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Pre and Postnatal Development of the Rabbit Thin Skin

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Abstract: Rabbits are a popular model in laboratory animal medicine especially in the fields of human dermatology, experimental pharmacology and experimental toxicology. The present study aimed to investigate and re-evaluate, by light and scanning electron microscopy, the pre and postnatal development of rabbit skin. Early at embryonicage 15days(E15), the developing skin formed only of a single or double-layered epidermis and a loosely arranged mesodermal cells; the future dermis. At E17, epidermal stratification became clear and formed of basal layer, intermediate layer and the superficial (peridermal) layer. Additionally, the outer mesodermal cells of the dermis became condensed below the basal cells of the epidermis and followed by the formation of hair placodes at embryonic period E19. At E 25 the stratification of developing epidermis became more distinct with an increase in the extension and differentiation of hair follicle. At late pregnancy at E28, the epidermis became fully differentiated and nearly showed mature structure where the intermediate layer of epidermis differentiated into spinous and granular layers and the appearance of the keratinized layer. Just before birth atE30, the hair shaft started to protrude through skin surface associated with the development of the sebaceous glands as an outgrowth from the hair follicle. Postnatally and at one week old rabbit, the skin appeared nearly mature, the dermis became thicker with well-developed dermal papillae and the majority of the free ends of hair shafts elongated and protruded upon the surface of epidermis. At two months old,the rabbit skin became fully matured with marked increase collagenous fibers within dermis. During pre- and postnatal development of rabbit skin we could not observe any sweat glands of the examined skin samples. We can conclude that the development of the rabbit thin skin and its appendages was mainly established during the prenatal period with few developmental events during postnatal period. The extensive fur covering and absence of the sweat gland of rabbits might be considered during intensive rabbit production to avoid stress upon rabbit thermoregulation mechanisms.

Key words: Rabbits · Skin · Hair · Development

INTRODUCTION

Rabbits (Oryctolaguscuniculus) are increasing in popularity as house pets and popular model in laboratory animal medicine due to its relatively large size and docile nature. Skin diseases of rabbits may be the most common cause for an owner's visit to the veterinarian and these diseases are also very common in laboratory colonies and may interfere with the quality and conditions of research [1]. Skin is the largest organ of the body and consists of a complex layered structure, which forms a permeable barrier between the body and the outside environment allow an exchange of heat, air, as well as fluids containing matter of very low molecular weight [2]. The skin is structured in two layers, namely the epidermis and the dermis. The dermis consists of connective tissue and contains nerves, blood and lymph vessels, hair follicles, sebaceous glands and sweat glands. The epidermis, on the other hand, contains cells in several stages of differentiation. During the differentiation process the cells migrate from the basal layer to the surface where they cornify [3]. The skin of rabbits and rodents can be viewed as a window to their general state of health. Observation of abnormalities of skin and pelage may dictate changes

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in diet or husbandry or lead to the diagnosis of other underlying disease [4]. The rabbit's skin has blood vessels immediately under the dermis and the fascia superficialis is well differentiated due to the elastic fibers and dense collagen content [5].

Skin development from single-layered surface ectoderm to multi-layered keratinized epidermis has been well documented [6]. Proper skin development is dependent on a series of inductive signals traveling between epithelial and mesenchymal progenitors [7, 8]. In mammals, embryonic skin epithelial cells are pluripotent, able to choose between epidermal and hair follicle cell fates. Epithelial progenitor cells in skin give rise to epidermis as well as the epithelial component of skin appendages, including hair follicles and associated sebaceous glands [9-11]. Commitment to follicle formation occurs when an underlying mesenchymal cue instructs overlying ectoderm to commit to forming an appendage [12-14]. Early-stage follicles consist of an epithelial thickening, called a placode and an adjacent mesenchymal condensate in the developing dermis. The follicle epithelium grows downward through the dermis and into the subcutaneous fat, where it surrounds the condensate derived hair follicle papilla to form the hair bulb [14, 15]. The final differentiated cell type to appear in the developing follicle is the oil-rich sebocyte, which arises from cells within the superficial hair follicle [16].

Morphogenesis, histogenesis and histochemistry of fetal skin of different animals received little attention, recently, as observed from the available literatures [17-21]. Although rabbits are wildly used as the main laboratory animal in the experimental human dermatology and as the main model used to study healing and regeneration processes of human skin. The detailed descriptions, including the scanning electron microscopy, about the prenatal and postnatal development of rabbit skin have been not reported yet. For these reasons the present study aimed to investigate by light and scanning electron microscopy the pre and postnatal development of rabbit skin.

MATERIALS AND METHODS

Embryos, fetuses, newborn and adult V-line white rabbits were obtained from the farm of Faculty of Agriculture, Alexandria University, Egypt. The age of fetuses and rabbits was estimated according to the records of the farm. Prenatally, the day of mating was considered the day one of embryonic life while the day of delivery was considered the day one of postnatal life.

