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An estrogen receptor pathway regulates the telogen–anagen hair follicle transition and influences epidermal cell proliferation

(epidermis/estrogen/keratinocyte/differentiation/mesenchymal epithelial interactions)

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ABSTRACT **The hair follicle is a cyclic, self renewing epidermal structure which is thought to be controlled by signals from the dermal papilla, a specialized cluster of mesenchymal cells within the dermis. Topical treatments with 17-β-estradiol to the clipped dorsal skin of mice arrested hair follicles in telogen and produced a profound and prolonged inhibition of hair growth while treatment with the biologically inactive stereoisomer, 17-α-estradiol, did not inhibit hair growth. Topical treatments with ICI 182,780, a pure estrogen receptor antagonist, caused the hair follicles to exit telogen and enter anagen, thereby initiating hair growth. Immunohistochemical staining for the estrogen receptor in skin revealed intense and specific staining of the nuclei of the cells of the dermal papilla. The expression of the estrogen receptor in the dermal papilla was hair cycle-dependent with the highest levels of expression associated with the telogen follicle. 17-β-Estradiol-treated epidermis demonstrated a similar number of 5-bromo-2′-deoxyuridine (BrdUrd) S-phase cells as the control epidermis above telogen follicles; however, the number of BrdUrd S-phase basal cells in the control epidermis varied according to the phase of the cycle of the underlying hair follicles and ranged from 2.6% above telogen follicles to 7.0% above early anagen follicles. These findings indicate an estrogen receptor pathway within the dermal papilla regulates the telogen–anagen follicle transition and suggest that diffusible factors associated with the anagen follicle influence cell proliferation in the epidermis.**

The hair follicle cycle is characterized by a period of follicle growth (anagen), followed by period of degeneration and rearrangement (catagen), and finally by a resting period (telogen) (1, 2). The hair follicle is a self-renewing system and is therefore governed by a slow cycling stem cell (1–5). Recently, the bulge activation hypothesis has been proposed which states that the follicular stem cell resides in the bulge area of the permanent portion of the hair follicle and that this stem cell is stimulated during early-anagen to divide and produce transient amplifying stem cells (4). It appears that a small group of highly specialized mesenchymal cells referred to as the dermal papilla provides the signal that initiates anagen and instructs the bulge follicular stem cell to divide (3, 4, 6–8). The transient amplifying stem cells or matrix cells proliferate and then differentiate under the influence of unidentified morphogens into the inner root sheath cells (cuticle, Huxley's and Henle's layers) and medulla and cortex cells, which together with the cuticle cells terminally differentiate into the mature hair fiber (3, 4, 6). The matrix cells have a finite life span and are thought to terminally differentiate as the follicle enters catagen. After degeneration of the lower follicle, the follicle enters telogen and remains in telogen until the dermal papilla signals the bulge stem cells to divide and the hair follicle cycle begins again. While the dermal papilla appears to be critical in the regulation of hair follicle cycle (8, 9) the actual signals that initiate and terminate the cycles of hair growth as well as those that induce the differentiation of the matrix cells remain poorly understood.

Developmental as well as recent transgenic studies in mice have identified a number of growth factors and growth factor receptors that appear to be important in morphogenesis and development of the hair follicle (10–21). While these factors influence morphogenesis and development of the follicle, the factors that initiate and regulate the cycle itself remain to be elucidated. A variety of steroid hormones can influence hair growth; for example, corticosteroids can inhibit hair growth while androgen treatment in man can stimulate hair growth in certain parts of the body and paradoxically also induce baldness (7). Testosterone and dihydrotestosterone have been extensively studied with regard to their involvement in male pattern alopecia; however, the role of androgens in the hair follicle cycle remains elusive. Estrogen appears to be able to influence hair growth; for example, pregnant women demonstrate a slower rate of replacement of spontaneous hair loss or plucked hair, presumably due to high levels of circulating estrogen (6). In rats, multiple subcutaneous injection of estradiol benzoate retards hair growth and reduces the thickness of rat skin as well as the size of the sebaceous gland (22). Whether these effects are directly mediated by estradiol within the follicle or are of an indirect nature through estrogen-induced modulation of pituitary hormones, for example, is not known. However, the hair follicle itself can form significant quantities of estrogens, particularly estrone from androstenedione and testosterone (23) and based on Scatchard analysis in human and mouse skin and ^{3}H-estrogen binding in rat skin, it appears that estrogen receptors are present in skin (24–26). Therefore, we have explored the role of estrogen and the estrogen receptor in the hair follicle cycle.

