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Review

Localization of sex steroid receptors in human skin

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Summary. Sex steroid hormones are involved in regulation of skin development and functions as well as in some skin pathological events. To determine the sites of action of estrogens, androgens and progestins, studies have been performed during the recent years to accurately localize receptors for each steroid hormone in human skin. Androgen receptors (AR) have been localized in most keratinocytes in epidermis. In the dermis, AR was detected in about 10% of fibroblasts. In sebaceous glands, AR was observed in both basal cells and sebocytes. In hair follicles, AR expression was restricted to dermal papillar cells. In eccrine sweat glands, only few secretory cells were observed to express AR. Estrogen receptor (ER) α was poorly expressing, being restricted to sebocytes. In contrast, ERß was found to be highly expressed in the epidermis, sebaceous glands (basal cells and sebocytes) and eccrine sweat glands. In the hair follicle, ERß is widely expressed with strong nuclear staining in dermal papilla cells, inner sheath cells, matrix cells and outer sheath cells including the buldge region. Progesterone receptors (PR) staining was found in nuclei of some keratinocytes and in nuclei of basal cells and sebocytes in sebaceous glands. PR nuclear staining was also observed in dermal papilla cells of hair follicles and in eccrine sweat glands. This information on the differential localization of sex steroid receptors in human skin should be of great help for future investigation on the specific role of each steroid on skin and its appendages.

Key words: Human skin, Sex steroid receptors Epidermis, Hair follicles, Sebaceous glands

Introduction

It is well known that sex steroids are involved in the regulation of several human skin functions, especially hair growth and sebaceous gland activity. The role of androgens in some abnormal conditions such as alopecia, acne and hirsutism has been extensively studied (Boudou et al., 1995; Chen et al., 2002). Estrogens have been recently shown to produce beneficial effects on the role and quality of cutaneous wound healing (Ashcroft et al., 1999). There is little information about the role of progesterone in skin functions. To identify in the skin the target structures and cells for sex steroids, one approach is to accurately localize receptors for each sex steroid using histological techniques, such as immunocytochemisty or in situ hybridization. The androgen receptor (AR), two estrogen receptor (ER) subtypes, ERα and ERß, and two progesterone receptor (PR) isoforms PRA and PRB have been cloned and fully characterized (Chang et al., 1988; Kastner et al., 1990; Mosselman et al., 1996). As far as we know, there has been no report on the localization of sex steroid receptors in the human skin as achieved by in situ hybridization. In the present review, we will summarize the available data on the localization of sex steroid receptors obtained by immunocytochemistry in the human skin.

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Androgen receptors (AR)

The androgen has been cloned in rat and human (Chang et al., 1988; Lubahn et al., 1988). Most of the polyclonal or monoclonal antibodies used for immunocytochemical localization have been developed against peptide sequence corresponding to a specific region of human AR. The localization of AR was very similar in the different skin areas, including the scalp (Blauer et al., 1991; Choudhry et al., 1992; Kimura et al., 1993; Liang et al., 1993; Bird et al., 1998). In the epidermis, AR immunoreactivity was detected in nuclei of interfollicular keratinocytes and pilose baceous duct keratinocytes . The stained nuclei were uniformly distributed throughout the different layers of the epidermis. In the dermis, about 10% of the fibroblasts exhibited AR immunostaining. In sebaceous glands, AR was expressed in nuclei of both basal cells and differentiated sebocytes (Fig. 1). In the hair follicle, no AR immunoreactivity could be detected in the epithelial cells of the outer root sheath including the bulge region

and the inner root sheath. On the other hand, the majority of dermal papillar cells expressed AR (Choudhry et al., 1992). In the sweat glands, Ar immunostaining was observed in nuclei of a small proportion of secretory cells (Fig. 2). No AR has been detected in blood vessels.