Light Microscopic Examination: Apparently healthy female rabbits, at pregnancy stages E15, E17, E19, E21, E25, E28and E30, were anaesthetized and slaughtered and then embryos and fetuses were removed. The fetuses at stages E15, E17 and E21, the whole fetuses were fixed in 10% neutral buffered formalin (for 2-5 days) and Bouin's solution (for 24-48 hours). In older fetuses, newborn and adult rabbits, skin samples from different regions were dissected out and were preserved in the same fixatives for the same periods. The skin samples were then dehydrated in ascending grades of ethanol, cleared in xylene, impregnated with melted paraffin wax. Finally paraffin blocks of the processed samples were prepared. Thin sections (5-7 μm thick) were cut and mounted on egg albumin-glycerin coated glass slides, dried in an electrical incubator (at 37 degree) for 30-60 minutes and stained with the following stains hematoxylin and eosin (H &E), Masson's trichrome technique, Van Gisson stain and Periodic Acid Schiff reaction (PAS).

Scanning Electron Microscopic (SEM) Examination: Samples from the skin of fetuses at E25 and E30 and from rabbit aged one and two months were used. The samples were immediately immersed in 4F1G (2% formaldehyde, 1.25%glutaraldehyde in 0.1 M sodium cacodylate buffer) fixative at pH 7.2 and stored at 4°C. The fixed samples were washed in 0.1 M cacodylate buffer containing 5%sucrose processed through tannic acid, dehydrated in graded ethanol series for 15 minutes in each of 50%, 70%, 80%, 90% and absolute alcohol. The critical point dried samples (with carbon dioxide) were then attached to stubs with colloidal carbon and coated with gold palladium in sputtering device. The samples were examined and photographed with JEOL SEM 5300 operating 15Kv in the Faculty of Medicine, Tanta University, Egypt.

RESULTS

Embryonic Age (E15): Light microscopy of rabbit fetus skin at the dorsolumbar region showed the developing skin in the form of a single- or double-layered epidermis and the underlying mesoderm of the future dermis appeared as a loose arrangement of cells without any particular pattern of arrangement. In the regions where epidermis was composed of double layer, it could be differentiated into basal layer of cuboidal or low columnar cells (stratum basale) and a superficial irregular or globular-shaped cell layer (periderm). The basal layer characterized by marked separation from the underlying mesenchymal cells of the developing dermis (Fig. 1A& B).

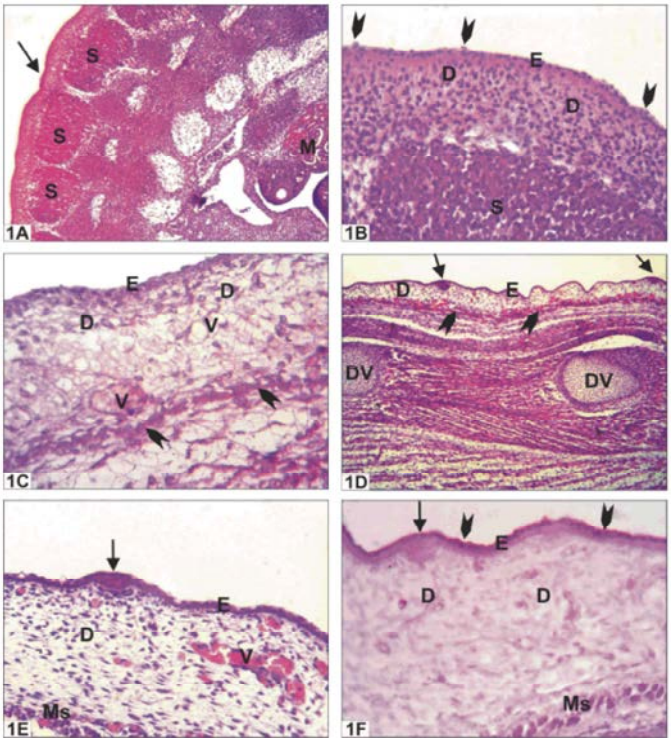

Fig. 1: Panel A, micrograph of rabbit fetus at E15 showed the developing skin (arrow) sommites (S) and mesonephros (M) (H&E.X 100). Panel B, was micrograph of rabbit skin at E15 showed developing epidermis (E), superficial cells or periderm (arrows), developing dermis (D) and sommit (S) (H&E.X 400). Panel C, micrograph of rabbit fetus at E 17 showed developing epidermis (E), superficial cells or periderm (arrows), developing dermis (D), blood vessels (V) and developing muscles (arrow heads) (H&E.X 400). Panel D, was micrograph of E19 showed developing epidermis (E), developing hair placodes (arrows), developing dermis (D), developing vertebrae (DV) and developing muscles (arrow heads) (H&E.X 100). Panel E, was a higher magnification of panel D showed developing epidermis (E), developing hair placodes (arrows), developing dermis (D), blood vessels (DV) and developing muscles (Ms) (H&E.X 400). Panel F, was micrograph of of E19 showed developing epidermis (E), developing hair placodes (arrows), PAS positive periderm (arrows heads) developing dermis (D) and developing muscles (Ms) (PAS.X40)

At E17, stratification became clear in some area of the developing epidermis where it was formed of basal layer, intermediate layer of flat or rounded cells and superficial layer (periderm). The superficial mesodermal cells of the dermis became more condensed under epidermis with many wide blood vessels in the dermis. Within the deep layer of the developing dermis the mesenchymal cell started to be differentiated into muscle cells (Fig. 1C).

