EFFECTS OF GLUCOSE AND INSULIN ON LIPOLYSIS RATES IN HUMAN FAT CELLS OF DIFFERENT SIZES

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1. Introduction

Previous investigations have shown that an increase in the glucose concentration of the incubation medium increases the lipolysis as well as the reesterification of the fatty acids [1, 2]. This effect of glucose can be inhibited by the addition of 2-deoxy-D-glucose [1, 2]. Jungas and Ball [3] have reported that in the absence of glucose, insulin exerts an antilipolytic effect. However, in the presence of glucose this effect of insulin is not seen [4].

Previous investigations from this laboratory have shown that the metabolism of glucose in human adipose cells is correlated to the cell size [5]. The purpose of the present investigation was to study the lipolysis in human fat cells of different sizes with or without the addition of glucose or insulin to the incubation medium.

2. Materials and methods

Biopsies of subcutaneous adipose tissue were obtained in connection with operations on patients with cholecystolithiasis or on patients undergoing exploratory laparotomy. Patients with diabetes mellitus, jaundice or malignant disease were excluded. All patients were fasted overnight. Anesthesia was induced with a short-acting barbiturate and maintained with halothane, nitrous oxide, and oxygen. The biopsies were usually obtained at the beginning of the operations. The ages of the patients varied from 31 years to 82 years and the body weights from 50 kg to 94 kg.

Mean cell size of the biopsies was determined as previously described [5]. Briefly, smaller tissue fragments were prepared from different parts of the biopsies and incubated for 60 min with collagenase (Sigma type 1, Sigma Chemical Company, St. Louis, Mo.). The diameters of 100 cells were measured with a calibrated ocular in a Zeiss photomicroscope. The mean of the cell diameters was considered as mean cell size.

After 30 min of preincubation tissue fragments, at a total weight of about 100–200 mg, were incubated in siliconized glass vials in 2.0 ml Parker medium 199 [6] which had been modified as follows:

a) no glucose present;
b) glucose concentration was 1.0 mM, and in some exriments;
c) 2-deoxy-D-glucose (Sigma Chemical Company, St. Louis, Mo.) was added at a concentration of 1.0 mM.

All incubations were performed in duplicates or triplicates with or without the addition of 0.1 I.U./ml recrystallized pork insulin (Vitrum AB, Stockholm) for 2 hr at 37° and pH 7.4 ± 0.2. The gas phase was air.

Aliquots of the incubation medium were taken after 0 min and after incubation for 120 min for glycerol determinations. Glycerol was determined enzymatically as described by Laurell and Tibbling [7]. Reagents for glycerol determinations were obtained from Boehringer, Mannheim. The release of glycerol was considered as an index of the lipolysis and is expressed in terms of the cellularity of the tissue fragments.

Lipids were extracted from the tissue fragments with chloroform–methanol (2:1, v/v), according to the method described by Folch et al. [8]. Glyceride-glycerol was determined on the chloroform phase as described by Carlson [9]. When the mean cell diam-

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Table 1
Effect of insulin on the release of glycerol by tissue fragments of human adipose tissue incubated in the presence or absence of glucose.

	Glycerol release (nmoles/ 10^5 cells/2 hr)	
	No insulin	0.1 I.U./ml insulin
No glucose present	7.55 ± 0.76	6.93 ± 1.17
1.0 mM glucose	20.6 ± 5.4	24.5 ± 5.1

eter and the glyceride-glycerol content of the tissue fragments are known the number of cells can be calculated according to the formula suggested by Goldrick [10]. The density of the fat cells was assumed to be that of triolein [11].

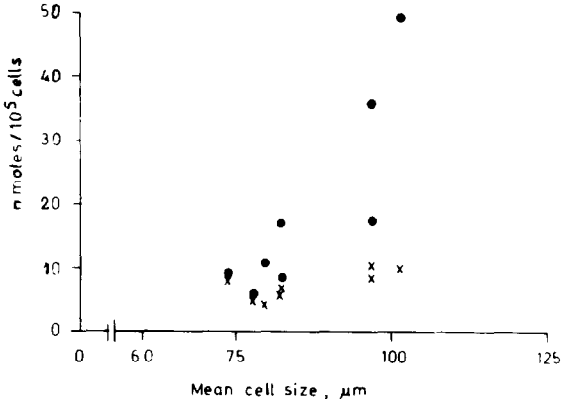

Fig. 1. Relationship between mean cell size and release of glycerol; X, in the absence of glucose; •, in the presence of 1.0 mM glucose.

3. Results

3.1. *Effect of insulin*

When the incubations were performed in the absence of glucose addition of insulin diminished the glycerol release (table 1). However, in the presence of glucose this antilipolytic effect of insulin cannot be seen. On the contrary, addition of insulin increased the lipolysis.

3.2. *Effect of cell size on lipolysis*

Fig. 1 shows the relationships between cell size and glycerol release in the presence or absence of glucose. In the absence of glucose the lipolytic rates were not significantly correlated to cell size ($r = 0.60$; $p < 0.1$). Similar results were obtained when the incubations were performed with 2-deoxy-D-glucose. Addition of glucose stimulated the lipolysis, and in particular in the larger adipose cells. In the presence of glucose the release of glycerol was significantly correlated to cell size ($r = 0.86$; $p < 0.01$).

4. Discussion

The results of the present investigation with human adipose tissue show that in a glucose-free medium insulin tends to diminish the lipolysis. This finding is in agreement with the investigations of Jungas and Ball [3] using rat adipose tissue. In addition, it was found that the antilipolytic effect of insulin was not only diminished in the presence of glucose but insulin actually enhanced the lipolysis. This finding was independent of the size of the fat cells. In their studies of rat adipose tissue Hall and Ball [4] have recently reported that the activity of lipolytic agents such as epinephrine is enhanced in the presence of insulin and glucose. It was suggested by these authors that the favourable action of glucose on the lipolysis is independent of any effects upon the reesterification process. Thus, glucose or products of glucose metabolism may exert a direct effect on the lipolytic process [4]. The findings of the present investigation that insulin exerts a favourable action on the lipolysis in the presence but not in the absence of glucose is presumably due to its stimulatory effect on glucose metabolism.

In the absence of glucose the basal rate of lipolysis was not significantly correlated to cell size. Addition of glucose increased the lipolysis but the larger the adipose cells the greater the increase in the lipolysis. Previous investigations from this laboratory have shown that the larger adipose cells metabolize glucose at greater rates than the smaller cells [5]. In view of the results of the present investigation it seems reasonable to believe that the greater rates of glucose metabolism are responsible for the increased lipolytic rates shown by the larger cells.

All incubations in the present investigation were performed in Parker medium 199 since it has been

shown that this medium is suitable for tissue culture of human adipose tissue [12]. In accordance with the suggestions of several investigators [13–15] glycerol is expressed in terms of the cellularity of the tissue fragments rather than the customary manner, i.e., per unit of tissue lipid.

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