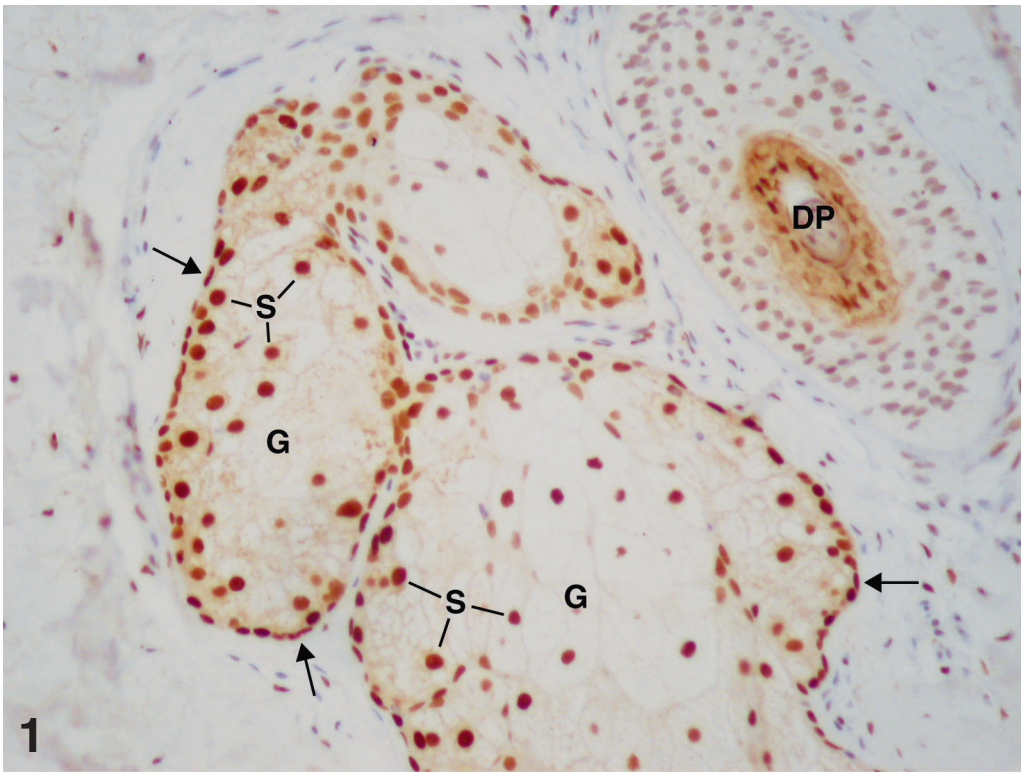

Fig. 1. Immunolocalization of AR in sebaceous gland (G) of human male forehead skin. Both basal cells (→) and sebocytes (S) exhibit nuclear staining. Nuclear staining can also be observed in dermal papilla (DP) cells in an adjacent hair follicle. x 380