With age progress and at E19, the stratification became more pronounced within the developing epidermis even though the single-layered epidermis existed in manyregions. The superficial layer of the dermis became denser under the epidermis and formed of large number of fibroblast-like cells and blood vessels. Muscle cells began to appear, clearly, within the CT under the skin especially in the deep region. The most characteristic feature of this stage was the initiation of the hair follicles development. The hair follicle was observed as local proliferation and condensation of epidermal basal cells forming a hair bud or placode. The hair bud appeared as ridge-like thickening as a result of propagation of the basal cells and the development of the intermediate layer. Marked aggregation of mesenchymal cells (fibroblast-like cells) of the dermis became condensed beneath each developing bud forming the future dermal papilla of hair follicle (Fig. 1D, E). The dermis increased in thickness with marked increase in collagen fibers and bounded by muscular tissue at its deep part. At this stage of development, the superficial cells (periderm) of the epidermis showed strong PAS positive reaction for the first time (Fig. 1F and 2A).

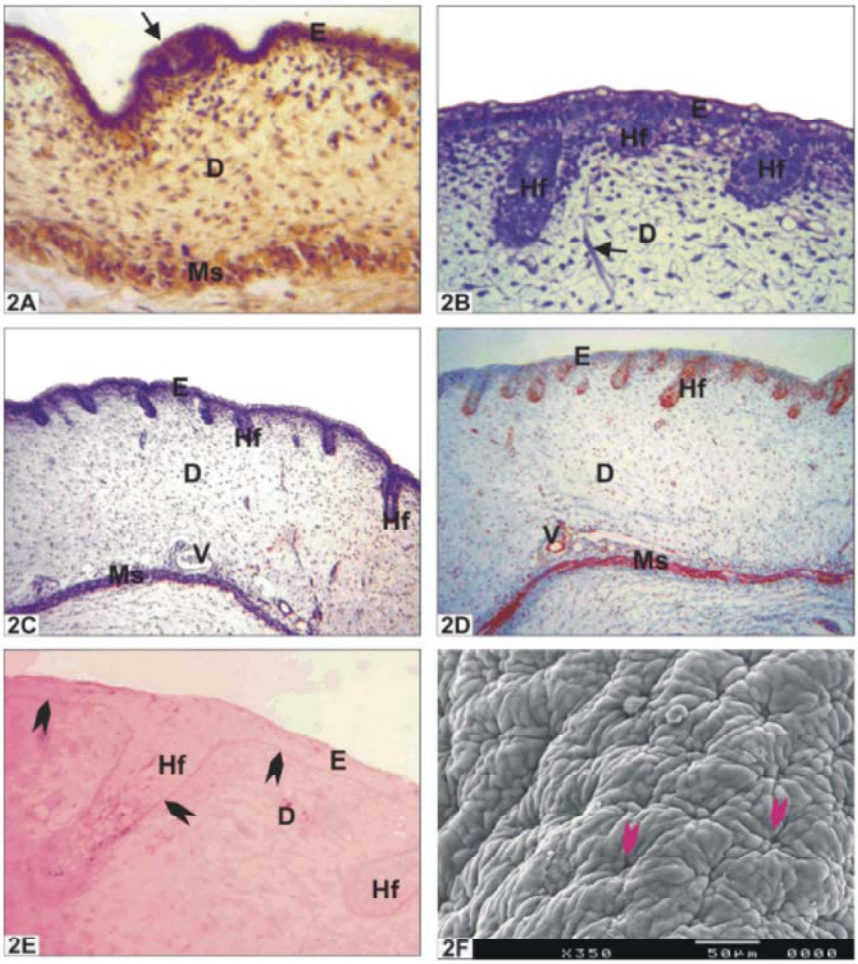

Fig. 2: Panel A, micrograph of E19 showed developing epidermis (E), developing hair placodes (arrows), developing dermis (D) and developing muscles (Ms) (Van Gisson. X 400). Panel B, micrograph of E21 showed stratified epidermis (E), dermis (D), developing hair follicles (Hf) and muscles (arrow) (H&E.X 400). Panel C, micrograph of E25 showed stratified epidermis (E), dermis (D), developing hair follicles (Hf), blood vessels (V) and developing muscles (Ms) (H&E.X 100). Panel D, micrograph of E25 showed stratified epidermis (E), dermis (D), developing hair follicles (Hf), blood vessels (V) and developing muscles (Ms) (Trichrome X100). Panel E, micrograph of E25 showed stratified epidermis (E), dermis (D), developing hair follicle (Hf) and basement membrane (arrow heads) (PAS.X 400). Panel F, scanning electron micrograph of embryonic stage E25 showed the skin surface with the invagination of developing hair follicles (arrow heads)

Later on and at E21, the stratification of the epidermis became the common feature of the epidermis everywhere and it was formed of the basal layer of columnar cells, the intermediate layer of flat or polygonal cells and the superficial (peridermal) layer of flattened cells. The hair follicles showed different stages of development where the older one became elongated and deeply invaginated into the dermis. The primitive follicle appeared as solid mass of cells where its peripheral cells were similar and continuous with the basal cells of the epidermis. These cords, as well as the developing epidermis, were surrounded by condensation of dermal mesenchymal cells in all examined rabbit fatal skin at this developmental stage. Some of these mesenchymal cells, close to the follicles, began to differentiate into muscle cells that were the first primordial structure of arrectorpili muscle of the hair follicle (Fig. 2B). The multi-layered arrangement of the epidermis became more established into the 3 layers without keratinization at E25. Different stages of hair follicles development could be noticed where the early developed hair follicles increased in length and extended deeply within the dermis.