MATERIALS AND METHODS

Topical Treatment with Estrogen and ICI 182,780 and Determination of Hair Follicle Phase. Female CD-1 mice, 4 or 5 weeks of age, were purchased from Charles River Breeding Laboratories. Mice were fed rodent chow (Agway Food, Granville Milling, Creedmoor, NC) and water ad libitum. The hair on dorsal region (≈4 × 2.5 cm area) was clipped with electric clippers. For hair growth studies, the mice were treated on the clipped dorsal surface with 10 nmol 17-β-estradiol, 10 nmol 17-α-estradiol, an inactive stereoisomer, or 10 nmol ICI 182,780, a pure estrogen receptor antagonist (Zeneca Pharmaceuticals, Cheshire, U.K.) in 200 μl acetone or acetone alone twice weekly for up to 16 weeks of age. Full hair regrowth is defined as the complete growth of hair over the entire clipped dorsal surface. For histological studies, additional groups of mice were treated as described above and every week

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Abbreviation: BrdUrd, 5-bromo-2′-deoxyuridine.

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three mice from each group were killed by cervical dislocation and the dorsal skin was excised. The dorsal skin was fixed for 24 h in a 10% neutral buffered formalin, processed, and embedded in paraffin. Sections (≈5 μm) were bichromically stained with hematoxylin/eosin for histological examination, and the specific phase of the hair follicle cycle was determined according to Dry (1) and Ebling *et al.* (7). We have used the terms early-anagen to refer to anagen I and II, mid-anagen to refer to angen III and IV, and late-anagen to refer to anagen V and VI. Additional treatment groups received BrdUrd by i.p. injection 1 h before killing the mice.

Localization of Estrogen Receptor in Mouse Skin. The dorsal skin area from 6- to 12-week-old untreated mice was excised and fixed for 24 h in a cold 10% neutral buffered formalin, then changed to cold 70% ethanol and processed and embedded in paraffin. Mouse uterus was collected and used as a positive control for estrogen receptor staining. Tissue sections (5 μm) were deparaffinized and placed in 3% H_2O_2 for 10 min to quench the endogenous peroxidase activity and then washed with automation buffer (Biomedia, Foster City, CA). The sections were treated with trypsin (0.15 mg/ml in automation buffer) for 4 min at room temperature followed by two washes with automation buffer and then incubated with DNase (0.25 mg/ml in automation buffer) for 3 min at room temperature followed by two washes with automation buffer. The sections were blocked with 10% normal goat serum and incubated with the prediluted ER-ICA, an anti-estrogen receptor rat monoclonal antibody (Abbott), overnight at 4°C. Slides were washed twice with automation buffer and incubated with biotinylated goat anti-rat IgG (Boehringer Mannheim) at a dilution of 1:50 for 60 min at room temperature. After washing with automation buffer twice, the sections were incubated with horseradish peroxidase-conjugated streptavidin (1:20 dilution, BioGenex Laboratories, San Ramon, CA) for 30 min at room temperature. The samples were washed with automation buffer followed by 0.05 M Tris·HCl (pH 7.5) and incubated with 3,3′-diaminobenzidine tetrahydrochloride for 10 min in the dark. Slides were rinsed with 0.05 M Tris·HCl buffer and some samples were counterstained in hematoxylin. Samples were dehydrated in a graded series of ethanol and xylene and permanently mounted with Permount. Uterine cells demonstrated characteristic nuclear staining. However, no estrogen receptor staining was observed when control antibody was used or when the primary antibody was omitted in both skin and uterine samples. In addition, we utilized another monoclonal antibody to the estrogen receptor, ER1D5 (Immunotech, Westbrook, ME) and obtained the same results as with ER-ICA.

BrdUrd Staining and Counting Methods. Mice received BrdUrd (100 mg/kg body weight in 200 μl PBS) administered by i.p. injection 1 h before killing the mice. Skin sections were stained for BrdUrd according to Eldridge *et al.* (27). Skin sections were deparaffinized in xylene, passed through graded alcohol, incubated in 2 M HCl for 30 min at 37°C, and washed in borate buffer for 3 min at room temperature. The sections were digested with 0.001% trypsin for 3 min at 37°C and then washed in distilled water for 1 min. Endogenous peroxidase was inhibited by preincubation with 3% H_2O_2 for 10 min. Slides were then washed with automation buffer for 5 min. The sections were blocked with horse serum for 20 min and incubated with primary antibody, anti-BrdUrd IgG (Becton Dickinson) at a dilution of 1:25 (diluted in 1% bovine serum albumin in automation buffer) for 1 h at room temperature. Slides were washed twice with automation buffer for 5 min and further processed using a Vectastain Elite mouse ABC kit (Vector Laboratories). Samples were then processed as described for estrogen receptor immunohistochemistry. At least 1000 interfollicular basal cells above phase synchronized hair follicles were counted per slide.

