In This Issue . . .

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Does the Ulceration of Mouse Skin Initiate Cancer?

It is well known, says Samuel Cohen of the University of Nebraska Medical Center, that ulcerating the rat bladder initiates the sequence of steps that lead to cancer, not reversible later. And, in mouse skin, it is known that if cancer is already initiated, ulceration acts as a promoter—it enables the already-initiated cells to become cancerous. So, Cohen and his colleagues Ryohei Hasegawa, Margaret St. John, and T. Scott Tibbels asked, Can ulceration of mouse skin also initiate cancer? They report in this issue that it cannot.

The investigators wounded mouse skin and then applied a promoter substance, 12-O-tetradecanoylphorbol-13-acetate, or TPA. If ulceration were a cancer initiator, then the skin cells would become malignant following the application of TPA. "Our hypothesis was that the cells would become cancerous," says Cohen. "We were surprised when they didn't."

Cohen and his associates then asked why ulceration is an initiator in the bladder but not in the skin. They suggest that because skin proliferates so rapidly, it may have a well-developed repair system to repair molecular damage. Skin, Cohen proposes, may be "used to damage" and better able to deal with it.

In addition, there are certain growth factors in urine, including epidermal growth factor and transferrin, that may act on the bladder cells so that they become initiated when they are wounded.

In any event, Cohen says, the group concludes that "short-term wounding by itself is not capable of leading to cancer. Something else is required."

Wound Healing and the Role of Fibronectin

In the past few years, it has become increasingly clear that fibronectin may be crucial for epithelial cell binding, during morphogenesis and wound healing. But what was not known was whether keratinocytes actually contribute fibronectin to the wound matrix. In this issue, Edward O'Keefe, David Woodley, Ronald Falk, W. Ray Gammon, and Robert Briggaman of the University of North Carolina School of Medicine provide evidence that they do.

About 5 years ago, when fibronectin was first studied, investigators thought that it was a fibroblast—but not an epithelial cell—binding protein. Fibronectin was discovered because it caused fibroblasts to attach to plastic. But, researchers reported, fibronectin did not enhance the attachment to or spread of epithelial cells on surfaces.

So the idea gained credence that fibronectin is not particularly important to epithelial cells. But the problem with this hypothesis, O'Keefe explains, is that the researchers did not study keratinocytes directly. Instead, they studied cells from a malignant epithelial cell line and extrapolated their results to normal keratinocytes. A few years ago, when O'Keefe and others began looking directly at fibronectin's effects on keratinocytes, they found that fibronectin causes these cells to attach to surfaces and spread.

The new studies showed that keratinocytes recognize fibronectin, spread on it, increase their motility on it, and lay it down as they move. O'Keefe, Richard Clark of the National Jewish Hospital in Denver, and others also found that keratinocytes synthesize and secrete fibronectin. And investigators are reporting that fibronectin may be important in wound healing and in embryogenesis. During embryogenesis, fibronectin "may direct epithelial organs to develop in the proper shape," O'Keefe says.

O'Keefe and his associates decided to use an in vitro skin equivalent model to determine whether fibronectin is actually laid down by keratinocytes during wound healing. They found that not only is it laid down but "it's there pretty early. Fibronectin is there in abundance before the basement membrane is assembled," O'Keefe says. He concludes that his findings are further evidence that fibronectin may be involved in wound healing. Keratinocytes may use fibronectin as a surface to migrate on and to cover the wound, he concludes.

Understanding Ichthyosis Vulgaris

About a year ago, Beverly Dale and Karen Holbrook of the University of Washington School of Medicine biopsied affected skin from patients with ichthyosis vulgaris and reported that the keratinocytes have no detectable profilaggrin or filaggrin. Now Philip Fleckman, Dale, Holbrook, and Virginia Sybert continued their studies of the molecular genetics of ichthyosis vulgaris by examining cultured cells. They report their results in this issue.

Although ichthyosis vulgaris is a relatively common inherited disease—occurring in as many as 1 in 250 to 300 individuals—it has not been entirely clear where the gene defect lies. Since the University of Seattle group has been unusually successful in culturing differentiated keratinocytes, they decided to look in vitro for evidence that the defect is in the filaggrin gene.

Profilaggrin and filaggrin are produced in the granular layer of the epidermis. When purified filaggrin is added to keratin filaments, large globules, keratohyaline granules, form. The hypothesis is that filaggrin is a matrix protein that acts as a scaffold for keratin filaments to organize in the stratum corneum and form the keratin pattern. Then, more permanent bonds form and filaggrin disappears from the matrix.

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Patients with ichthyosis vulgaris do not have the granular layer in their affected skin. And although their unaffected skin has a granular layer, it has smaller than normal amounts of profilaggrin and filaggrin, Fleckman reports.

When Fleckman and his colleagues cultured keratinocytes from normal volunteers, the cells differentiated, formed granules, and made profilaggrin. But when they grew keratinocytes from patients with ichthyosis vulgaris, the cells differentiated but were morphologically abnormal. "They did not have granules and did not make profilaggrin," Fleckman says. Moreover, he reports, when they cultured cells from a carrier for ichthyosis vulgaris who was himself clinically unaffected, the cells made only a small amount of profilaggrin. These cells, in vitro, were intermediate between normal cells and cells of clinically affected patients. "In culture, the defect is unmasked," Fleckman says.

Now that they have established this in vitro system, the group suggests that it can be used to study what profilaggrin and filaggrin do and how the expression of the profilaggrin gene is controlled. Moreover, Fleckman points out, ichthyosis vulgaris is only one of a number of skin diseases whose only manifestation is in the epidermis. It should thus be possible to study these other epidermal-specific diseases with cultured epidermal cells.

Determining Prognoses in Psoriasis

A group of investigators at Cornell University Medical College and Rockefeller University has evidence that molecular features of epidermal cells can predict whether a patient with psoriasis has responded to treatment. They report their results in this issue.

The group, consisting of Lisa Staiano-Coico, Alice Gottlieb, Lance Barazani, and D. Martin Carter measured the DNA and RNA content of epidermal cells from involved and uninvolved psoriatic skin and from normal skin. They also tested their epidermal cell samples with a monoclonal antibody that indicates that cells are basal in nature.

They found that the number of cycling epidermal cells is increased in involved and uninvolved psoriatic skin. RNA levels, which measure protein synthesis, are also high in cells from involved psoriatic skin, and the monoclonal antibody marker indicates that lesional skin has increased numbers of basal cells.

When patients with psoriasis are successfully treated, the RNA levels of their epidermal cells go down and the monoclonal antibody stain indicates that there are fewer basal cells. The investigators stress the importance of looking at RNA levels because DNA levels remain elevated even though patients respond to therapy. "Since it is mainly RNA that increases in psoriasis, drugs that affect RNA metabolism might be a way to treat the disease," Gottlieb says.

But the immediate clinical application of their work is to provide prognoses. They report in their paper on 10 patients. The RNA content of epidermal cells of lesional skin remained relatively low after treatment in 9 of them. The tenth patient, however, although not distinguishable from the others on the basis of clinical signs, had very high levels of RNA in his epidermal cells. He had a severe relapse within a month after cessation of therapy.

The DNA, RNA, and antibody tests must be done in an academic center, according to Gottlieb. But they are not technically difficult to perform and, Gottlieb says, "almost every medical school has the equipment to do them." Even though the relationship between RNA and active psoriasis remains to be determined, the tests may prove useful "to tell patients whether they can expect a prolonged remission," Gottlieb says.