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RESEARCH ARTICLE

Gross Morphological and Ontological Studies on Pituitary of Camel (Camelus dromedarius)

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ABSTRACT

Thirty normal male and female camels (*Camelus dromedarius*) with three age groups (2-4, 5-10 and 11-onward years) were selected for pituitary gross morphological and ontological studies. Body weight and pituitary were recorded; Mean body weight of camels and mean relative pituitary gland weight as a percent of body weight were calculated. In camels of age greater than 11 years, body weight significantly correlated with relative pituitary weight (r=-0.754*), overall correlation of female body weight and pituitary weight irrespective of their age was highly significant (r=0.888**). Whereas, the body weight of female camel between 5-10 years of age reflected a significant correlation with relative pituitary weight (r=0.882*). The cells of adenohypophysis were arranged in the cluster and formed the pars distalis and pars intermedia. Overall acidophil in the pituitary of camels were higher than chromophobes and basophils while basophils were the lowest in number as compared to other two types of cells. It is concluded that body weight of camel is suggestive of its correlation with pituitary weight, sex and age.

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INTRODUCTION

Camel mostly lived in semiarid as well as arid area of Pakistan where availability of feed as well as water is poor. In camel like in human and all other animals is known to be not only regulating most of the important physiological functions pertaining to growth, production and reproduction. Almost similar observations were recorded by Matinez-Lage (2011) and Morieb and Hoehn (2012). Some authors did look into hypothalamus and adrenal glands of camel (Ye *et al.*, 2014, 2015, 2017). Very recently Wenling *et al.* (2018) studied the pituitary gland from camel and obtained data pertaining to biology, anatomical and histological observations.

Pituitary gland of both male and female camel was seen elongated and overall a dense irregular connective tissue was observed surrounding the pituitary gland of camel (Jaspal *et al.*, 2011). They also depicted that pituitary weight increased significantly as the animals aged and male camel had much heavier pituitary as compared to female. *Bacterian dromedarius* differentiation of hypothalamo-pituitary system is completed when 2-3 years old and the pituitary weight is maximal at six year of age (Bezrukov, 1970). However, Verzar (1966) reported that pituitary gland weight did not

decline with age despite a reduction in the number of GH and PRL cell. Dorst, (1968) and Li *et al.*, (2008) reported related work on pituitary from animals like in buffalo, sheep, pigs, and in dogs.

Keeping in view the lack of details and research references on differential cell count of adenohypophysis and correlation of body with relative weight of pituitary of camel. The present research work was under taken to depict the correlation of body weight with relative pituitary gland weight as a percent of body w eight of male and female camel with advancement of age.

MATERIALS AND METHODS

Thirty male and female camels (2-4, 5-10 and 11-onwarsd years) irrespective of any physiological condition were collected from slaughter house at Kot Kamboh, Lahore and Zanith Associates (Pvt.), Lahore. Animals were divided into six groups of five animals each. The age of animals was determined from the history provided by the camel owners and by teeth examination (Wilson, 1984; Schwartz and Doili, 1992). The Pituitary glands were extirpated within three to four hours after the slaughtering to avoid the diffusion of and loss of hormones.

The pituitary glands were recorded for their weight (Mohammad, 1982). Weight (g) was recorded by using electrical balance. The body weights of all experimental camels were calculated after recording certain body dimensions using a plastic tape according to the following equation as described by Kohler-Rollefson *et al.* (2001):

Body Weight $(Kg) = 50 \times SH \times CG \times HG$

Where,

SH (Shoulder Height)=Height of the shoulder (in meters). CG (Chest Girth)=Distance (in meters) around the camel's chest measured in front of the hump and behind the front legs and chest pad.

HG (Hump Girth)=Distance (in meters) around the camel's body measured at its widest point from the top of the hump around the belly.

Pituitary gland weight as a percent of body weight ($\%BW\times10^{-3}$) of camel (*Camelus dromedarius*) was calculated.

Samples for light microscopy were fixed, dehydrated, cleared embedded in paraffin, section and stained with special stained. The staining procedure for pituitary cells has been reported by Jaspal *et al.* (2011).

Adenohypophyseal cell counts for acidophils, basophils and chromophobes were performed on male and female camel within each age group. The counts were made at X1000 with ocular micrometer. Nine (9) fields were selected from each slide counting each from left, right and middle.

Statistical analysis: Data was analyzed statistically by two-way analysis of variance. Comparison and differences between young and old as well as male and female were analyzed. Duncan Multiple Range test was applied to see the difference between mean values. A correlation coefficient, *=significant (p<0.05); **=Highly significant (p<0.01) was also used see the relationship of the body weight and relative pituitary gland weight as a percent body weight (Steel *et al.*, 1997) with age and gender.