RESULTS

To determine if 17-β-estradiol could influence hair growth, the dorsal hair of 6-week-old CD-1 female mice was clipped with electric clippers and the dorsal surface treated twice weekly with topical applications of either 10 nmol 17-β-estradiol, 10 nmol 17-α-estradiol, an inactive stereoisomer, or acetone vehicle from the 6th week of age to the 16th week of age. Treatment with 17-β-estradiol had a potent inhibitory effect on dorsal hair growth. As shown in Fig. 1, 100% of the acetone-treated mice as well as the mice treated with the inactive stereoisomer, 17-α-estradiol, demonstrated full hair regrowth by 13 weeks of age. In contrast, mice treated with 17-β-estradiol did not demonstrate any full hair regrowth by 16 weeks of age. Some of the 17-β-estradiol-treated mice did demonstrate a partial patchy hair regrowth that involved less than 10% of the total clipped area. At 12 weeks of age, 20% of the 17-β-estradiol-treated mice demonstrated such partial patchy hair regrowth and by 15 weeks of age 40% of the 17-β-estradiol-treated mice demonstrated such partial patchy hair regrowth. Treatment of older mice with 17-β-estradiol also blocked hair regrowth (data not shown). These results demonstrate that topical treatment with 17-β-estradiol potently blocks hair growth.

The hair follicle cycles of mice are highly synchronized from birth to 12 weeks with temporally fixed peroids of anagen, telogen, and catagen (28). The second syncronized telogen phase in CD-1 mice begins at ≈6 weeks of age and lasts until ≈9 weeks of age at which time the hair follicles synchronously enter the third anagen. Histological analysis was conducted on mouse skin from mice treated twice weekly with 17-β-estradiol or acetone from the 4th week of age to the 15th week of age. Each week, skin samples from three mice from each group were collected for histology. Representative skin histology sections from acetone- and 17-β-estradiol-treated mice at 7, 9, and 11 weeks of age are shown in Fig. 2. At 7 weeks of age all follicles were in telogen in both the acetone- and the 17-β-estradiol-treated mice, as this age represents a period of synchronized telogen (Fig. 2 *A* and *B*). By 9 weeks of age the hair follicles of the acetone control mice were in early-anagen, while the hair follicles of the 17-β-estradiol mice remained arrested in telogen (Fig. 2 *C* and *D*). By 11 weeks of age the hair follicles of the acetone treated mice were all in mid- to late-anagen or catagen while the hair follicles of the 17-β-estradiol-treated mice remained in telogen (Fig. 2 *E* and *F*). At the termination of the experiment, when the mice were 15 weeks of age, the hair follicles of the 17-β-estradiol-treated mice continued to be arrested in telogen (data not shown). These data demonstrate that the topical application of 17-β-

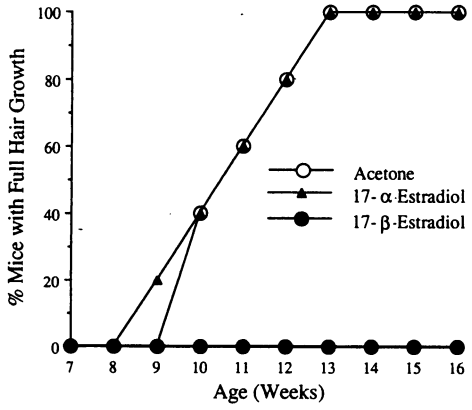

FIG. 1. Topical 17-β-estradiol blocks hair regrowth in mice. Six-week-old mice (5 mice/group) were treated on the clipped dorsal surface twice weekly with topical applications of 10 nmol 17-β-estradiol, 10 nmol 17-α-estradiol, or acetone vehicle for 10 weeks.