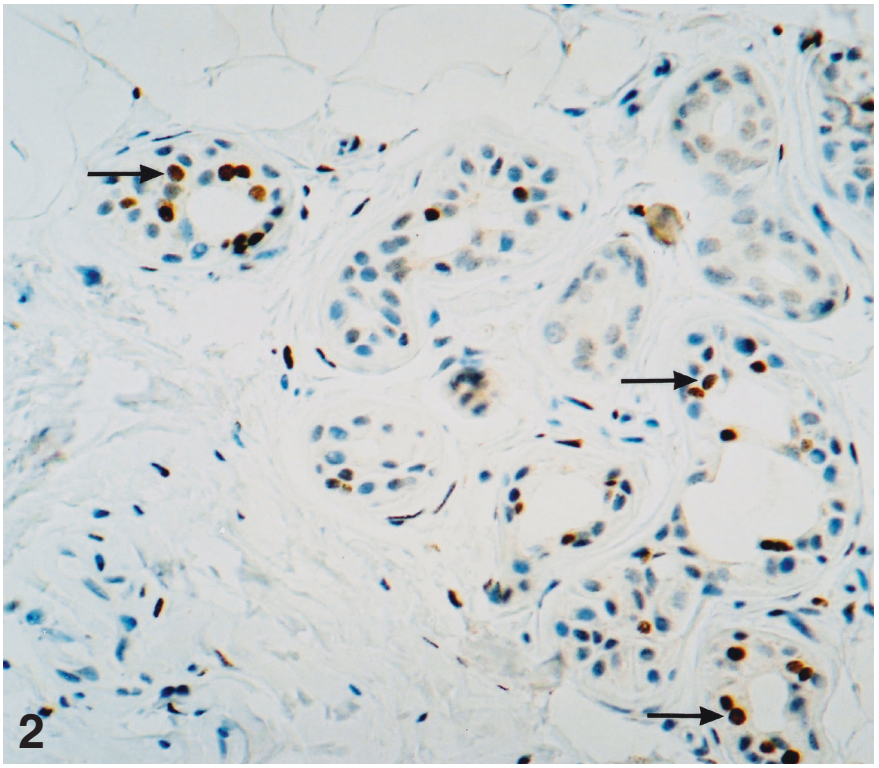

Fig. 2. Immunolocalization of AR in eccrine sweat glands of human male forehead skin. Nuclear staining is present in a few secretory cells (→). x 320

The expression of AR in the epidermis indicates that keratinocytes are target cells for androgen action in skin. The role of androgens on keranocytes activity remains to be clarified. The presence of AR-expressing keratinocytes within the pilosebaceous ducts support the hypothesis that androgens can directly influence direct keratinization in acne process (Choudhry et al., 1992).

It is well known that androgens stimulate cell proliferation and differentiation in rat and human sebaceous glands (Sweeney et al., 1969). Localization of AR in both basal and sebocytes in human sebaceous glands suggest that androgens may directly influence both cell proliferation and lipogenesis. The rate of sebum production is higher in adult men than in adult women (Cunliffe et al., 1969), but so far there has been no report on sex-related variations in sebaceous gland AR expression. An age-related decline in sebaceous gland AR has not been observed (Cunliffe et al., 1969), suggesting that the decrease in sebum production with age in both sexes is probably related to variations in circulating or locally produced androgens (Labrie et al., 2000) rather than in AR expression.

It has been reported that androgens influence pubertal development of sweat glands but do not regulate sweat secretion in adults (Rees and Shuster, 1981). The recent results indicating the expression of AR in some sweat gland cells in adults suggest that androgens might influence sweat gland activity throughout adult life.

Androgens are involved in the development of hair in the face and trunk as well as the sexual hair (Ebling, 1990). In hair follicles, AR expression has been detected only in the dermal papilla cells. These findings clearly indicate that the dermal papilla is the site of androgen action on human hair growth. The dermal papilla plays an essential role in the formation and maintenance of the germinative matrix of the hair follicle (Horne et al., 1986). The restriction of AR to the dermal papilla is indicative that androgens modulate interactions between mesenchymal (dermal papilla) cells and epithelial (matrix) cells. This may be analogous to androgen action in the embryonic prostate where AR expression is restricted to mesenchymal cells (Lasnitzki and Mizuno, 1980; Cooke et al., 1991; Choudhry et al., 1992)

Estrogen receptors (ER)

New knowledge about estrogen action has been advanced by the sequencing and cloning of the ER (Green et al., 1986) and of a new ER (Mosselman et al., 1996). The classical ER is referred to as ERα and the new one has been named ERß. Both ERα and ERß bind 17ß-estradiol with high affinity (Kuiper et al., 1997). It is likely that antibodies to ER previously used for immunocytochemical localization studies could recognize both receptor subtypes because of the close structural analogy between ERα and ERß. Using specific antibodies to ERα and ERß, we showed major differences in the tissue and cell distribution of ERα and ERß (Pelletier, 2000; Pelletier et al., 2000). Since the discovery of ERb, there have been only two reports on the specific localization of ERα and ERß in human skin (Pelletier, 2000; Thornton et al., 2003). All the results summarized below have been obtained by immunocytochemistry.

