

Research article

Histological and morphometric study of the skin appendages of local breed sheep *ovis ovis*

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Abstract

The aim of this study is provide histological and morphometric data information to the skin appendages of the local breed sheep regarding to the individual variation in different animal species to use in another sciences. The study carried out on ten healthy ram skin and the samples were taken during autumn season from fourth different anatomical sites (ventral ear surfaces, neck, middle dorsum, tail.). The study revealed that there were a number of skin appendages including(wool follicles and skin glands Sebaceous and sweat glands), two types of wool follicles were observed in the dermis (reticular layer) of the skin of local breed sheep in the form of primary and secondary wool follicles, the wool follicles in sheep were considered as a compound type in which there were more than one wool fiber emerged from one pore on the skin surface. The follicles was arranged as groups consist mainly of three follicles (Trio), in addition to (Dio) and (Mono) arrangement, in this study we noticed the greater diameter of the primary and secondary wool follicles in the neck region and the deeper primary wool follicles present in neck region ,while the deeper secondary wool follicles present in middle dorsum. In respect to the sebaceous glands, the present results indicate that they attached with the both types of wool follicles in which they opened at the upper part of the wool follicles via short duct the larger sebaceous gland found in neck region all the sweat glands of the skin of sheep were of Apocrine type, these glands which have large secretory parts and narrow duct were accompanied the primary wool follicle but the bigger sweat gland present in middle dorsum.

Keywords: Sheep, Wool follicles, Sebaceous glands, Sweat glands.

Introduction

The skin of the mammals consist of two layers ,the surface layer called (epidermis) or corneum and underlying layer called (dermis) or (corium) and it involved several epidermal derivatives (skin appedages) their arrangement and distribution differ from region to another, the most important of them are (hair follicles ,sebaceous glands, sweat glands) (1).The hair shaft is involved three layers from external to internal(cuticle, Cortex, Medulla),the cuticle is consist of one layer of highly keratinized squamous cells

and the cortex is contain keratinized cells joining with each other by desmosomes and it contain pigmented granules, while the medulla comprise soft keratin (2,3), hair follicles consist of two ,sheathes the internal sheath called (Dermal root sheath) which is originated from the dermis layer besides that, the external root sheath termed the (Epidermal root sheath) it arise from epidermis layer (1,4).Wool follicles of sheep skin classified by several articles (5,6,7) into primary wool follicles and secondary wool

follicles, primary type is the bigger and deeper follicles into the dermis layer also it associated with sebaceous and sweat glands, but the secondary follicles has small diameter and it found near the surface layer of the skin and it complained only by sebaceous glands. Many authors mention the type of hair follicles in different domestic animals like in horse and cattle they are simple follicles(8), even though the compound hair (wool) follicles seen in camel and sheep(9). Sebaceous glands is another skin appendages it is simple branched alveolar glands emarginated from the epithelial of the hair root canal (10,11), so the most sebaceous glands accompanying with hair follicles, their secretion (sebum) released to the outer surface of the skin through the follicles (11). this glands has two type of cells (basal and secretory) cells, all cells of this gland hydrolysis and released with lipid droplet because it has (Holocrine type secretion) (12,13). The sebum act as antifungal, antibacterial and thermal insulation material and play important role in formation of vitamin-D (14). Sweat glands is more important skin appendages, it's simple tubular coiled gland, coiled secretory portion present in deep portion of the dermis layer of the skin and the duct of this gland reach to the outer surface passing through the epidermis layer (15). The cells of secretory segment of sweat gland is larger than cells of canal portion and they are simple cuboidal cells surrounding by myoepithelial cells, but the canal lining by stratified cuboidal cells (16). the function of the sweat gland is thermoregulation and maintain the water-ion balance of living body organism (17, 18). In mammals sweat gland dividing into two type

Results

The current study revealed that the skin of sheep comprise two important layers. The outer layer called (epidermis) and the inner layer termed (dermis). The skin appendages (wool follicles, sebaceous glands, sweat glands) present in dermis layer Figure (1).

(Apocrine and Merocrine) glands according to the histological structure and function of it, but the Apocrine sweat gland is the most numerous type distribution (19,20).