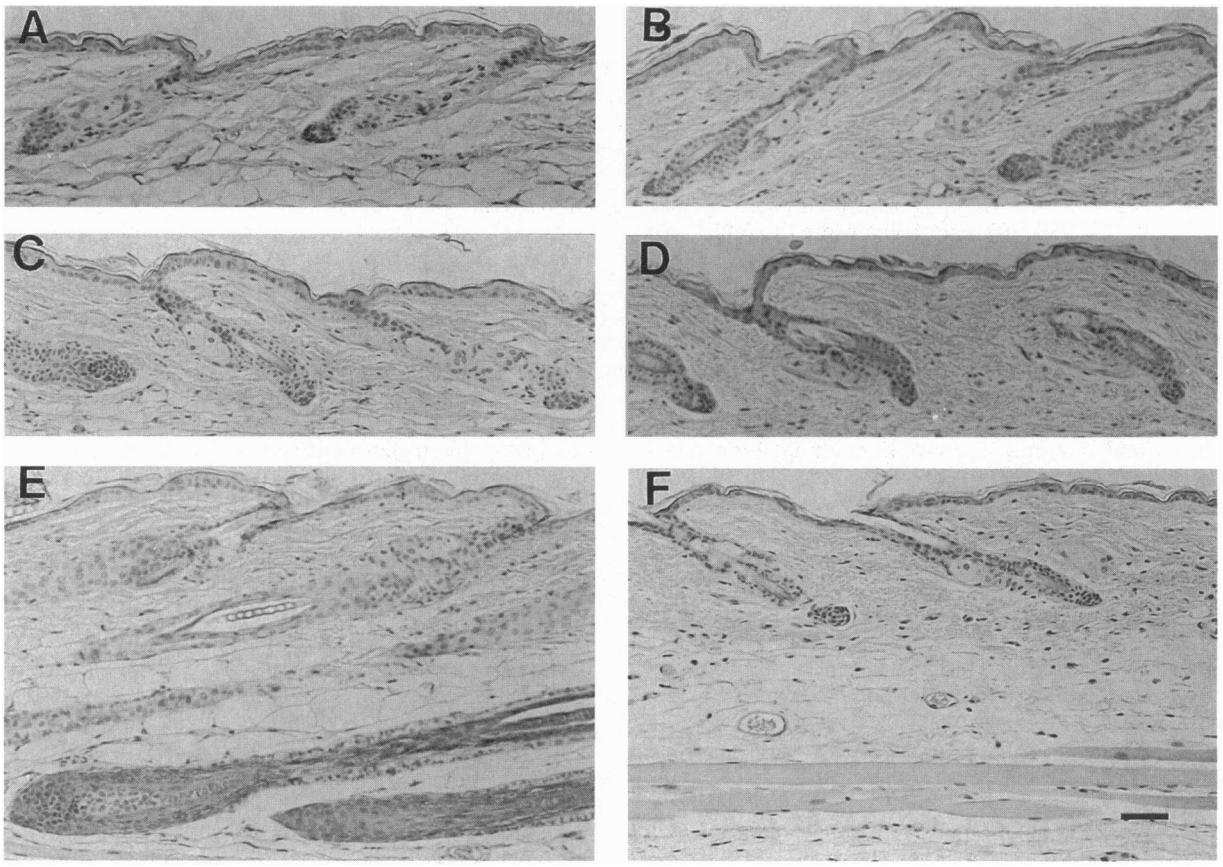

FIG. 2. Topical 17-β-estradiol arrests the follicle in telogen. Four-week-old mice were treated on the clipped dorsal surface twice weekly with 10 nmol 17-β-estradiol or acetone up to 11 weeks. Histological samples were prepared from three mice per group per week: (*A*) 7-week-old acetone-treated mouse, (*B*) 7-week-old 17-β-estradiol-treated mouse, (*C*) 9-week-old acetone-treated mouse, (*D*) 9-week-old 17-β-estradiol-treated mouse, (*E*) 11-week-old acetone-treated mouse, and (*F*) 11-week-old 17-β-estradiol-treated mouse. (Bar = 50 μm.)

estradiol prevents hair growth by arresting the hair follicle in the telogen phase of the hair cycle.

To begin to determine a potential role for the the estrogen receptor in the observed effects on hair growth, mice were treated with the pure estrogen receptor antagonist ICI 182,780. Twice weekly treatment with 10 nmol ICI 182,780 or acetone alone to the clipped dorsal skin was begun when the mice were 6 weeks of age. By 9 weeks of age, 70% of the ICI 182,780-treated mice demonstrated full hair regrowth while no hair growth was observed in the acetone treated mice (Figs. 3 and 4). By 10 weeks of age all of the ICI 182,780-treated mice developed a full coat of hair while only 20% of the acetone-treated mice demonstrated full hair regrowth. At this time the ICI 182,780-treated mice were visually indistinguishable from mice whose hair was not previously clipped. These data indicate that ICI 182,780 caused the telogen follicle to enter anagen during what should have been the second synchronized telogen phase. To confirm this notion, skin was collected and prepared for histological analysis at 7, 8, 9, 10, and 11 weeks. At 7 weeks of age, the ICI 182,780-treated mice demonstrated