RESULTS

The mean body weight of camels (*Camelus dromedarius*) according to their age and sex is represented in Table 1. Mean differences in different age groups and sex were significant ($P \le 0.01$). Body weight of camel did increase with their age. Male camel showed significantly high body weight as compared to female. Camel at 11-

onward years irrespective their sex showed significantly high body weight. In camels of age greater than 11 years, body weight correlated with relative pituitary weight. The mean relative pituitary gland weight as a percent of body weight in male and female camel (*Camelus dromedarius*) at different age groups was also calculated (Table 2). Camels showed a significant increase ($P \le 0.05$) in relative pituitary gland weight with their age irrespective of gender.

The overall correlation of female body weight and pituitary weight irrespective of their age was calculated. The overall body weight significantly correlated with relative weight (r=0.888**). The body weight of female camel between 5-10 years of age reflected a correlation with relative pituitary weight. During 5-10 years female body weight correlated with relative weight (r=0.882*).

Morphologically acidophil were found to be round, located throughout the adenohypophysis. Basophils were large, round/irregular cells in the center of adenohypophysis. Whereas chromophobe were of different shapes (Fig. 1). Female camels have significantly high cell count as compared to male (Fig. 2). Overall mean cell count of acidophils was significantly high as compared to chromophobe and basophils (Table 2). Age×cell type interaction observed acidophils to be significantly high at age 5-10 years and 11- onward years old camels. Lowest cell counts of basophils were found to be in young as well as in old camel (Fig. 3). Overall mean acidophils count was significantly high and lowest count of basophils was observed in male and female camel (Fig. 4).

Table 1: Mean body weight (Kg±SE) of camel (Camelus dromedarius) at different age groups and gender

Gender	2-4 Years	5-10 Years	II – Onward	Overall Mean
			Years	
Male	373.10±31.44	644.00±20.63	744.70±15.30	587.27±31.93 ^A
Female	341.40±24.59	607.00±15.28	675.70±11.13	541.60±28.58 ^B
Overall	357.25±19.76 ^C	625.85±13.16 ^B	710.20±12.14 ^A	564.43±21.45
Mean				

ABC, similar alphabets on overall means in row do not differ significantly at $P \le 0.01$.

Table 2: Mean relative pituitary gland weight as a percent of body weight (%BW×10³) of camel (*Camelus dromedarius*) at different age groups and gender

gi oups and gender							
Gender	2-4 Years	5-10 Years	II – Onward	Overall Mean			
			Years				
Male	0.20±0.010	0.26±0.006	0.26±0.002	0.24±0.007			
Female	0.21±0.006	0.27±0.013	0.28±0.003	0.25±0.007			
Overall Mean	0.20±0.005 ^C	0.26±0.007 ^E	³ 0.27±0.002 ^A	0.25±0.005			
ABC, similar	alphabets	on overall	means in row	do not differ			
significantly at P<0.05.							

Table 3: Mean adenohypophyseal cell count (number ±SE) of male and female camel (Camelus dromedarius) at different age groups

Parameters	2-4 Years		5-10 Years		I I-onwards Years		OII M
	Male	Female	Male	Female	Male	Female	- Overall Mean
Acidophils	27.78	32.19	30.81	30.85	31.96	29.7	30.55
	±0.54 ^d	±0.65 ^a	±0.45 ^b	±0.48 ^b	±0.51 ^a	±0.39°	±0.24 ^A
Basophils	15.41	21.37	23.11	23.33	23.3	21.93	21.41
	±0.25 ^k	±0.20 ^j	±0.41 ⁱ	±0.37hi	±0.37 ^{hi}	±0.23 ^j	±0.25 ^C
Chromophobes	21.48	24.3	26.04	25.63	26.3	25.07	24.8
	±0.28 ^j	±0.27gh	±0.338ef	±0.26 ^{ef}	±0.24e	±0.32 ^{fg}	±0.17 ^B
Overall Mean	21.56	25.95	26.65	26.6	27.19	25.57	25.59
	±0.61 ^C	±0.56 ^B	±0.43 ^A	±0.40 ^A	±0.46 ^A	±0.40 ^B	±0.21

a-j, similar alphabets on means do not differ significantly at $P \le 0.05$. ABC, similar alphabets on overall means in row do not differ significantly at $P \le 0.01$. ABC, similar alphabets on overall means in column do not differ significantly at $P \le 0.01$.

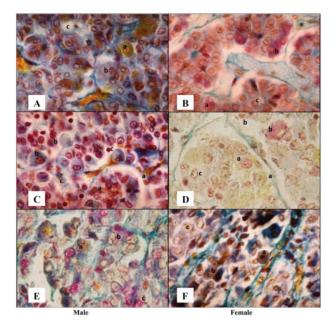


Fig. 1: Photomicrograph of Camel (*Camelus dromedarius*) adenohypophysis. The sections show parts of several cords of cell containing, (a) acidophils (orange-yellow / yellow); (b) basophils (magenta red/radish-purple) and (c) chromophobes (unstained or very low yellow). Fine connective tissue (green) separates the cords. A 4µm sections stained with phosphotungstic acid haematoxylin-aldehyde fuchsin-light green-orange G combination of stains. (A) 2-4 years male (B) 2-4 years female (C) 5-10 years male (D) 5-10 years female (E) 11-onward years male (F) 11-onward years female.