With antibodies to ER which were not described as being specific for either ERα or ERß, Esmaeli et al. (2000) reported that immunostaining was present in sebocytes in sebaceous glands in only 20% of the skin specimens while Maclean et al. (1990) found that the ER staining was restricted to dermal fibroblasts. On the other hand, Hodgins et al. (1998), also using not characterized ER antibodies, found no ER staining in female suprapubic skin.

Using specific antibodies to ERα and ERß, Thornton et al. (2003) have recently studied the distribution of both ER subtypes in male and female human scalp skin. No specific staining for ERα was observed in the epidermis (Fig. 3A). ERα immunoreactivity was found in sebaceous glands, particularly in partially differentiated sebocytes, but was almost undetectable in eccrine sweat glands. In the hair follicle, no specific nuclear staining was observed. In our own studies on ER immunolocalization in human skin, we did not find any structures exhibiting specific staining for ERα (Pelletier, 2000). In contrast, ERß was found to be highly expressed in the epidermis with all the keratinocyte layers being labelled (Fig.3B). In sebaceous glands, staining for ERß could be observed in differentiated sebocytes as well as basal cells. ERb is widely expressed in the hair follicle. Strong nuclear staining was detected in dermal papilla cells, inner sheath cells, matrix cells, and outer sheath cells including the buldge region (Fig. 4). As shown in Fig. 5, strong nuclear expression of ERß was found in eccrine sweat gland cells. Weak cytoplasmic staining was also consistently observed in these secretory cells. While AR and ERα staining was not expressed in blood vessels, strong staining for ERb has been observed in endothelial cells, muscle cells and fibroblasts in capillaries, veins and arteries. In these cell types, the expression of ERß was mostly found in nuclei with some weak cytoplasmic staining.

The recent ER localization studies have shown that ERß is widely expressed in human skin and its appendages, whereas ERα is only found in very few subsets of structures (Pelletier, 2000; Thornton et al., 2003). It has been previously shown that keratinocytes are estrogen-sensitive (Viae et al., 1989). Since only ERß is expressed in keratinocytes, it then appears that the effects of estrogens on the epidermis are mediated throughout activation of ERß. In the sebaceous glands, both basal cells and sebocytes express ERα and ERß with a much higher expression of ERß. In humans, sebum production is generally lower in females than males (Pochi and Strauss, 1974). Recent studies using human sebaceous glands in culture have shown that physiological levels of estradiol significantly decreased lipogenesis without affecting the rate of cell division

(Guy et al., 1996). The role of estrogens in the regulation of eccrine sweat glands which express only ERß is still unknown.

ERß protein is widely expressed in the hair follicle with strong immunostaining being observed in different compartments, including dermal papilla cells and cells of