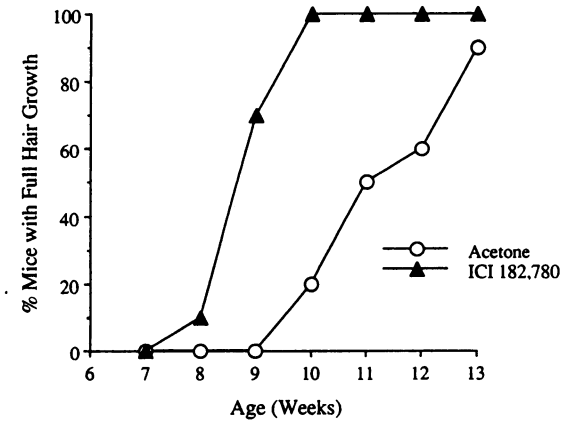

FIG. 3. Estrogen receptor antagonist ICI 182,780 induces hair growth. Six-week-old mice (10 mice per group) were treated on the clipped dorsal surface twice weekly with 10 nmol ICI 182,780 or acetone for 7 weeks.

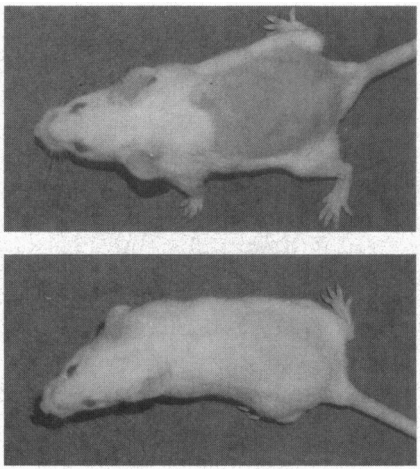

FIG. 4. Characteristic example of the effect of estrogen receptor antagonist ICI 182,780 on hair growth. Mice were treated as in Fig. 3. (*Upper*) Nine-week-old acetone-treated mouse. (*Lower*) Nine-week-old ICI 182,780-treated mouse.

follicles that were already in early- to mid-anagen while the follicles of acetone-treated mice were synchronized in second telogen (Fig. 5 *A* and *C*). By 8 weeks of age the follicles of ICI 182,780-treated mice were in mid- to late-anagen while the hair follicles of the acetone-treated mice were in telogen (Fig. 5 *B* and *D*). These results indicate that the estrogen receptor antagonist ICI 182,780 initiates the transition of a telogen follicle into anagen. At 11 weeks of age, the follicles of mice treated with ICI 182,780 entered telogen while the follicles of control mice were in mid- to late-anagen or catagen (data not shown) indicating that ICI 182,780 does not prolong anagen. Intraperiteneal injection of ICI 182,780 at a dose equal to that used in the dermal application was without effect on hair growth indicating that the ICI 182,780 effect was within the skin and was not of a systemic nature.

Immunohistochemical staining for the estrogen receptor in mouse skin revealed intense and specific staining of the nuclei of cells within the dermal papilla of a telogen follicle as shown in the counterstained sample (Fig. 6*A*). Noncounterstained samples (Fig. 6*B*) are provided to better demonstrate the areas and levels of estrogen receptor expression. Estrogen receptor expression within most telogen dermal papillas demonstrated a polarity, as nuclei of cells within the lower half of the dermal papilla stained intensely while very little staining was observed in the upper half of dermal papilla. Male mice also demonstrated intense estrogen receptor staining in the nuclei of the dermal papilla of the telogen follicle (data not shown). In addition, the expression of the estrogen receptor was hair cycle dependent as there was weaker staining of the dermal papilla of early-anagen follicles (Fig. 6 *C* and *D*) and no detectable staining in dermal papilla of mid- to late-anagen (Fig. 6 *E* and *F*) or catagen follicles (data not shown). Very light estrogen receptor staining was observed in the cells of the outer root sheath in the isthmus of the telogen follicle as well as in some nuclei of dermal fibroblasts.

