Accelerated Triglyceride Secretion

A METABOLIC CONSEQUENCE OF OBESITY

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ABSTRACT A new animal model was developed to determine the effect of obesity upon endogenous triglyceride secretion. Desert sand rats (*Psammomys obesus*), rodents which become spontaneously obese and hyperinsulinemic when given ad lib. chow, were given intravenous Triton to allow in vivo measurement of triglyceride secretion rates (TGSR). In a group of 18 fasted animals of varying body weight and degrees of obesity, TGSR correlated significantly with body weight ($r = 0.68$, $P < 0.01$) indicating that obesity was associated with accelerated endogenous release of triglyceride. In these same animals, basal plasma insulin levels correlated significantly with body weight ($r = 0.78$, $P < 0.001$) and TGSR correlated significantly with mean plasma insulin levels ($r = 0.73$, $P < 0.001$), suggesting that hyperinsulinemia may have been the mechanism through which obesity enhanced TGSR. No correlation was found between basal triglyceride level and either body weight, basal insulin, or TGSR which suggested that individual triglyceride removal rates among the animals may have been variable. To test this hypothesis, seven animals were studied prospectively before and after induction of obesity. There were significant increases ($P < 0.02$) in all parameters, i.e., weight, plasma insulin level, TGSR, and basal triglyceride level. Thus, when each animal was used as its own control, thereby minimizing the postulated factor of variable individual triglyceride removal, increments in basal triglyceride were shown to accompany the development of obesity, hyperinsulinemia, and accelerated triglyceride secretion. These data from studies in the sand rat offer in vivo evidence that obesity leads to accelerated triglyceride secretion, an effect which may be mediated by hyperinsulinemia, and which can be invoked as one possible mechanism to explain hypertriglyceridemia associated with obesity in man.

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INTRODUCTION

Body weight appears to be one of a variety of factors influencing circulating triglyceride levels in man. Triglyceride levels are higher following equilibration after experimental weight gain and lower after weight reduction (1). In addition, obesity is thought to be a contributing factor to the elevated triglyceride levels observed in patients with endogenous hypertriglyceridemia, and consequently, weight reduction has been advised in the treatment of this disease (2).

The characteristic elevation of both basal and stimulated insulin levels in obese subjects (3) is one conceivable mechanism through which obesity might express its postulated hypertriglyceridemic effect since insulin has been reported to enhance hepatic triglyceride synthesis in vitro (4–6). However, correlations between circulating triglyceride and insulin levels in man have been relatively weak (7). Since an isolated measurement of plasma triglyceride does not distinguish between triglyceride secretion and removal but only represents the balance between the two, a technique which measures triglyceride secretion may more accurately define the possible influence of obesity and its associated hyperinsulinemia upon regulation of circulating triglyceride level.

For several reasons, it is difficult to determine the influence of obesity upon rates of triglyceride secretion in humans. These points include the need to manipulate caloric intake prospectively over an extended period of time to establish desired levels of obesity and hypertriglyceridemia and the lack of universal agreement re-

TABLE I

Values of Body Weight, Fasting Plasma Glucose, Plasma Insulin, Plasma Triglyceride and TGSR in Sand Rats, either Lean or of Varying Degrees of Adiposity

Sand rats	Weight	Glucose	Pre-Triton insulin	Mean insulin‡	Pre-Triton TG_0	45 min post-Triton TG_1	90 min post-Triton TG_2	TG production rate
	g	*mg/100 ml*	*μU/ml*	*μU/ml*	*mg/100 ml*	*mg/100 ml*	*mg/100 ml*	*mg/min*
1	125	80	44	53	90	133	161	0.0545
2	130	109	12	17	62	87	121	0.0383
3	130	62	32	31	102	155	217	0.0777
*4	130	94	90	65	122	382	443	0.1109
5	141	80	80	39	51	78	99	0.0369
6	145	83	28	75	65	139	228	0.1135
7	146	95	58	61	35	62	88	0.0392
8	151	88	16	32	58	98	127	0.0552
*9	154	100	16	31	36	185	256	0.0724
10	156	74	44	44	36	120	154	0.1072
11	163	99	92	224	144	275	350	0.1784
12	167	92	86	99	78	127	218	0.0915
13	170	69	122	207	58	140	240	0.1340
14	175	94	94	135	35	109	148	0.1022
15	177	92	53	38	27	75	134	0.0799
16	215	90	72	149	74	286	370	0.3076
17	240	92	188	183	33	175	294	0.2450
18	268	92	181	147	55	128	—	0.1385

* Samples TG_1 and TG_2 drawn at 120 and 240 min, respectively.