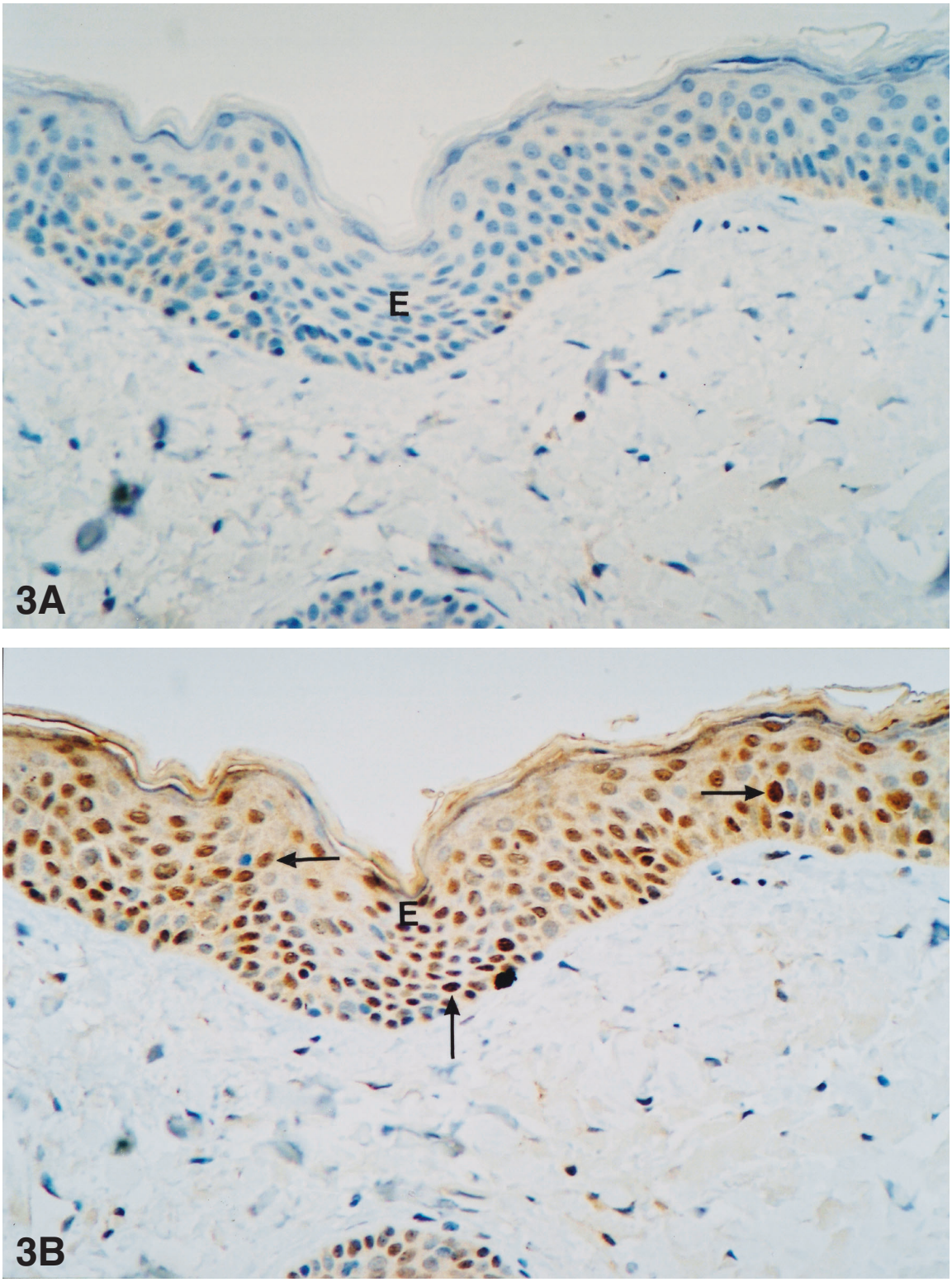

Fig. 3. Immunohistochemical localization of ERα **(A)** and ERß **(B)** in consecutive sections through the epidermis (E) of female abdominal skin. **A.** No ERα immunostaining can be detected. **B.** Nuclear ERß labelling is present in most of the keratinocytes. x 380

the specialized bulge of the outer root sheath. This bulge region contains the stem cells for hair follicles keratinocytes (Cotsarelis, 2000). It has been suggested that some skin carcinoma may originate from the bulge region and that estradiol might trigger the development of such tumors (Cotsarelis, 2000) Estrogens appear to be able to influence hair growth; for example, in pregnant women, there is a slower rate of replacement of spontaneous hair loss, possibly due to high levels of circulating estrogens. In mouse skin, it has been shown that follicular growth is blocked by estrogen, presumably by estrogen-related negative regulation of keratinocyte growth factor expression in the dermal papilla (Oh and Smart, 1996). The direct effect of estrogens on hair

