Effect of Cell Size on *in Vitro* Fatty Acid and Glyceride–Glycerol Biosynthesis in Rat Adipose Tissue[1] (36843)

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Recent experiments have demonstrated that the metabolic activity of adipose tissue is related to the size of the fat cells (1, 6, 12, 15). Salans and Dougherty (12) reported that in the rat insulin-stimulated incorporation of glucose into triglycerides (TG) decreased as the fat cells enlarged. In contrast to this observation glucose conversion into TG in human adipose tissue increased as fat cell size increased (1, 14, 15).

In the rat adipose tissue is an important site for the *de novo* synthesis of fatty acids (FA) (9); however, this does not appear to be true for human adipose tissue (1, 13). Björntorp and Karlsson (1) have shown that radioactive glucose conversion into TG in human adipose tissue is confined almost exclusively to the glyceride–glycerol (GG) moiety. Therefore, the increase in TG synthesis observed as the human fat cell enlarges is primarily a result of an increased glucose conversion to GG. There is a paucity of information on the effect of fat cell size on glucose conversion into FA and/or GG in the rat. The study reported here was undertaken to evaluate the effect of fat cell size (and/or animal age) on FA and GG synthesis in the rat.

Methods. Male Sprague-Dawley rats[2] were fed a commercial diet[3]. On the day of the experiment, the animals were decapitated and both epididymal fat pads were rapidly excised and weighed. To determine *in vitro* TG, FA and GG biosynthesis, pieces of adipose tissue (about 100 mg) were incubated as previously described (8). The Krebs–Ringer bicarbonate buffer contained 10 μmoles of glucose (0.1 μCi glucose-U-^{14}C) and 0.1 units porcine insulin[4]/ml. The procedures for isolation and counting of the radioactive FA and GG have been described (7). TG biosynthesis was estimated by calculation (sum of FA and GG synthesis).

Fat cells were isolated from another piece of adipose tissue (10) and the lipid/DNA ratio in the free cells was used to estimate cell size (5) as previously described (11). Tissue for both lipogenic capacity and cell size determinations was obtained from the distal portion of the fat pads.

Results. Body weights and fat pad weights of the five groups of rats are presented in Table I. Fat cell size increased as a function of the growth. A 4-fold increase in body weight resulted in a corresponding 4-fold increase in fat cell size. These results are in close agreement with values obtained by others (12, 16).

Conversion of glucose-U-^{14}C to TG decreased 3-fold as fat cell size increased 4-fold (Table I). Glucose conversion into FA was markedly decreased (7-fold) whereas glucose conversion into GG did not change, as the fat cell enlarged (Fig. 1). It is apparent that the decrease in TG synthesis observed as the rat fat cells enlarged was due primarily to the diminished rate of FA biosynthesis.

Discussion. In the present study fat cells were obtained from rats of different sizes (and ages). Consequently it is not possible to separate the effects of cell size from the effects of animal age. However, Salans and

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[2] Spartan Research Animals, Inc., Haslett, MI.

[3] Wayne Lab-Blox.

[4] Generously supplied by Dr. R. Chance, Eli-Lilly Research Laboratories, Indianapolis, IN.

TABLE I. Body Weight, Fat Pad Weight, Fat Cell Size and *in Vitro* Incorporation of Glucose-U-^{14}C into Triglyceride by Adipose Tissue of Rats.

Criteria	Group[a]				
	1	2	3	4	5
Body wt (g)	128 ± 2[b,a]	255 ± 3[b]	342 ± 5[c]	449 ± 5[d]	578 ± 9[e]
Fat pad wt (g)	0.60 ± 0.03[a]	2.18 ± 0.08[b]	3.78 ± 0.33[c]	6.25 ± 0.82[d]	9.05 ± 0.61[e]
Fat cell size (mg × 10^{-5} lipid/cell)	4.26 ± 0.28[a]	8.69 ± 0.84[b]	14.68 ± 2.34[c]	15.64 ± 1.39[c]	19.64 ± 2.31[c]
Triglyceride synthesis/10^6 fat cells[c]	1983 ± 250[a]	1856 ± 261[a]	1723 ± 193[a]	906 ± 132[b]	733 ± 106[b]

[a] Groups 1, 2, 3, 4 and 5 contained 10, 9, 9, 10 and 9 rats, 5, 8, 10, 13 and 30 wk of age, respectively.

