done based on the assumption that the relative position of the bulge along the length of dissected follicles is constant. However, as shown in **Fig 1**, neighboring follicles whose bulges are located in a similar depth can have lower follicles of widely varied length. Another question is: if the stem cells are below the bulge, can one identify them morphologically? As can be seen in **Fig 1**, the outer root sheath cells located below the bulge are morphologically uniform and there is, so far, no evidence for cellular heterogeneity in this area. One also needs to know how the stem cells located in the transient portion of the follicle escape destruction during telogen. Finally, if the stem cells are below the bulge, what kind of microenvironmental heterogeneity may account for the stem cell "niche"? Thus, overall, Rochat's [12] data are largely consistent with ours in emphasizing the importance of keratinocytes in the vicinity of the bulge. The difference in the detailed localization may have arisen from the difficulty in localizing precisely the bulge zone by microdissection (versus, e.g., the precise identification of the slow-cycling cells by autoradiography coupled with histology [2]).

In sum, existing data from diverse approaches support the bulge location of follicular stem cells. Additional data are clearly needed to resolve some of the apparent inconsistencies raised by the recent data by Rochat *et al* [12]. In a preliminary series of experiments, we have generated a monoclonal antibody to the bulge keratinocytes. Such antibodies, particularly if they are directed against exposed cell surface epitopes, may allow us to isolate and to study the growth properties of such cells.

This work was supported in part by grants awarded by NIH (AR 41669) and the National Alopecia Areata Foundation to RML and T-TS.

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Hair Follicle and Fiber Reconstruction

Amanda J. Reynolds, Clifford M. Lawrence,* and Colin A.B. Jahoda

Department of Biological Sciences, University of Durham, Durham; and *Department of Dermatology, Royal Victoria Infirmary, Newcastle, United Kingdom

The subcutaneous hair follicle is a vital component of mammalian skin, housing numerous specialized cell types that are naturally adapted to extensive tissue remodeling because of their involvement in cyclic fiber production. Considerable demand exists for the ability to create (transplant, stimulate, or substitute for) hair follicles *in vivo*, often when the fibers they produce are considered to be inadequate. A representative *in vitro* model for this appendage is also widely sought, because it embodies potential for exploitation in many areas of biomedical research. This paper outlines two of our approaches to recreating hair follicles: *in vitro*, where cells, growth factors, and some form of supportive element must necessarily be supplied in an appropriate manner, and *in vivo*, when the requirement is for an initial stimulus to the developmental cascade of interactions that ultimately results in follicle formation.

Reprint requests to: Dr A.J. Reynolds, Department of Biological Sciences, University of Durham, South Road, Durham DH1 3LE, UK.

CULTURED FOLLICLE RECONSTRUCTIONS

Our *in vitro* approach has involved trying to identify, isolate, manipulate, and then appropriately recombine what we considered to be the least committed (or most flexible) follicle cell populations. Our aim was to create an environment that could provide some degree of structured support for the four main types of constituent cells, but also allow enough flexibility for cell or tissue interactions that might recapitulate embryonic-like developmental events. Small pieces of germinative epidermal tissue were associated (for 3–6 d) with pre-established mixed papilla and sheath dermal cell monolayers (under standard culture conditions). All three cell populations were then scraped into sticky composite masses and rapidly transferred to the central cavity of pre-prepared follicle tubes (all cells were completely destroyed before outer root sheath epidermis was re-established from cultured cells). Each of these follicle cell–filled structures was then positioned vertically within collagen gel and epidermal culture medium, so that about three quarters of the length became submerged but the uppermost portion remained exposed.