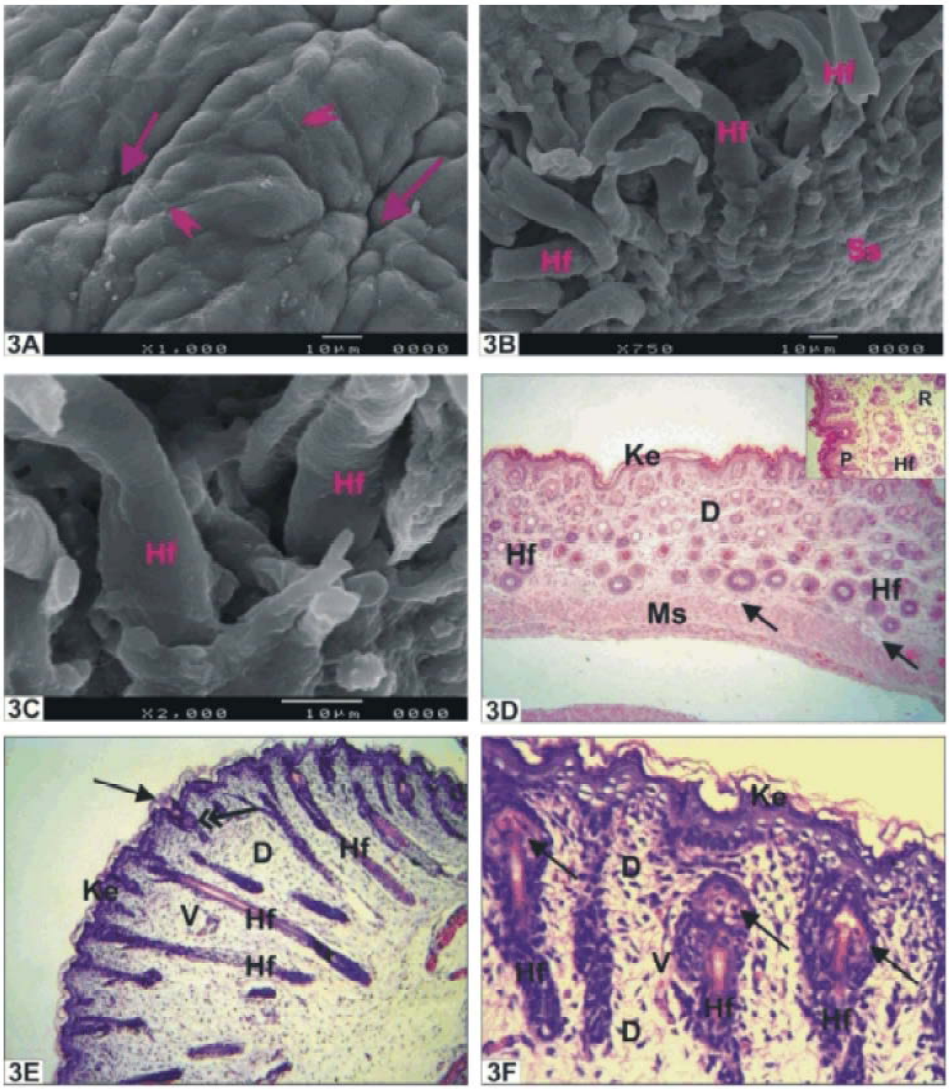

Fig. 3: Panel A, a higher magnification of scanning electron micrograph of E25 showed the invagination of developing hair follicles (arrows) and the cell boundary of superficial cells (arrow heads). Panel B, scanning electron micrograph showed the skin surface and cut edges of developing rabbit skin at embryonic stage E25 showed skin surface (Ss) and developing hair follicles (Hf). Panel C, a higher magnification of Panel B showed the extended hair follicles (Hf). Panel D, micrograph of E28 showed stratified keratinized epidermis (E), dermis (D), developing hair follicles (Hf), developing fat cells (arrows) and muscles (Ms) (H&E.X100). The inset is a higher magnification of panel D showed the hair follicle (Hf), papillary layer of dermis (P) and reticular layer (R) (X 400). Panel E, micrograph of E30 showed stratified keratinized epidermis (E), dermis (D), developing hair follicles (Hf), initially protruded hair shaft (arrows), developing sebaceous gland (double head arrow) and blood vessels (V) (H&E.X 100). Panel F, was a higher magnification of panel E and showed stratified keratinized epidermis (E), dermis (D), developing hair follicles (Hf), developing sebaceous gland (arrows) and blood vessels (V) (H&E.X 400)