[b] Mean ± SEM. Mean values on a line followed by the same superscript letter do not differ significantly ($p > .05$).

[c] Triglyceride synthesis was calculated as the sum of fatty acid and glyceride–glycerol synthesis from Fig. 1. Results expressed as nanomoles glucose-U-^{14}C converted into triglyceride/10^6 fat cells × 2 hr at 37°.

Dougherty (12) have recently shown that the relationship between cell size and TG synthesis in the rat was similar regardless of whether cells of different sizes were obtained from animals of different body weights and ages or from different portions of the epididymal fat pads of animals of the same weight and age.

It is clear that glucose conversion into TG (in the presence of insulin) is diminished as the rat fat cell enlarges (12 and present study) and that the decrease is primarily due to a diminished conversion of glucose to FA (Fig. 1). If it is assumed that 1 mole of glycerol is required to esterify 3 moles of FA, then it can be calculated (from Fig. 1) that 817 nmoles excess GG were synthesized/10^6 cells in the 128 g rats whereas the value was increased to 1043 nmoles/10^6 cells in the 578 g rats. Thus it appears that GG synthesis (over and above that needed to esterify newly formed FA) in both rat and human (1, 14, 15) fat cells is stimulated as the cells enlarge. It has been shown that both basal [rat and human (6, 14)] and epinephrine-stimulated [rat (6)] lipolysis increased as the fat cell enlarged. This enhanced lipolytic rate presumably increases the need for α-glycerophosphate in the larger cells. Di Girolamo and Rudman (3) have reported that glucose conversion to GG increased and glucose conversion to FA decreased as rats age; however, their results were expressed on a tissue weight basis.

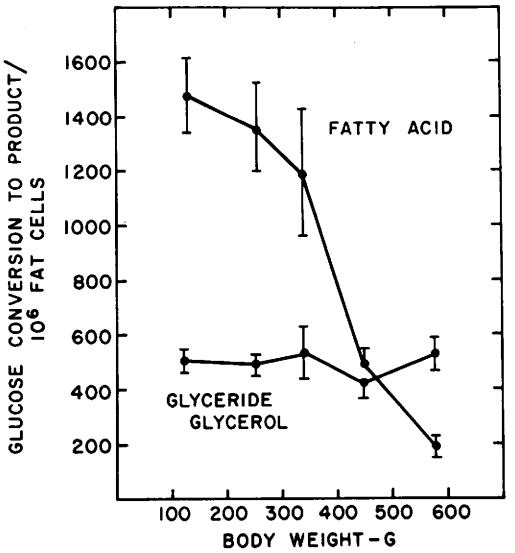

FIG. 1. Nanomoles glucose-U-^{14}C converted into fatty acids or glyceride–glycerol/10^6 fat cells × 2 hr at 37°. Each point represents the mean ± SEM for 9 or 10 rats.

The reason(s) for the marked decrease in FA synthesis as the rat fat cell enlarges and/or ages is not readily apparent. Larger fat cells are less sensitive than smaller cells to both lipogenic (12, 14) and lipolytic (5) hormones. It is not possible to distinguish whether the altered hormone sensitivity of

the fat cell occurs as the result of or is the cause of the altered metabolic activity. It has been suggested that intracellular FA levels function in regulating adipose tissue activity (2). Lipolytic hormones stimulate glucose conversion into GG and inhibit conversion into FA (4). The extent and nature of the interrelationships between lipogenesis, lipolysis and fat cell size (and/or age) remain to be elucidated.

Summary. Glucose-U-^{14}C conversion into triglycerides (in the presence of insulin) decreased as rat fat cells enlarged (and/or aged). The results of this study show that the diminished lipogenic capacity results from the marked decrease in glyceride–fatty acid biosynthesis while glucose conversion into glyceride–glycerol was unaltered by fat cell size.

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