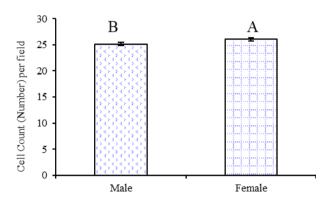


Fig. 2: Overall mean adenohypophyseal cell count (number±SE) of male and female camel (*Camelus dromedarius*) irrespective of their age and cell types AB, similar alphabets do not differ significantly at P≤0.01.

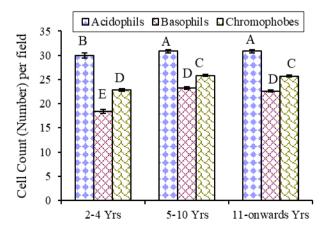


Fig. 3: Overall mean adenohypophyseal cell count (number \pm SE) of camel (*Camelus dromedarius*) at different age groups irrespective of their gender A-E, similar alphabets do not differ significantly at P \leq 0.01.

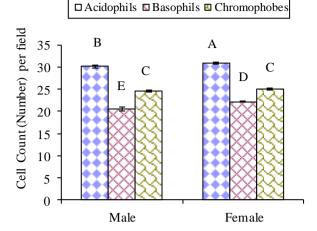


Fig. 4: Overall mean adenohypophyseal cell count (number±SE) of male and female camel (*Camelus dromedarius*) irrespective of their age groups A-E. similar alphabets do not differ significantly at P<0.05.

DISCUSSION

Adenohypophysis is a critical component of the neuroendocrine system in all vertebrates which is essential for the maintenance of homeostasis, metabolism, reproduction, growth and lactation. The synthesis and secretion of trophic hormones from hypothalamus for distinct endocrine cell types of the pituitary gland are very well recognize by for different physiological system as well as for positive and negative feedback loops from peripheral region.

Pituitary weight and morphological characters of the hypophysis could be greatly influenced by age and sex. The variations in mean body weights of camel arise from factors like age and gender. Relatively higher figures of mean body weight have been recorded for Bikanarie, and Qatar camels in Arabian Penisula (Wilson 1984). The present study indicate that body weight of camel did increase with their age. Male camel showed significantly high body weight as compared to female. Camel at 11-onward years irrespective their sex showed significantly high body weight. These variations closely agree with findings of Wilson (1984).

In camels of age greater than 11 years, body weight correlated with relative pituitary weight (r=-0.754*). A significant increase (P≤0.05) in relative pituitary gland weight with their age irrespective of gender reveals that increase in pituitary gland weight during old age may be for normal physiological phenomenon as body mass increases, pituitary gland weight also increases proportionally. A significantly high difference in body weight of male camel as compared to female, may require massive pituitary gland. An increase in pituitary gland weight during old age may be due to the normal physiological phenomenon as body mass increases, pituitary gland weight also increases proportionally. Higher mean weights of pituitary glands from adult male animals than those of heifers has also been reported by Namboothripad and Luktuke (1978). Pousty (1977) revealed that male camel has massive pituitary gland as compared to female. Shirasawa and Yoshimara (1982) reported that body weight and pituitary weight increase with advancing age as a result of multiplication of pituitary cells (mitotic division) from the undifferentiated to the mature cell types to various hormone producing cells related to the repeated growth, reproduction and production. They concluded that the mitotic rates of the six cell types are age dependent. Nagamalleswari *et al.* (2004) also revealed an increasing tendency in the biometrical values (length, width, weight and thickness) of goat pituitary with the advancement of age.

It is found that pituitary of camel rests upon the hypophyseal fossa of the sphenoid bone in the center of middle cranial fossa attaching to the hypothalamus by hypophysis stalk. Hegazy *et al.* (2004) noted that pituitary gland of female camel is subdivided into adenohypophysis and neurohypophysis. The adenohypophysis comprises pars distalis, pars tuberalis and pars intermedia, and inter glandular cleft was separating the pars distalis from pars intermedia. Ye *et al.* (2018) investigated that cells of adenohypophysis are arranged in clusters in order to formed the pars intermedia and pars distalis.

This is a normal physiological phenomenon, as there is an increase in body mass there will be more need of hormone to engage and fulfill the body needs. These results are in comparison to buffaloes where it was observed by Muhammad and Khan (1984) that increase in pituitary weight was related to increase in weight gain of animal.