Materials and Methods

Ethical approval

The Animal Ethical Committee of Veterinary Medicine College, University of Al-Qadisiyah, Iraq, has approved the present study under permission No: 440

Ten adult ram aged 1-1.5 years were clinically healthy used for histological and morphometric study, the samples were collected from AL-Qadisiyah abattoir during autumn season from fourth different anatomical sites (ventral ear surface, neck, middle dorsum, tail), after the animals were slaughter, we prepared the samples by removing wool of skin sheep genially by special cream for (5-10) minute. Specimen of the skin was taken and fixed in 10% formalin, then washing for two hour and dehydration by ascending series of ethanol 70%, 80% , 90%, 100% in which two times 1-2 hour, clearing by using xylene twice times for half hour and embedding in paraffin wax two times for two hour after that blocks sectioned by rotary microtome with 5-6 micrometre in thickness and staining with Harris hematoxylin and Eosin stain (H&E) and the (mean and the standard error) of the diameter of primary and secondary wool follicles and the depth of them into dermis layer and also the length and width of sebaceous glands and the secretory portion of sweat glands measured by coulometer-chamber and documented by digital camera (21, 22, 23).

Histological examination of skin samples shown existing of compound wool follicles in the region, which has abundant wool fibres like in neck and middle dorsum area. They have (Trio) arrangement of primary wool follicles and (Dio or Trio) arrangement of

secondary wool follicles Figure (2). moreover we noticed the (Dio) arrangement of the primary follicles with group of secondary follicles and besides that (Mono) arrangement of primary follicles (one wool follicle with group of secondary follicles) Figure (2). In the existing study we noticed in low density wool follicles sites like in ventral surface of ear (Mono) arrangement of primary follicles. Generally all studied area revealed the primary follicles larger than secondary follicles Figure (3) and the sebaceous and sweat glands associated with primary wool follicles ,while only sebaceous glands seen with secondary follicles. The histological section shown the hair follicles are tubular invaginations of epidermis, the developers down growths of the epidermis into the dermis. Hair is made up of columns of dead keratinised cells (!). In the middle of wool, follicles there is wool fibre consist of three keratinized layers. The external layer called cuticle it involved one layer of highly keratinized cells Middle layer contain keratinized cells the long axis of them are parallel with the root of follicle and has elongated nucleus ,while the third internal layer comprise cuboidal or flat cells with round nucleus and existing of pores between cells Figure (4). There are two root sheath surrounding the wool fibre are (internal and external) (4,5), the internal sheath contain three layers of keratinised cells (Henle's layer consist of one layer of keratinized cells while Huxley's layer has 1-3. Layers of keratinized cells hold (Trichohyaline granules) and the last layer termed inner cuticle layer contain one layer of cells, these cells distingrates that the duct of sebaceous gland enter into the wool fibre. This study noticed the external root sheath cells connected with stratum Basal and st. Spinosum layer of epidermis Figure (6), this sheath separated from dermis by glassy membrane. The terminal wide segment of wool follicle named wool bulb surrounded The dermal papilla Figure (7), the diameter of primary follicles changed in different studied area, the higher diameter

measured in neck region (325.55) μm Table (1), but the lesser diameter recorded in ventral surface of ear (85.44) μm T.(1), also the size of secondary follicles varied in fourth studied sites, greater diameter of it seen in neck region (87.16) μm T.(1), while smaller one shown in tail samples (54.65) μm T.(1). The current study revealed the depth of wool follicles into dermis layer differ from one region to another, higher depth of primary follicles noticed in neck site (1825.56) μm T. (2) and lesser depth in ventral surface of ear (316.07) μm . T.(2) However bigger depth of secondary follicles present in middle dorsum (870.25) μm T.(2) and smaller depth seen in ventral surface of ear (94.20) μm T.(2). In all examined regions observed sebaceous glands, it's simple branched alveolar Glands which consist of small basal low cuboidal cells resting on basement membrane with dark nucleus, but the central cells has bigger size contain several layers of dark nucleus cells and light cytoplasm Figure (8). In all considered histological sections revealed sebaceous glands accompanied with primary and secondary wool follicles. Figure (1). Sebaceous glands have variable size, so the larger sebaceous glands shown in neck area (241.81) μm T.(3) and smaller sebaceous gland noticed in ventral ear surface (110.75) μm T.(3). Sweat gland is another skin appendages examined in our study, they are apocrine type in all studied area, and this gland has straight canal and coiled secretory portion. The duct of sweat gland consist of double layer of cuboidal cells with central nucleus Figure (9). the duct of this canal penetrate the primary wool follicle before it opened into the outer surface of the skin, the secretory portion of this gland has wide lumen lining with by one layer of cuboidal cells surrounding with myoepithelial cells Figure (10), this glands present in different sites with varied size, so the longer sweat gland appear in middle dorsum region (225.91) μm T. (4) and the shorter gland present in ventral surface of ear (94.15) μm T.(4).

