Number of cells released from skin by enzymic digestion

Experiment	Cells released per 10 mg tissue (total)	Non-viable cells per 10 mg tissue	Viable cells (%)
Trypsin + EGTA + chondroitinase	140,000	43,500	69
Trypsin + EGTA (control)	31,000	16,700	45

Materials and methods. Albino mice (5–6 days old) were killed by decapitation and a small piece of skin removed from the mid-dorsal area. Pieces of dermis containing the lower hair follicle bulbs (approx. 0.5 × 0.5 × 1 mm) were dissected from the under side of the skin, blotted, weighed, and transferred to 1 ml aliquots of trypsin solution containing 0.3% trypsin (Difco I:250) and 0.05% EGTA in sterile Hanks' solution (calcium- and magnesium-free). Incubation was carried out for 10 min at 37 °C in sealed tubes with gentle shaking every 2 min. Pieces of this tissue were then incubated for a further 10 min in 1 ml of the trypsin solution (control) or, after washing in Hanks' solution, transferred to 1 ml of Hanks' containing 5 units of chondroitinase ABC (Sigma) and incubated for 10 min at 37 °C with gentle shaking. After this, the contents of each tube were sucked up and down for 30 sec with a Pasteur pipette, and the resulting cell suspensions diluted with an equal volume of 0.1% nigrosine in Hanks' solution. This dye stains the cytoplasm of damaged cells, but is not taken up by viable cells[4]. After thorough mixing, a drop of each suspension was placed in the chamber of a haemocytometer and the cells (viable and non-viable) counted.

Results and discussion. The results (see Table) show that nearly 5 times as many cells were released by trypsin plus chondroitinase as compared to trypsin alone, and also that the percentage of viable cells was much higher in the first case (69% as compared to 45%). Microscopic examination of the residual tissue when the new method was used showed only long strands of collagen and parts of lower hair shafts – no intact follicle bulbs were seen. In contrast, the control digest contained in the residual tissue a great number of recognizable follicle bulbs. Some fresh pieces of dermis were also incubated with chondroitinase alone (5 units per ml of Hanks' solution) and, after 30 min at 37 °C it was apparent that some dissociation of follicle bulbs had occurred, but not to the degree found using trypsin plus chondroitinase. The use of the 2 enzymes sequentially thus offers a means of obtaining single viable cells from hair follicles representative of the whole hair bulb.

[3] B. SYLVÉN, Expl. Cell Res. *1*, 582 (1950).
[4] J. P. KALTENBACH, M. H. KALTENBACH and W. B. LYONS, Expl Cell Res. *15*, 112 (1958).

CONGRESSUS

France
29th International Meeting on Electrical Phenomena at Membrane Level

in Saclay, 12–15 October 1976

The main topics are: 1. Bioenergetical study of coupling mechanisms. 2. Electrical phenomena at exitable membrane level. The scientific program and registration information will be available by: Dr. C. Troyanowsky, General Secretary, Société de Chimie physique, 10, rue Vauquelin, F–75231 Paris Cedex 05, France.

Federal Republic of Germany
First International Congress for Research on Medicinal Plants

in München, 6–10 September 1976

Organized under the auspices of Gesellschaft für Arzneipflanzenforschung, the Phytochemical Society of England and the International Association for Plant Tissue Culture the Congress will be devoted to the following topics: A) New natural products and plant drugs with pharmacological or therapeutical activity. B) Plant tissue culture and its bio-technological application. Further information for section A: Dr. P. Wolff or Prof. H. Wagner, Institut für Pharmazeutische Arzneimittellehre, Karlstrasse 29, D–8000 München 2, Federal Republic of Germany. For Section B: Prof. E. Reinhard, Pharmazeutisches Institut, Auf der Morgenstelle 8, D–74 Tübingen, Federal Republic of Germany.

Canada
Third International Symposium on Pharmacology of Thermoregulation

in Banff, 14–17 September 1976

The Symposium will be held at the Banff Centre and further details about registration may be obtained by the organizers: Prof. K. E. Cooper, Division of Medical Physiology, Faculty of Medicine, The University of Calgary, Calgary, Alberta, Canada T2N 1N4; or by Prof. P. Lomax, Department of Pharmacology, UCLA School of Medicine, Los Angeles, California 90024, USA; or by Prof. E. Schönbaum, Peelkensweg 4, 4274 Venhorst N. Br., The Netherlands.

CORRIGENDUM

MARIANNE E. SCHWAGER-HÜBNER and M. C. GNÄDINGER: *Synthesis of Sulfated Glycosaminoglycans by Three Cell Types of the Rabbit Cornea in Culture*, Experientia *32*, 15 (1976). On page 16 in the first paragraph 2 lines have been omitted. The two first sentences should read as follows:

After 21 days of incubation, 50 μCi $Na_2{}^{35}SO_4$ (NEN, specific activity 859 mCi/m*M* were added to the medium of each culture for 4 days. At the end of the exposure to the precursor, the medium was withdrawn and the cells homogenized.