In the course of these studies we observed that the epidermis of 17-β-estradiol-treated mice was significantly thinner ($P < 0.01$; 10.1 ± 1.8 μm) than the acetone-treated mice (14.1 ± 1.3 μm). To determine if 17-β-estradiol inhibited cell proliferation in mouse epidermis, 4-week-old mice were treated with 17-β-estradiol or acetone alone twice weekly until the mice were 15 weeks of age. Every 3–7 days histological samples from each group were prepared and immunohistochemical staining for BrdUrd was conducted. Each sample was evaluated for the phase of the hair follicle and the number of interfollicular basal epidermal cells in S-phase was determined based on BrdUrd immunohistochemical staining. Accurate quantitation of BrdUrd S-phase-positive cells within the hair follicle itself was not possible due to the fact that the entire follicle was not always in the same plane of section. Epidermal samples from 17-β-estradiol-treated mice demonstrated a similar number of BrdUrd S-phase-positive interfollicular basal epidermal cells as the interfollicular epidermis from vehicle-treated control mice whose follicles were in telogen (Table 1). When intact telogen follicles from acetone- or 17-β-estradiol-treated mice were observed in the plane of the section, very few BrdUrd S-phase-positive cells were observed in the follicles (data not shown). In vehicle-treated control skin the number of BrdUrd S-phase positive cells in the basal cells of epidermis increased from 2.6% above telogen follicles to 7% in the epidermis above early-anagen follicles and decreased to 4.9 and 3.8% above mid- and late-anagen follicles, respectively (Table 1). When intact anagen follicles were observed many BrdUrd S-phase-positive cells could be observed in the bulb area and outer root sheath. These results indicate that the phase of the underlying hair cycle influences the labeling index of the basal cells of the epidermis and suggest that diffusible factors associated with the anagen follicle influence the proliferation of basal cells in the epidermis.

DISCUSSION

The growth and cycling of the mature hair follicle is regulated through complex and intricate interactions between the epithelial cells of the follicle and mesenchymal cells of the dermal papilla (3, 4, 6–9). Implantation studies with dermal papilla or cultured dermal papilla cells have demonstrated that these papilla can initiate the growth of deactivated follicles and specify the type of hair fiber produced (8, 29); however, the nature of the dermal papilla-derived signals remain unknown. It has been suggested that diffusible factors derived from the dermal papilla or surface molecules on the dermal papilla cells regulated the follicle cycle as well as the differentiation of the matrix cells (3, 4, 6–9, 29). While numerous growth factors can

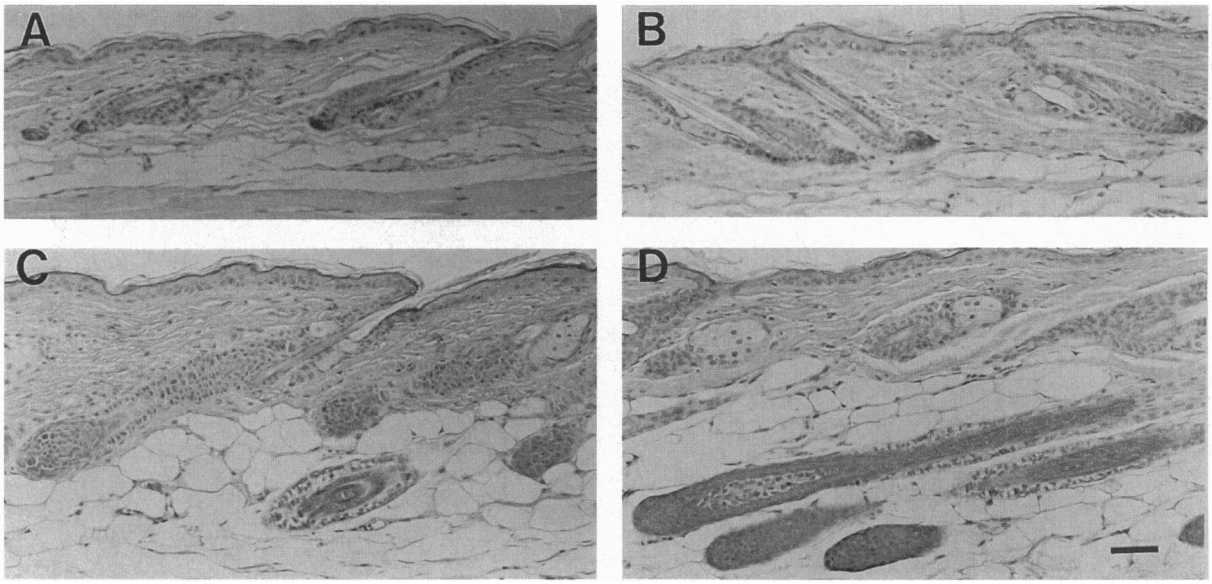

FIG. 5. Estrogen receptor antagonist ICI 182,780 initiates entry into anagen. Six-week-old mice were treated on the clipped dorsal surface twice weekly with 10 nmol ICI 182,780 or acetone for 5 weeks. Histological samples were prepared from three mice per group per week: (*A*) 7-week-old acetone-treated mouse, (*B*) 8-week-old acetone-treated mouse, (*C*) 7-week-old ICI 182,780-treated mouse, and (*D*) 8-week-old ICI 182,780-treated mouse. (Bar = 50 μm.)

