH 2.5	Capsule	+++
	Trabeculae	+++
	Cortex	++
	Zona glomerulosa	++
	Zona fasciculata	++
	Zona reticularis	+
	Medulla	++
	Inner medulla	+
	Outer medulla	+++
Central vein	+	

Note +++ = Intense or Strong, ++ = Moderate, + = Weak and - = Negative

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