The hair follicles were surrounded by fibrous sheath, derived from the dermis, associated with blood capillaries but the hair shaft did not develop yet (Fig. 2C, D). At this stage weak PAS reaction, restricted mainly within basement membrane, some peridermal cells and some cells of the developing hair follicles, was observed (Fig. E). SEM of this stage confirmed light microscopy where the outer surface of the skin still non-keratinized and showed several invaginations of the primordia of hair follicles. The superficial cells of the epidermis showed undulation and

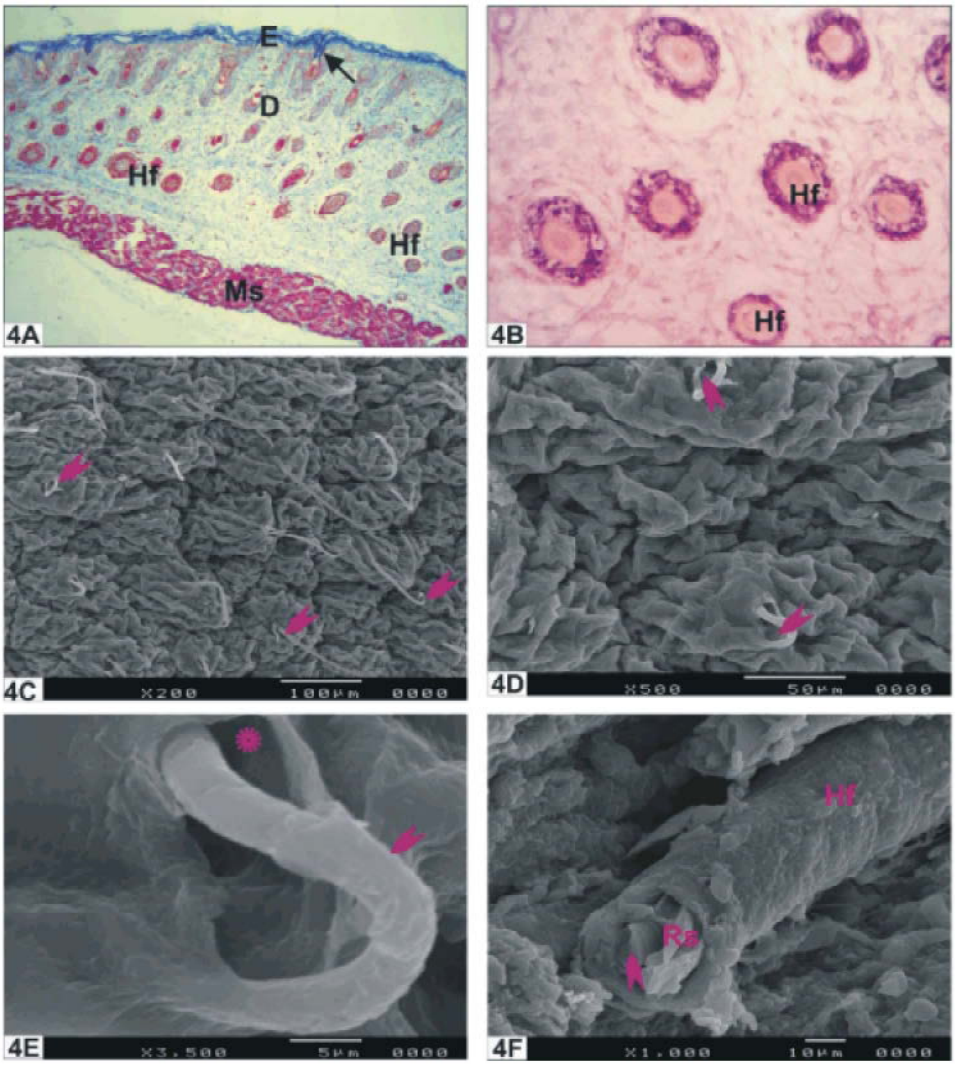

Fig. 4: Panel A, micrograph of E30 showed stratified keratinized epidermis (E), dermis (D), developing hair follicles (Hf), extended keratinization within the hair canal (arrow) and muscles (Ms) (Trichrome. X 100). Panel B, micrograph of E30 showed a positive reaction of root sheaths of hair follicles (Hf) (PAS. X400). Panel C, scanning electron micrograph of E30 showed the skin surface and protruded hair with its folded end (arrow heads). Panel D, was a higher magnification of panel C showed the folded end of protruded hair (arrow heads). Panel E,was a higher magnification of protruded hair showed the hair (arrow head) and the opening of the hair canal (star). Panel F, scanning electron micrograph of the developed hair follicle at stage E30 showed the hair follicle with its covering fibrous sheath (Hf), root sheaths (Rs) and the hair shaft (arrow head)

the cell boundary was clear in the form of shallow ridges (Fig. 2F, 3A). Deeply within the dermis, different stages of hair follicles extended toward the deep part of the dermis (Fig. 3B, C).