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Table (1):-explain the mean diameter (μm) of the primary and secondary wool follicles of the sheep skin

Tail	Middle Dorsum	Neck	Ventral ear surface	Anatomical regions
200.30±5.15	220.26±7.60±7.60	325.55±5.95	85.44±3.92	Diameter of primary follicles Mean±SE
54.65± 5.70	79.50±5.70	87.16±5.65	57.22±3.25	Diameter of secondary follicles Mean±SE

Table (2):-explain mean depth (μm) of primary and secondary wool follicles of sheep skin

Tail	Middle Dorsum	Neck	Ventral ear surface	Anatomical regions
1505.22±10.418	1725.23±70.15	1825.56±90.72	316.07±30.21	Depth of primary follicles Mean±SE
654.20±36.96	870.25±42.21	840.54±60.20	94.20±17.21	Depth of secondary follicles Mean±SE

Table (3):-explain the long axis and width(μm) of sebaceous gland of the sheep skin

Tail	Middle ear	Neck	Ventral ear surface ear	Anatomical regions
180.00±5.90 *41.60±5.10	225.00±8.70 *35.00±5.50	241.81±11.20 *55.37±9.40	110.75±030 *42.00±3.12	Long axis * width of sebaceous gland

Table (4):-explain the long axis and width (μm) of sweat gland of sheep skin

Tail	Middle dorsum	Neck	Ventral ear surface	Anatomical regions
155.47±10.22 *70.23±11.33	225.91±31.24 *68.20±19.82	205.12±31.24 *70.22±17.39	94.15±7.35 *36.70±7.49	Long axis and Width of sweat gland Mean ±SE



Figure(1):-Vertical section of the skin of neck region shown:- (A)epidermis layer (B)dermis layer (C)wool follicle (D)sebaceous gland (E)sweat gland. (H&E)stain 100 x

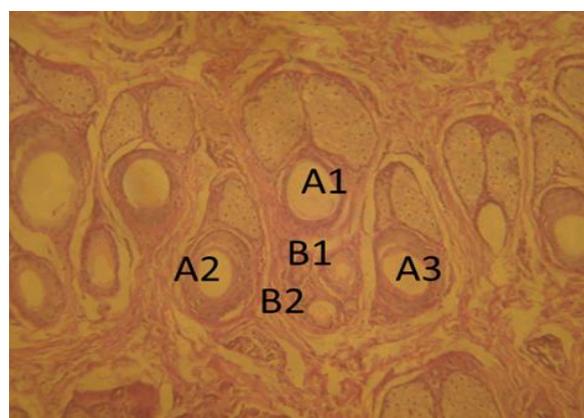


Figure 2:-Vertical section of the skin of middle dorsum region shown the(trio) arrangement of the primary wool follicles :-((A1,A2,A3) primary wool follicles,(B1,B2) secondary wool follicles (H&E)stain 100x

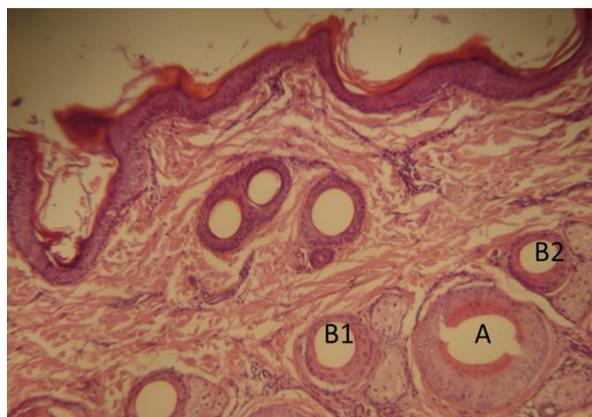


Figure (3):-Vertical section of the skin of the ventral ear surface region shown :-(A) primary follicle (B1,B2) secondary follicle (H & Estain)100 x.