It has also been concluded by Gilmore *et al.* (1941) that hypophyseal weight in bovine increases up to the age of seven years. Pousty (1977) studied that one humped Iranian female camel were with heavier pituitary glands than male.

Heavier pituitary glands were observed in a lean sheep line as compared to with a heavy body weight (Fleming et al., 1997). Heavier pituitary glands have also been reported in swine lineages selected for low fat and they have attributed these differences to an increase in cell number rather than cell size. Bassett et al. (1951) reported that pituitary glands of dairy cows (6-10 years) were heavier as compared to those of steers (4-5 years) and these changes may have been attributed due to its their functional atrophy. In addition, Ganguli and Yadava (1975) also recorded that in 2-3 years' male buffalo, the mean weight of pituitary gland was 0.55g and in10-13 years' older females the gland was 0.88g. They attributed the increase in pituitary weight to the increase in length, thickness and breadth of the pituitary gland with the age of animal. Pousty (1977) reported that in camel (male and female) pituitary gland weight and volume increased with their age. Average weight ranged 1.0-2.7 g and volume 1.0-2.9 cm³ in camel irrespective their gender.

Jubb and Kenedy (1970) reported that in nulliparous domestic animals the size of pituitary gland was twice to its normal volume and the sella correspondingly enlarged. The anterior pituitary of yak constituted 67% of total weight (0.21 g) as well as total volume (3cm) of the gland; the rest (33%) was posterior pituitary and infundibulum stalk (Nath and Bhattacharya, 2003). Pituitary glands from healthy non-descriptive female goats (*Capra hircus*) of day-old to 12-month-old during prepubertal period did show an increasing tendency in the biometrical values in terms of length, width, weight and thickness of pituitary with the advancement of their age (Nagamalleswari *et al.*, 2004). Ye *et al.* (2018) reported

that hypophysis of Chinese camel was about 1.54 g in weight and its AGW/BW was calculated as 0.0045 g/kg, therefore it indicates a similarity with herbivores that had similar body size to Bactrian camel.

The cell of pituitary gland does play a major role in organizing and regulating vital physical functions as well as general wellbeing. So pituitary is referred as the body's 'master gland'. Moreover, it does regulate other hormones and secretion to regulates the activity of most other vital organs. Martinez-Lage, (2011) and Marieb and Hoehn, (2012) also indicated that pituitary cell secretes a variety of hormones into the circulation to act as messengers to transmit physiological messages from the pituitary to different cells present for distant regulating their activity.

The increasing total number of pituitary cells with advancing age may be associated with high concentration of pituitary hormones. Fat-Halla and Ismail (1980) observed variations in the cell number, cell size, cell nucleus size and distribution of cell types in the anterior pituitary of camel (*Camelus dromedarius*) at different ages.

Morphologically acidophil were found to be round, located throughout the adenohypophysis. Basophils were large, round/irregular cells in the center of adenohypophysis. Whereas chromophobe were of different shapes.

Male and female camel at 5-10 years of age and male camels older than this have a significant high cell count. Overall mean cell count of acidophil was significantly high as compared to chromophobe and basophils. Acidophils were significantly high at age 5-10 years and 11- onward years old camels. Variation in cell number. size and distribution of cell types in adenohypophysis of camel occur at different age groups (Fat-Halla and Ismail, 1980). Lowest cell counts of basophils were found to be in young camel. Overall mean acidophils count was significantly high in female and lowest count of basophils was observed in male camel. The highest acidophils count in female rather than male camel could be due to the normal physiological phenomenon to meet prolactin level. The presence of higher acidophil count reinforces the findings of Biswal et al (1966) in the Glandula Pituitaria of bull and bullock and Prasad and Singh (1980) in nonpregnant Indian buffalo. Increase in acidophil number may be due the fact for more need of growth hormone for body development and prolactin specifically mammary parenchymal growth in female with increase in age, chromophobe number increased. This may be due to the phenomenon of apoptosis or may be due to less need of some hormones at different physiological stages. Acidophil number increased in both male and female but chromophobes number was least among all the categories of cell types. Similar results were depicted in nonpregnant Indian buffalo (Bubalus bubalis) by Prasad and Singh (1980).

Conclusions: The pituitary weight of male camel as compare to female did show that as the animal grow older there seems a low mitosis followed by hypertrophy. This included by the change in the correlation of body weight and pituitary weight in terms of its growth indicating in width, length and circumference. This information will provide some insight into the important cellular and subcellular data in camel based on age and gender. The

study will provide credible, gross morphological and ontological data of pituitary gland of camel of different age groups which will contribute to the future research for the relationship of pituitary weight to body weight and differential cell count in this organ.

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Authors contribution: SASJ collected the pituitary for gross morphology, staining and morphometric study. ZUR execute and conducted the research. AMC helped in the write-up and preparation of the manuscript.

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