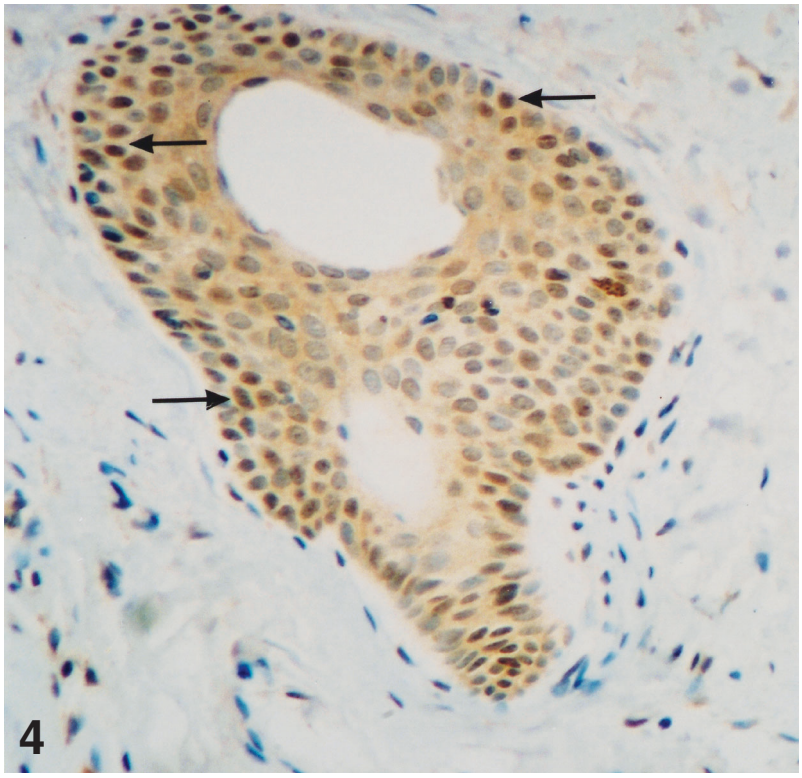

Fig. 4. Immunohistochemical localization of ERß in a hair follicle of female abdominal skin. Nuclear staining is observed in the majority of cells of the external root sheath. (→) x 380

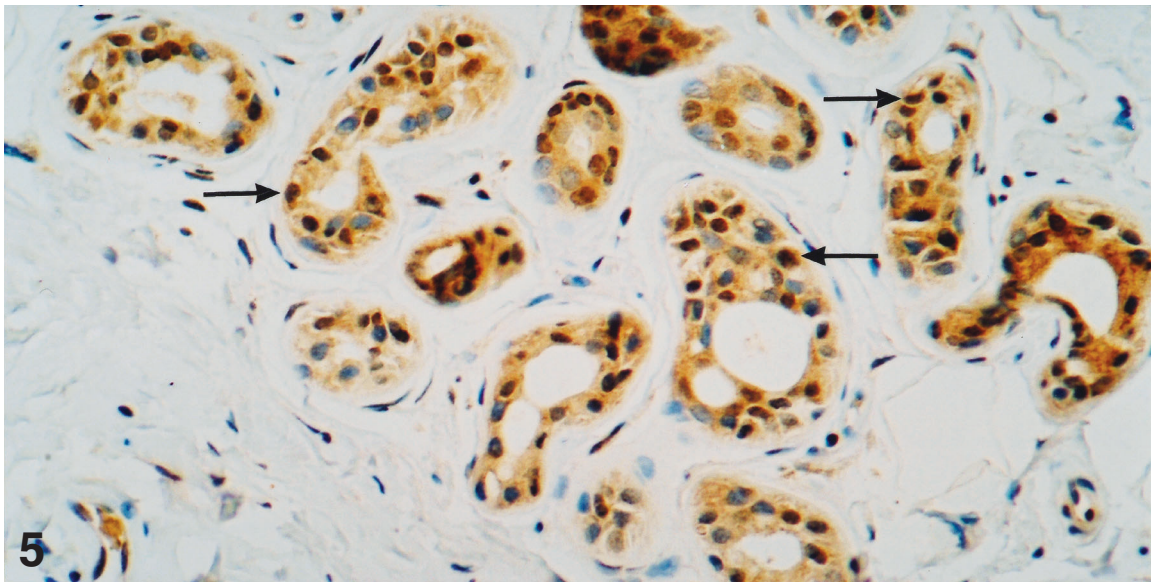

Fig. 5. Immunolocalization of ERß in eccrine sweat glands. Strong nuclear staining is seen in the majority of secretory cells. Cytoplasmic staining is also present in the same cells. x 380

growth is likely mediated by ERß.