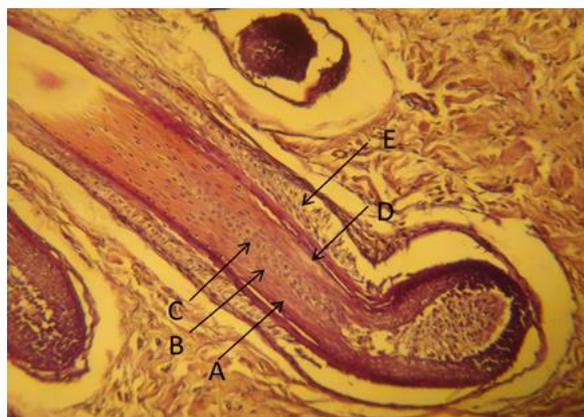


Figure (4):-Vertical section of the skin of neck region shown :-(A) cuticle (B)cortex (C)medulla (D)internal root sheath (E) external root sheath.(H&E) stain 200x



Figure (5): Vertical section of the skin of tail shown: (A)internal root sheath (B)external root sheath (C)wool fiber. (H&Estain) 100 x.

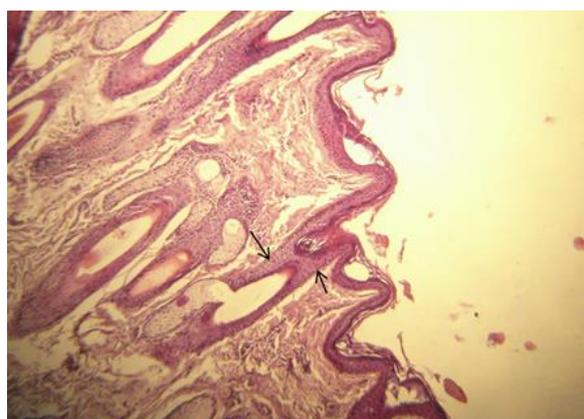


Figure (6): verticle section of the neck region shown external root sheath connected with st.basale and st. Spinosum (→) (H&E)stain 100 x



Figure (7): verticle section of the middle dorsum region of the skin shown:-(A) wool bulb (B)dermal papilla (H&E)stain 200 x

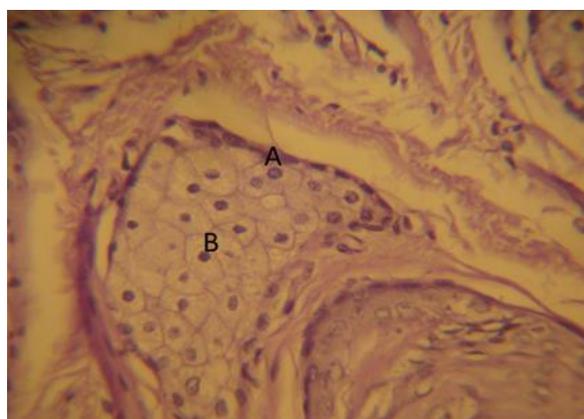


Figure (8): Vertical section of the tail region of sheep skin shown:-(A) one layer of basal cells (B) several layers of central cells.(H&E)stain 200 x

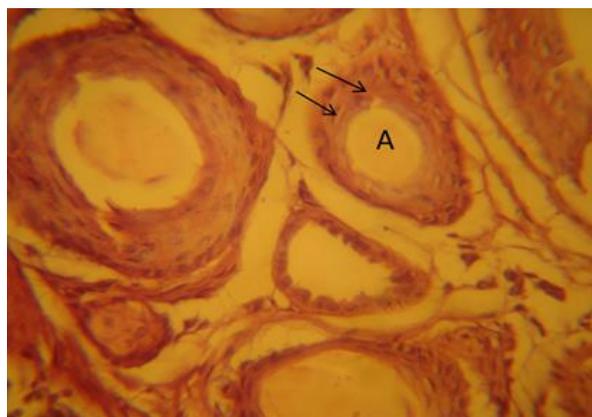


Figure (9): Vertical section of neck region of sheep skin shown :- (A) the duct of sweat gland consist of two layers of cuboidal epithelial cells (→) (H&E) stain 200 x