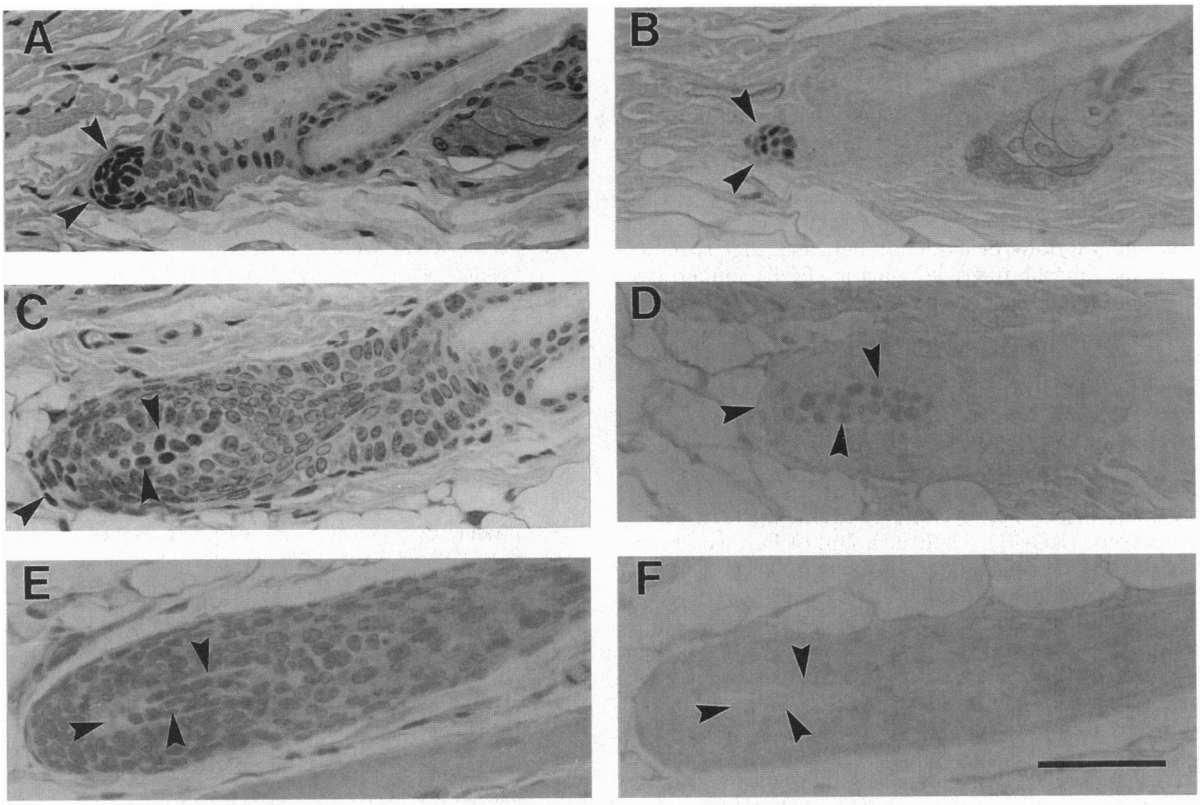

FIG. 6. Immunohistochemical localization of estrogen receptor in mouse skin. Estrogen receptor immunohistochemical staining was conducted with or without hematoxylin counterstaining. (*A*) Telogen hair follicle with counterstaining. (*B*) Telogen hair follicle without counterstaining. (*C*) Early-anagen hair follicle with counterstaining. (*D*) Early-anagen hair follicle without counterstaining. (*E*) Late-anagen hair follicle with counterstaining. (*F*) Late-anagen follicle without counterstaining. Arrowheads point to the dermal papilla cells. (Bar = 50 μm.)

influence the development of the hair follicle (10–21), much less is known about the dermal papilla-derived signals that initiate and terminate the hair cycle and how the dermal papilla itself is regulated. Our results provide evidence that an estrogen receptor pathway within the dermal papilla regulates the telogen–anagen transition of the hair follicle. This conclusion is based on following experimental results: (*i*) 17-β-estradiol blocks hair growth and arrests hair follicles in the telogen phase of the hair cycle; (*ii*) 17-α-estradiol, an inactive stereoisomer, does not block hair growth; (*iii*) estrogen receptor antagonist ICI 182,780 causes the hair follicle to exit telogen and enter anagen thereby initiating hair growth; and (*iv*) the estrogen receptor is expressed in skin and its expression is localized to the nuclei of cells of the dermal papilla of the telogen follicle. The fact that estrogen is produced by the hair follicle itself (23) and the estrogen receptor is predominately expressed in the dermal papilla suggest that estrogen is an endogenous paracrine regulator of hair follicle cycle. The identification of regulatory molecules such as estrogen and estrogen receptor antagonists that modulate the transition from a telogen to anagen follicle should aid in the delineation of the intricacies of mesenchymal/epithelial interactions of the follicle and allow for the development of effective pharmacological agents to treat male pattern alopecia, diffuse alopecia, chemotherapy-induced alopecia, as well as hirsutism. In addition, an understanding of the signals that regulate the follicular stem cell could have important implications for cutaneous neoplasia, as the follicular stem cell may represent the target cell for chemical carcinogens and the precusor cell of skin cancer (30–33).

