BIOLOGICAL SCIENCES

Differences in Growth *in vitro* of Adipose Cells from Normal and Obese Patients

OBESITY is a bodily state in which an excessive accumulation of fat occurs, but it is not known whether it is the lipid content per cell¹ or the total number of adipose cells² which increases. Bray³ found that adipose cells from obese patients were larger than those from normal patients, and concluded that the gain or loss of weight in adults occurs chiefly through changes in the size of existing fat cells. Fat cells *in vivo*, however, are greatly influenced by the nutritional state of the patients, and observations in such conditions do not indicate whether there are acquired and/or inborn differences in adipose cells from normal and obese patients.

We have therefore examined these cells for growth, size and cellular reproduction *in vitro*, where they are not affected by the nutritional state of the patients.

Subcutaneous adipose tissue was obtained from patients, aged 35–50, undergoing abdominal surgery. Cells were dispersed with collagenase according to Rodbell⁴ and centrifuged at low speed. The sedimented cells were cultured on glass cover slips in Leighton tubes containing 1.5 ml. of lactalbumin hydrolysate growth medium supplemented with 20% foetal calf serum, in an atmosphere of 5% CO₂–95% air at 37° C. One day after the establishment of the culture, the culture medium was replaced with fresh medium to remove red blood cells. The cells were then differentially stained with oil red O and Harris haematoxylin solution. The number and size of cells were evaluated respectively by direct microscopic counting and by planimetric measurement from photographs of the cells.

The cells in early cultures resembled fibroblasts and contained some lipids; after 2 months they became balloon shaped —a characteristic of adipose cells—and accumulated large amounts of lipids. In two samples from obese patients, the cultures maintained for 2 months each produced a large mass, several cell layers thick. Microscopy showed that these masses were very similar to the original adipose tissue, and contained a large quantity of lipids.



Fig. 1 The growth curves of adipose cells from normal patients: Ro, △; Hu, □, and Fo, ○; and obese patients: Ma, ▲; Si, ■, and Og, ●, *in vitro*.



Fig. 2 The distribution of adipose cells (percentage of population) from normal patients: Ro, △; Hu, □, and Fo, ○; and obese patients: Ma, ▲; Si, ■, and Og, ●, in different size ranges: a, (6-0.05) inch², b, (0.05-0.10) inch²; and c, (0.10-0.15) inch², after magnification × 32.2.

The results of growth of adipose cells *in vitro* (Fig. 1) from normal patients indicated no increase in cell number with time. In contrast, adipose cells from obese patients rapidly increased in number in the first 4–6 days of culture and then stopped dividing. Measurements of the size of adipose cells (Fig. 2) showed that cells from normal patients rapidly increased in size with time, whereas cells from obese patients did not increase in size until they reached the stationary phase.

The study of the effect of hormones and other growth factors on the cellular growth and reproduction of adipose cells in culture may give further information on the peculiarities of these cells. These aspects are currently being investigated.

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- ¹ Sims, E. A. H., Goldman, R. F., Gluck, C. M., Horton, E. S., Kellcher, P. C., and Horton, E., in *Physiopathology of Adipose Tissue* (edit. by Vague, V.) (Exerpta Medica Foundation, Amsterdam, 1969).
- ² Hirsch, J., Knittle, J. L., and Salans, L. B., *J. Clin. Invest.*, 45, 1023 (1966).
- ³ Bray, G. A., Amer. Coll. Physicians Fifty-second Ann. Session (1970).
- 4 Rodbell, R., J. Biol. Chem., 239, 375 (1964).

Conformation of Synthetic Polypeptide Monolayers

PROTEINS and synthetic polypeptides spread at air/water interfaces are thought to exist in extended chain conformations^{1,2}. From the results of deuterium exchange in monolayers, and infrared spectroscopy and electron diffraction on collapsed films, Malcolm³⁻⁵ reported that the α -helical