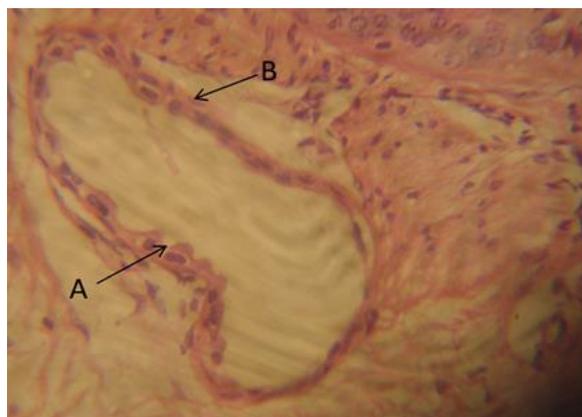


Figure (10):- vertical section of ventral ear surface region shown :- (A)the secretory portion of sweat gland lining with one layer of basal cells (B)myoepithelial cells (H&E)stain 200 x.

Discussion

The present histological study revealed the dermis layer of sheep skin in heavy growth wool follicles regions (neck and middle dorsum) contain the (Trio), (Dio) and (mono) arrangement n of wool follicles, our result respected with (24)in American sheep,(25) in goat skin. Different articles described the sites with low growth wool follicles they hold (mono) arrangement primary wool follicles like in ventral ear skin (26) in Iran Lori sheep. Another research's in the skin of ox and buffalo (27), (28) in the skin of camel and (29) in the skin of human revealed present of simple type hair follicles, this result agreement with our finding. Primary wool follicles mean the rough wool fibres ,but the secondary follicles reflected the smooth wool fibres ,the same results seen in the study of the(Merino Australian) sheep (30).Our study present variation in the diameter of wool follicles in selected area .generally the diameter of primary follicles is larger than the diameter of secondary follicles, in neck region primary and secondary follicles noticed bigger than another studied sites, our finding similar to that in local breed ox (31) and they ensure the diameter of primary follicles are higher in dorsal surface than the ventral surface of the skin area. Also the sex of animals affected in the diameter of follicles, in female the

primary follicles diameter (2036.02) μm while in male (2633.90) μm ,so in female has lesser diameter than male (26). The current study seemed the density of hair follicles differ from one region to another ,so the neck, middle dorsum, and tail regions have higher density of wool fibres ,but the ventral ear surface has few wool follicles, the same results noticed in the study of the skin of camel, small ruminant and ox (31,32,33).Our study observed the sebaceous gland associated with the upper segment of the wool follicle, the sebaceous gland duct penetrate the follicle, many authors described the same result (13 and 24), the study of the skin of one humped camels and the skin of black goat (25,34) they noticed the sebaceous glands in some selected area are lobulated and this result disagreement with our finding. Another articles (35, 36) described sebaceous gland present as a groups accompanying with modified sweat glands in special regions like in infra-orbital ,inguinal and between fingers of animal. The size of sebaceous glands varied in the four anatomical designated sites ,in neck and middle dorsum existing biggest than ventral ear surface and tail and we are agree in this finding with (28).In the study the skin of Egypt buffalo remarked pair of sebaceous glands accompanying with hair follicles, this

observation not approved in our study. The present study in the skin of local breed sheep revealed abundant distribution of Apocrine sweat gland in all selective regions ,it's simple tubular coiled gland ,this result were reported in (37) in ox skin and (35) in local breed goat, also the study of horse skin (38) ensured our finding ,they noticed present of Apocrine sweat gland in most anatomical area except in upper and lower lips skin ,but our pattern is differ with (39) in the study of human skin ,they seen Apocrine sweat gland in only in genital and axillary regions. Our study observed variation in the diameter of secretory portion of sweat glands (T.4) they

appeared bigger in middle dorsum region than another anatomical selected area, but this finding not applied with many research's like (40,41) in the study of ram and gazelle skin, they noticed present of this gland with higher diameter in scrotum region. The current study present the duct of sweat gland opened into the primary wool follicle above the orifice of sebaceous gland ,and there is a relationship between the size of secretory segment and diameter of the duct of sweat gland ,the same observation seen in (27, 40), besides they noticed a Giant sweat glands in scrotum and muzzle regions.

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