Progesterone receptor (PR)

PR is a dimer composed of two receptor proteins, PRA and PRB, which each binds progesterone. In a recent extensive review in the physiological role of progesterone, Graham and Clarke (1997) reported the expression of PR in a large variety of tissues and cells, but there was no mention that skin could express PR. By radio-receptor assay, PR has already been detected in pubic hair in pre- and postmenopausal women (Schmidt et al., 1990). So far there have been very few studies on the immunolocalization of PR in human skin and in most of them no PR immunostaining has been observed (Hodgins et al., 1998; Kohlberger et al., 1998; Esmaeli et al., 2000). In fact, the only positive report came from (Wallace and Smaller, 1998) who found nuclear and cytoplasmic staining for PR in eccrine sweat cells and sebocytes in sebaceous glands. In 30% of cases of

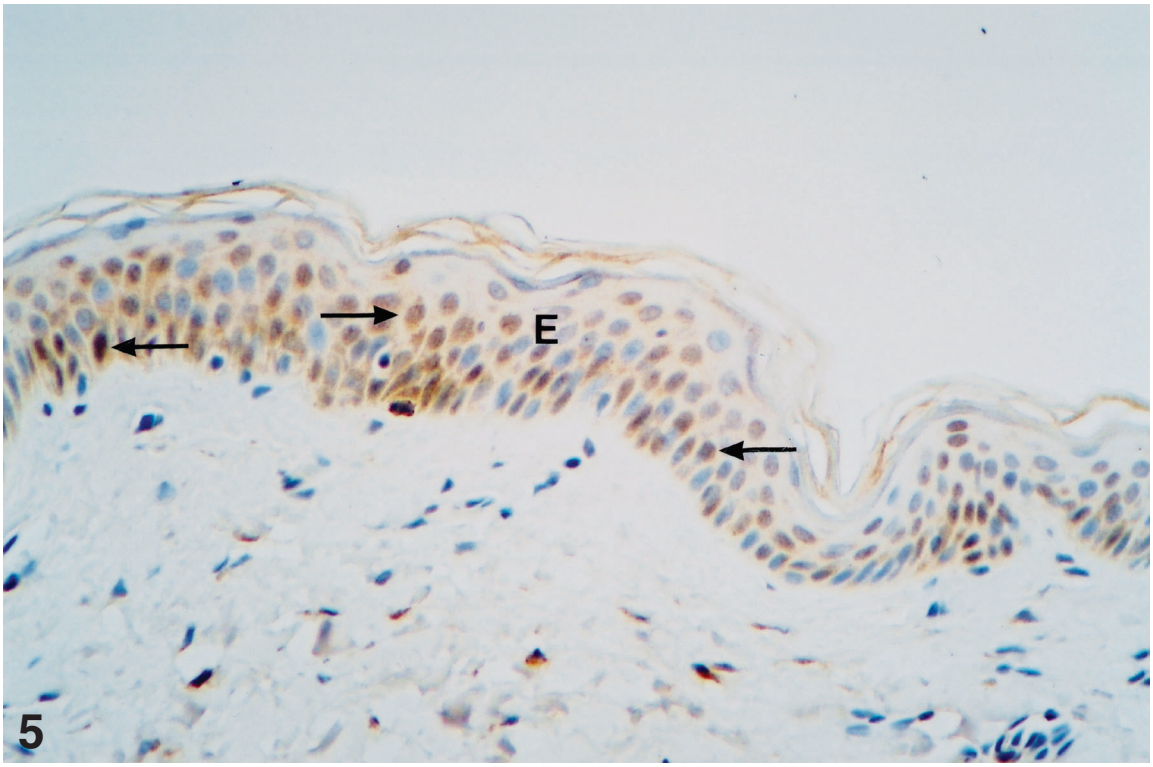

Fig. 6. Immunohistochemical localization of PR in the epidermis of male forehead skin. Nuclear staining is observed in keratinocytes (Æ) throughout the different layers of the epidermis (E). x 380

