

total protein content, the snake and turtle sera do not differ in their means or in their variation. However, for albumin content they do differ significantly.

The albumin values obtained in this study compare favorably with the electrophoresis data of Deutsch and McShan, who found that turtle sera contain more low mobility components (suggestive of globulins) than do snake sera. In fact, the albumin proportions of the majority of the snake sera were twice those of the turtles.

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The Effect of Serum Ultrafiltrate on Cultivated Mast Cells and Fibroblasts from Human Skin¹

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In a previous study of the cultivation of human tissue mast cells undertaken in this laboratory (1), serum ultrafiltrate was shown to stimulate cytoplasmic granulation when used as a constituent of the medium (chick embryonic extract, with or without cord serum). At the same time it was found that in the presence of cord serum the ultrafiltrate promoted proliferation, whereas in the absence of cord serum it inhibited proliferation, caused marked differentiation, and led to the death of the cells. Since fibroblasts from normal human skin were not cultivated as controls during the previous investigation, the significance of the effect of serum ultrafiltrate could not be evaluated. The present paper reports the results of a 6-month comparative study of the effect of serum ultrafiltrate on cultivated mast cells and fibroblasts, both of human origin.

Following the procedure employed in the earlier work, identical Carrel-flask cultures were prepared from a skin-lesion biopsy of an infant with urticaria pigmentosa and a biopsy of normal human adult skin. Supravital staining with toluidine blue confirmed the identity of the cells in the original cultures from the

urticaria pigmentosa tissue, and in subsequent subcultures, by producing metachromasia, the accepted criterion for mast cells. No metachromasia was noted in the normal skin cultures, or in subsequent subcultures, when these were stained in the same manner. Three test media were used: (1) 40% serum ultrafiltrate, (2) 40% cord serum, and (3) 20% serum ultrafiltrate and 20% cord serum. The balance of each medium consisted of 4% embryonic extract and 56% Tyrode's solution.

The fibroblasts from the normal skin cultures grew, as described by Gey and Gey (2), in a dense radial network in all media, and mitosis occurred frequently. Some fine cytoplasmic granulation was noted in cultures maintained on medium 2. In cultures using media 1 and 3, the cytoplasm remained free of any granulation. This is consistent with the findings of Simms and Stillman (3) in their studies of the effect of serum ultrafiltrate. Growth was sufficiently rapid to permit subculturing every 3 or 4 wk.

The mast cells responded differently to each of the three test media. In cultures maintained on medium 1 the growth was sparse, and the large spindle-shaped cells divided only by amitosis. Differentiation was noted during the first week. The cells formed thickened stalks, and the cytoplasm was densely packed with coarse granules. After 2 wk no further proliferation occurred, and the cells became rounded; at 3 wk they were dead. Media 2 and 3 stimulated excellent growth, and the cells formed a dense radial pattern. Both mitotic and amitotic divisions were noted in these cultures. The cells were of great size, with large regular nuclei, often containing two or more nucleoli. Proliferation was slower than in the fibroblast cultures. It took 6-7 wk to attain adequate outgrowth for subculturing. In cultures placed on medium 2 the cytoplasmic granules varied greatly in size and number, although granulation, in general, was sparse. In cultures using medium 3, however, the cytoplasmic granules were uniformly coarse and so profuse that in some cases they obscured the nuclei. All the above results are in agreement with our previous study.

When fragments of the two tissue strains were grown side by side in the same Carrel flask with medium 3, no alteration of their individual characteristics was observed. Serum ultrafiltrate appears to have a differential effect upon the cytoplasmic granulation in the cells from the two tissue strains used in this study. The fibroblasts remained free of any cytoplasmic granulation, whereas cytoplasmic granulation in the mast cells was uniformly stimulated.

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