0022-202X/95/$09.50 • SSDI0022-202X(95)00131-4

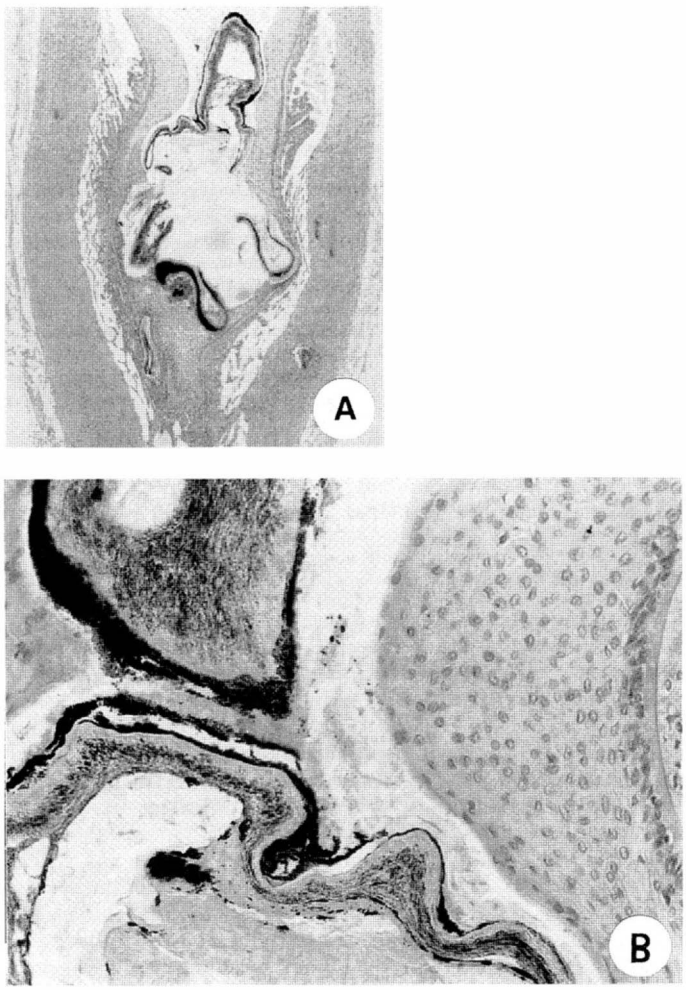

Figure 1. Histology of reconstructed follicle tissue after 3 weeks in culture. Irregularly curved pieces of fibrous material can be seen to have formed within the central cavity of the follicle structures (*A*). At higher magnification the induced fibrous tissue, although misshapen, can be seen to have a structural composition that is comparable to normal hair fiber (*B*).

After 2–3 weeks in culture, our results revealed that the dermal sheath and epidermal outer root sheath cell populations had formed impressive, highly ordered arrangements on either side of the glassy membrane. Moreover, a considerable amount of irregularly distributed fibrous material had been produced, with apparently the same structure and composition as that of normal hair **(Fig 1)**. Papilla-like structures also developed within our follicle reconstructions, and what seemed to be residual papillary tissue was frequently observed in cavities located to the centre of induced portions of "fiber."

Future assessment of the short-term status of the recombinations using cell-labeling techniques should help to reveal the extent to which each individual cell population contributes toward the new follicle structure. More specifically, the use of labeled cells could confirm whether the induced fibres were of germinative and/or outer root sheath epidermal origin.

INDUCTIONS *IN VIVO* INVOLVING HUMAN FOLLICLE TISSUE

Further studies aimed to investigate whether (as has been widely speculated) the capacity to induce follicles and fibers *de novo* is restricted to dermal papillae from highly specialized vibrissa-type sensory follicles. In this context, we had already shown that human papillae can at least stimulate fiber production when they are implanted into the upper portions of amputated (and therefore fiber-growth inactivated) athymic mouse vibrissa follicles. However, our most recently initiated study aims to provide a clearer, more comprehensive assessment of human papilla inductive capabilities by investigating their behavior *in vivo* following their implantation into human skin. In other words, this approach eliminates any complicating influences that could be attributed to other species or indeed to other follicle types.

The preliminary findings from this current work, involving human tissue recombination, are promising. We have noted that (as established during comparable experiments with rodent material) human hair follicle dermal papillae display complex levels of interaction with the surrounding tissue at the site of their implantation. Furthermore, and clearly integral to the papilla-induced interactions, large, non-local hair fibers are seen to emanate from within the wound sites.

In conclusion, the results from the manipulations that we have described above support two important statements, namely, that 1) cultured cells from adult hair follicles retain enough embryonic- or stem cell–type interactive potential to reform follicle-like tissue arrangements, which, in turn, are capable of generating appendage-specific differentiation
put forward, dermal papillae from human hair follicles exhibit similar inductive capabilities *in vivo* to those isolated from rodent mystacial regions, suggesting that at least some fundamental control mechanisms may be common to all follicle types and, perhaps, to all species.

We would like to acknowledge the support contributed by the B.B.S.R.C. (A.F.R.C. Stem Cell Initiative Programme), and the University of Durham Research Fellows Committee.

Proteoglycans and Associated Proteins of the Mammalian Hair Follicle

John R. Couchman and Diana L. du Cros

Department of Cell Biology, University of Alabama at Birmingham, Birmingham, Alabama, U.S.A.

0022-202X/95/$09.50 • SSDI0022-202X(95)00132-5