Nearly complete differentiated epidermis formed of keratinized stratified squamous epithelium was observed at E28. Additionally the epidermis was clearly thickened and consisted of several distinct strata during this stage. The dermis became differentiated into superficial papillary and deep reticular layers. The hair follicles, which increased greatly in number, became nearly fully differentiated where the external root sheath and the internal root sheath could be demarcated specially the older ones. No hair shafts could be seen protruding from the epidermal surface at this stage (Fig. 3D).

At E30, the epidermis appeared thicker and almost completely developed with increased thickness of keratinization. The dermis became thicker and the dermal

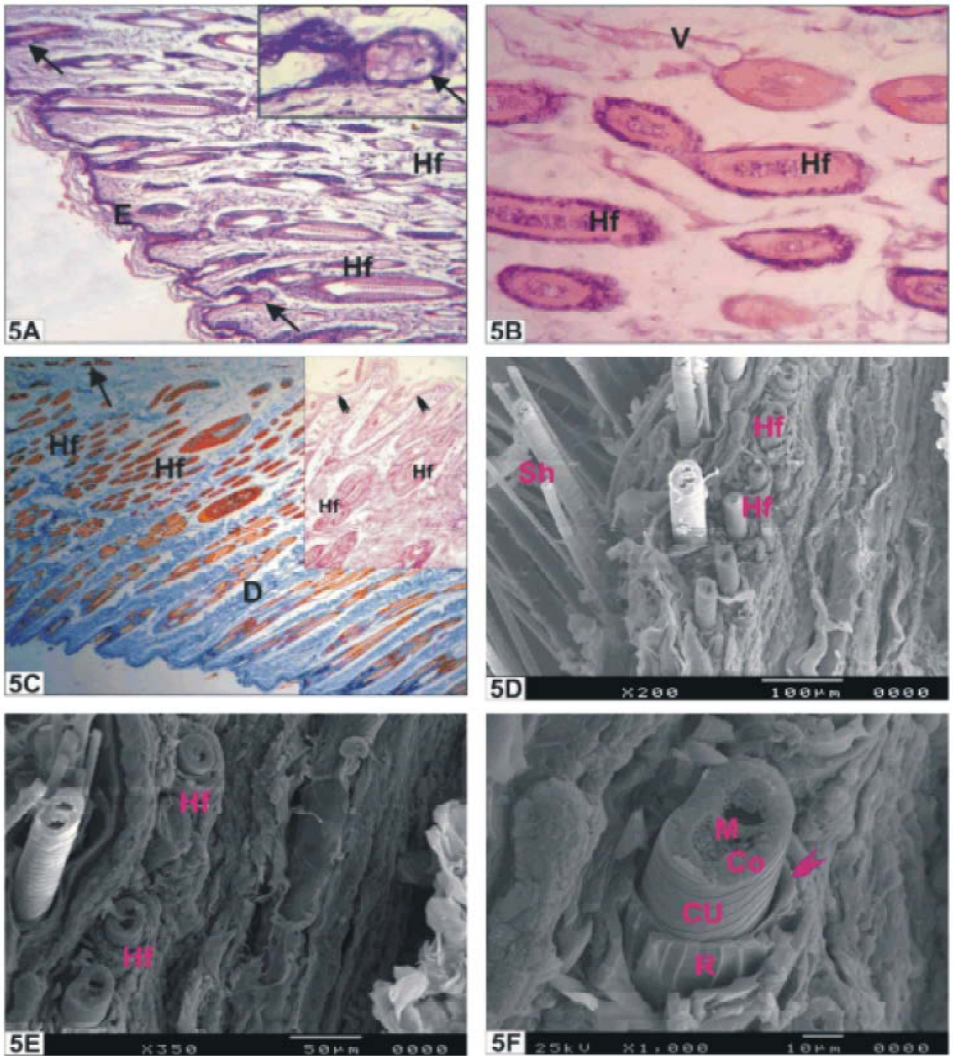

Fig. 5: Panel A, micrograph of one week old rabbit skin showed stratified keratinized epidermis (E), developing hair follicles (Hf), advanced developed sebaceous gland (arrow) (H&E.X 100). The inset was a higher magnification of panel A showed the sebaceous gland (arrow) (X400). Panel B, was PAS reaction of the dermis of the one week old rabbit showed positive reaction within the root sheaths and the hair medulla (Hf) and blood vessels (V) (PAS. X400. Panel C, micrograph of two month old rabbit skin showed stratified keratinized epidermis (E), extensive collagen fiber within dermis (D) deeply extended hair follicles (Hf) and muscles (arrows) (Trichrome. X 10). The inset was micrograph of two month old rabbit showed PAS positive reaction in the hair follicle (Hf) and the basement of epidermis (arrows head) (PAS.X400). Panel D, was scanning electron micrograph of two month old rabbit skin showed the extensive surface hair (Sh) and hair follicle within the cut edge (Hf). Panel E, was a higher magnification of the cut edge of panel D showed the fully developed hair follicle (Hf). Panel F, was a higher magnification of the fully developed hair follicle showed the root sheaths (R), cuticle of the inner root sheath (arrow head), medulla of the hair shaft (M), cortex of the hair shaft (Co) and the outer cuticle of the hair shaft (Cu)