Table 1. Epidermal cell proliferation is influenced by the phase of the underlying hair follicle

Hair follicle phase (*n*)	BrdUrd S-phase cells/1000 basal epidermal cells (%)
17-β-Estradiol-treated skins (4)	37 ± 6 (3.7)
Vehicle-treated skins telogen (14)	26 ± 10 (2.6)
Early-anagen (14)	70 ± 12* (7.0)
Mid-anagen (5)	49 ± 16* (4.9)
Late-anagen (3)	38 ± 19 (3.8)

At least 1000 interfollicular basal epidermal cells were counted per skin sample. Values represent the mean ± SD. The number of skins examined is indicated by the number enclosed by parentheses.
*Value is significantly different from telogen value at $P < 0.01$.

The cyclic nature of estrogen receptor expression in the dermal papilla indicates that additional factors must control the expression of the estrogen receptor itself. The estrogen receptor is detected at its highest level in the dermal papilla of a telogen follicle, which is consistent with its role in telogen arrest. Earlier investigations found that testosterone and estrogen bound to proteins in the supernatant fraction of rat skin and this binding varied during the hair follicle cycle (34). The factors that regulate the expression of the estrogen receptor are not known. Perhaps signals originating from the epithelial cells of the early- to mid-anagen follicle may be important in the down-regulation of estrogen receptor expression in manner analogous to the interactions that occur when the papilla is activated (4), or perhaps estrogen synthesis within the follicle itself is cyclic and can influence the expression of the estrogen receptor. Further studies will be required to understand the complexities of regulation of the receptor within the dermal papilla.

Due to the early synchronous nature of the mouse hair follicles between 6 and 12 weeks we were able to observe that the phase of the underlying hair follicle dramatically correlates with the proliferation of basal epidermal cells. These obser-

vations suggest the existence of a diffusible factor(s) that simultaneously induces proliferation of the matrix cells of the hair follicle as well as the proliferation of basal cells in the interfollicular epidermis. Since these two proliferative events, follicular growth and epidermal cell proliferation, are coordinated and both are blocked in estrogen-treated mouse skin suggests that estrogen may negatively regulate the putative dermal papilla diffusible factor or that the anagen follicle itself is producing a mitogenic stimulus.

An accumulating body of evidence indicates that specific members of the fibroblast growth factor (FGF) family are critical in hair follicle development, epidermal differentiation and proliferation, and wound repair (10–17). Acidic FGF (aFGF), basic FGF (bFGF), and FGF-5 and their receptors have been localized to the hair follicle, or immediately adjacent to the hair follicle, and each of these FGFs have been shown to inhibit hair growth and follicle development (10–14). FGF-7 or keratinocyte growth factor (KGF) expression has been localized to the dermis (35) and dermal papilla (17), and exogenously administered KGF stimulates follicle growth (17). The KGF receptor (KGFR), a splice variant of FGF receptor 2 has been localized to the basal and suprabasal keratinocytes as well as the hair follicle and dermal papilla (17, 36, 37). Studies in transgenic mice utilizing a keratin K14 promoter to target the expression of KGF to the stratified epithelium of mice demonstrated a hyperthickening of the epidermis and alterations in hair follicle morphogenesis (15). More recently, studies utilizing transgenic mice that contain a dominant-negative KGFR under the regulation of K14 promoter demonstrated a greatly reduced proliferation rate in the epidermis and abnormalities in the morphology of the hair follicle with hair growth retardation (16). These reported effects of the dominant-negative KGFR as well as the effects of exogenously administered KGF are similar to what we have observed in estrogen-treated and estrogen antagonist-treated mice, respectively, and suggest that estrogen may negatively regulate KGF expression in the dermal papilla. Other dermal papilla derived signals which are possible targets for estrogen regulation include hepatocyte growth factor (38), cell adhesion proteins (3), and extracellular matrix components (39).

While further studies are required to discern the downstream pathway through which the estrogen receptor regulates the telogen–anagen transition, our study provides new fundamental insights into the role of estrogen and the estrogen receptor in mesenchymal/epithelial interactions that are critical in the regulation of the hair follicle cycle.

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