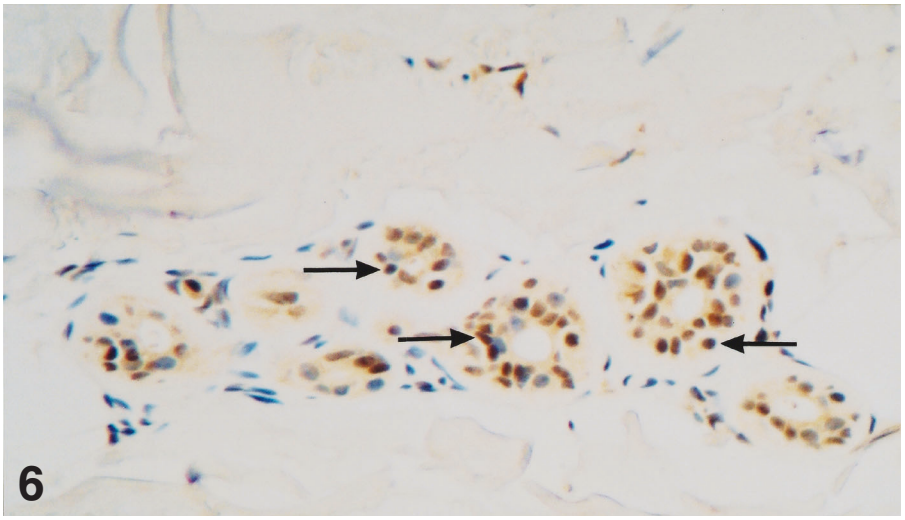

Fig. 7. Immunohistochemical localization of PR in the eccrine sweat glands of male forehead skin. Nuclear staining can be seen in most secretory cells. x 320

androgenic alopecia, they observed nuclear and cytoplasmic staining for PR within the dermal papilla of hair follicles. Using antibodies which react with both PR-A and PR-B, we recently found PR staining in nuclei of some keratinocytes throughout all the layers of the epidermis (Fig. 6) and also in nuclei of basal cells and sebocytes in sebaceous glands. Nuclear staining was also observed in eccrine sweat gland cells (Fig. 7). In hair follicles, we did not detect any PR immunoreactivity.

The physiological of progesterone on skin and its appendages remains to be elucidated. It has been reported that chronic administration of progesterone suppressed estrogen receptor concentration in the monkey skin (West et al., 1990). We have recently observed that 20α-hydroxysteroid dehydrogenase the enzyme which catalyzes the conversion of progesterone into its inactive form 20α-hydroxyprogestgerone was highly expressed in sebaceous glands in the mouse skin (Pelletier et al., unpublished data). The enzyme could regulate the availability of circulating progesterone for PR and thus control the influence of progesterone on sebaceous gland cell activity.

Concluding remarks

It has been long known that sex steroid hormones display distinct influences on the various structural elements of the skin. Recent immunocytochemical studies, especially those concerning ERß localization have contributed to establish the localization of steroid receptors in human skin and its appendages, including hair follicles, sebaceous glands and eccrine sweat glands. It now clearly appears that the epidermis, dermis as well as skin appendages are all expressing receptors for the major sex steroids, namely estrogens, androgens and progestins. It is of interest to note that, in the different skin compartments, there is generally a differential expression of each receptor. Such a specific localization can be related to the different effects exerted by each hormone. These results provide information for future investigation on the roles of each steroid on skin functions.

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