papillae were more developed. The external root sheath and the internal root sheath,surrounding the cortex of the developing hairs, were obviously demarcated in the majority of the hair follicles. Furthermore some of the hair shafts elongated and began to protrude upon the surface of epidermis. The main feature of this stage was the differentiation and development of the sebaceous glands where it appeared as small cellular budding from the upper third of the growing follicles (Fig. 3E, F). The hair follicle surrounded by intensive collagenous fibrous sheath and

the cells of outer and inner root sheaths of the hair follicle showed strong positive PAS reaction (Fig. 4A, B). By SEM, the cornification of the skin surface was distinct with clear folds of different heights confirming the result reported by LM. Some hairs appeared with folded ends on the skin surface, where the shaft was bent upon its self during leaving the opening of its canal (Fig. 4C, D,and E). Within the dermis the hair follicles appeared in the mature form with extensive fibrous sheath (Fig. 4 F).Till this stage the development of sweat gland could not be detected.

Early postnatal and at one week old rabbit, the skin appeared nearly mature with extensive hair coat. The dermis became thicker and the dermal papillae were more developed. The majority of the hair shafts extended and protruded through the hair canal upon the surface of epidermis (Fig. 5A). The sebaceous gland became enlarged and more developed (Fig. 5A).The outer root sheath of the hair follicles and the medulla of the hair showed positive PAS reaction (Fig. 5B). At two months old, the rabbit skin was fully developed with marked increase of the dermal papillae and epidermal ridges. The outer collagenous sheath of hair follicle become more intensive while the PAS reaction of the outer and inner root sheath was markedly reduced than that of the previous stage. The dermis was relatively thicker with marked increase in its collagenous fibers (Fig. 5C). SEM showed fully matured hair structures. The keratinized hair had central large vacuolated cells that form the core of the hair shaft or medulla that was surrounded by the cortex followed by the cuticle that was composed of proximally directly keratinized cells. The inner layer of the inner root sheath became differentiated into the distally directed cuticle of the inner root sheath. This arrangement of cells composed the cuticle of the hair and cuticle of the internal root sheath interlocked the opposing free edges of these cells (Fig. 5D, E, F).

DISCUSSION

In this study we showed by light and scanning microscopy the pre- and postnatal developmental events of the rabbit's thin skin. As long as we know, the current study considers the first scanning microscopical observation of fetal and adult rabbit skin. As early as at E15, the epidermis of rabbit was formed mainly of a single layer of cells with few regions of double layered of cells, stratum basale and periderm. The observation of periderm cells of developing skin of rabbits was also reported in rabbits at the same embryonic age [21], while in rat, the epidermis was identified as a double-layered, an inner (basal) and outer (periderm) layer at E12 [20]. The periderm layer was reported also in the developing epidermis of human [17, 22], rats [19, 23] and mice [24].

Although the periderm has no clearly defined role during development, its disappearance coincides with the completion of barrier formation in human and mouse skin [22, 25,26]. In rabbit, the ultrastructure of peridermal cells is characterized by few cytoplasmic projection and glycogen sorting cells between ages 15-18 days only [21], while in rat the survival of glycogen sorting cell of developing epidermis is observed until birth [19]. The role of the periderm in the uptake of glucose from the amniotic fluid, production of the amniotic fluid, or both was previously suggested by many authors [23, 27]. This suggestion isn't accepted by [21] in rabbit because their study revealed periderm cell with few cell organelles. An important feature of the developing epidermis is its lack of cornified envelope, which appears shortly before the end of the embryonic period [28]. Up to this specific point in time, the emerging epidermis is covered by a protective epithelium, which is considered the main barrier between the developing embryo and the surrounding amniotic fluid. This protective epithelium called the periderm consists in mammals of a monolayer of flattened epithelial cells [17]. Cells of the periderm have been shown by electron microscopy to be tightly joined by sites of close membrane contacts characteristic of tight junctions [29]. The impermeability of the periderm and the barrier function of these tight junctions structures have been shown for fetal rats by the use of lanthanum-containing solution [30].

At embryonic age E17, epidermal stratification became clear in some area of the developing epidermis where it was formed of several layers. Additionally, the outer mesodermal cells of the dermis became condensed below the basal cells of the epidermis. This event was followed by the initiation of the hair follicle development at embryonic period E19 through the formation of hair placodes. In the mice, primary hair follicle morphogenesis begins at approximately E14.5 [16, 31], in rat the primary hair follicle was observed at E16 [20], while in dog the hair follicle observed at E40 [32]. in buffalo, the hair follicles of shoulder, trunk and thigh regions start to appear at the end of 3rd month and last to the end of the 4th month and the hair emerged at the 7th month [18]. While in camel, the first hair follicle initiation is reported in 15 cm CVRL camel fetuses and the addition of new follicles lasts until 50 cm CVRL fetuses [33]. In human, the early developed hair follicle is reported at 14 week old fetus [27]. This variation in the hair follicle development might be attributed to the difference in gestation period between the different species investigated.

The present observations might confirm several studies that reported the relations between mesenchymal cells of dermis and the development of the hair follicles. Our result about aggregation and condensation of the dermal cells beneath the epidermis was in agreement with previous study in rabbit [21]. They reported that, only few mesenchymal cells of the developing dermis were noticed between E10-15, then considerable numbers of dermal mesenchymal cells are aggregated beneath the epidermis between E15-18 that is associated with appearance of the hair placodes at E18. Placode formation might stimulated by signals sent from the underlying mesenchyme, the dermal cells. The mesenchymal cells aggregate immediately underneath the epidermis and mark the location of the new hair follicle [34]. These aggregates or dermal condensates are the precursors of the dermal papilla, the permanent mesenchymal part of a hair follicle [35]. Additionally, dermal papilla (DP) cells are thought to instruct matrix progenitors during hair growth and bulge stem cells during adult hair regeneration in the hair cycle [36, 37], but the precise molecular mechanisms of DP niche function remain elusive. Likewise, during embryonic hair follicle formations the precursors of DP cells in dermal condensates are thought to instruct epidermal placode cells that contain the future hair follicle stem cells [38]. The occurrence of dermal aggregates and thus the formation of hair follicles, is controlled in a strict spatiotemporal manner and the signals involved in this process have been extensively reviewed [34, 39-42].

At E21, the stratification of the epidermis became established where it was formed off basal columnar cells, intermediate flat or circular cell layer and the superficial or peridermal. In rabbit [21] reported similar result at embryonic day 20 and they added that at this age the appearance of intermediate layer in the epidermis together with the aggregation of mesenchymal cells beneath the epidermis are common features of the developing skin of rabbit. The present study coincides with the observation of [21], in E25-26 rabbit fetus, in that the peridermal cells degenerate and exfoliate from the epidermis and this is associated with the differentiation of the horny layer of the epidermis. Additionally, the formation of hair shaft without protrusion outside the skin surface is another point of agreement at this developmental stage. At E18.5 mouse fetus, the inner root sheath develops into the hair channel and the outer root sheath maintains contact with the basement membrane [35]. In rat fetus, the epidermal maturation was marked by the appearance of cornified cells below the degenerated periderm at E20 [20].

At E28, the epidermis became fully differentiated and nearly showed mature structure where the intermediate layer of epidermis differentiated into spinous and granular layer and the appearance of the keratinized layer. While at E30 the keratinized epidermis became fully differentiated and the hair shaft began to protrude through skin surface. In a partial agreement, [20] in 6-days-old rat observed keratinized hair penetrates the epidermis with marked increased thickness of the stratum corneum. The main feature of embryonic E30 was the development of the sebaceous glands as an outgrowth from the hair follicle. Sebaceous glands are formed late in mouse development and appear when follicles elongated into hair pegs [35]. In rat, the sebaceous gland developed as an outgrowth of the follicular outer root sheath during early postnatal development and at the same time, the arrectorpili muscle was recognizable as bundles of smooth muscle fibers attached to the connective tissue sheath surrounding the hair follicle [20]. Additionally, the last differentiated cell type to appear in the developing follicle is the oil-rich sebocyte, which arises from cells within the superficial hair follicle [16]. Over the course of several days, the expanding pool of sebocytes forms a gland located outside of the hair follicle, with sebocytes releasing their contents into the hair canal not far below the skin's surface [43].

Early postnatal, the skin appeared in the mature form with extensive hair coat with enlarged and well developed sebaceous glands. With increasing the age and at about two months old rabbit was characterized by extensive outer collagenous sheath of hair follicle, relatively thicker dermis than the previous stage and marked increase collagenous fibers within dermis and subcutaneous tissue. This observation coincides with the result of [5] who reported that the skin of rabbit is characterized by well differentiated the fascia superficialis due to the elastic fibers and dense collagen content. Moreover, our postnatal investigation on the rabbit skin showed the epidermal layers, except the stratum corneum, were relatively thin. These observations are in agreement with what is reported by [20] that the differentiated epidermal layers were very thin, with prominent stratum corneum, in the skin of 15-days-old rat. They added, the skin of a 55-days-old rat epidermal strata were very thin, while the stratum corneum amounted to 3- to 4-fold of the epidermal thickness. During our pre and postnatal study we could not localized any sweat glands of the examined skin samples of rabbit. This observation partially coincides with the study of [21] who did not report any comment

about sweat gland during his prenatal studies. Thus the rabbit might use variable mechanisms for thermoregulation other than sweat glands. In non-sweating animals like rabbits, heat dissipation is carried out by altering the breathing rate to increase vaporizing of high moisture through the respiratory air. In addition the nasal mucosa and ear play a big role in thermoregulation of rabbit [44]. This character of rabbit as non-sweating animal allowed the uses of it as main laboratory animal in the field of experimental pharmacology and toxicology [45].

Finally we can conclude that the development of the rabbit skin and its appendages was mainly established during the prenatal period with few developmental events during postnatal period. The epidermal and dermal cells interaction might relate directly to the initiation of the hair follicle development. Extensive fur covering and absence of the sweat gland of rabbits might be considered during intensive rabbit production to avoid stress upon rabbit thermoregulation mechanisms.

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