‡ Mean of the three values obtained before and after Triton injection.

garding the validity of any single method for determination of triglyceride secretion in man. On the other hand, *Psammomys obesus,* the desert sand rat, is ideally suited for this type of study since in this animal model excess caloric intake quickly induces obesity (8, 9) and endogenous triglyceride secretion rates (TGSR)[1] can be determined through the use of Triton (10).

METHODS

Experimental animal and colony characteristics. The sand rat is a rodent which inhabits deserts of the Middle East, normally feeds on low caloric plants, and is lean. In the laboratory setting, increased caloric intake provided by ad lib. laboratory chow has been reported to induce obesity in these animals (8, 9). The sand rats used in this study were trapped in Egypt and transported to Seattle, Wash., where they were maintained in a clean environment which utilized a laminar flow unit. Nutrition consisted of spinach, tap water, salt blocks, and Purina lab chow (56% carbohydrate, 5% fat) (Ralston Purina Co., St. Louis, Mo.) Two basic types of diets were utilized: (*a*) weight-maintaining (30 g of Purina lab chow daily) and (*b*) ad lib. (unlimited access to Purina lab chow). All of the rats studied were adult animals. Fasting plasma glucose levels ranged between 62-109 mg/100 ml (Table I) and hematocrits between 38–56%.

Physiologic methodology. Before all experiments, the animals were fasted overnight for 12 h. Halothan inhalation induction and intraperitoneal pentobarbital were used to establish general anesthesia. Temperature was maintained at 37°C through the use of heat lamps and rectal probe monitoring. Hind foot veins were used for injections and blood samples were collected from the retro-orbital capillary bed. Plasma volumes were determined through use of 0.2 cm^3 of undiluted Evans blue dye; the percent absorbance of a plasma sample collected 5 min after injection of the dye was determined by spectrophotometry.

Triton WR-1339, a nonionic detergent, was injected intravenously and the subsequent increases in triglyceride levels were used to calculate TGSR (10). The mechanism of action of Triton upon triglyceride metabolism is thought to be through its detergent properties which bind triglyceride, thereby preventing removal of approximately 80–90% of circulating triglyceride (11). Hence, increments observed in circulating triglyceride after administration of Triton have been interpreted to be a result of triglyceride secretion. A pre-Triton sample was collected immediately before Triton injection; post-Triton samples were collected at 45 and 90 min after the Triton injection (Fig. 1). To insure a dose of Triton which would provide a maximal effect, a Triton dose-triglyceride secretion response curve was determined using adult lean nonlipemic animals and doses of 0 (saline control), 30, 60, 90, and 120 mg. TGSR were determined by the equation:

$$TGSR = \frac{\frac{TG_1 - TG_0}{T_1} + \frac{TG_2 - TG_0}{T_2}}{2} \times PV_E$$

where TGSR = triglyceride secretion rate; TG_0 = basal

[1] *Abbreviations used in this paper:* TG, triglyceride; TGSR, triglyceride secretion rate.

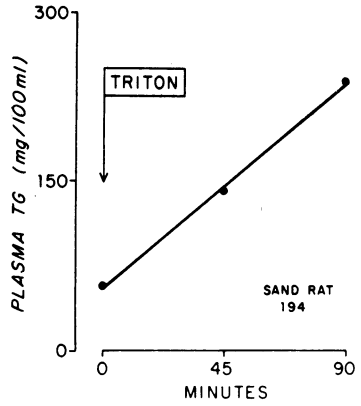

FIGURE 1 Method of determining triglyceride secretion (milligrams per minute) using intravenous Triton (120 mg).

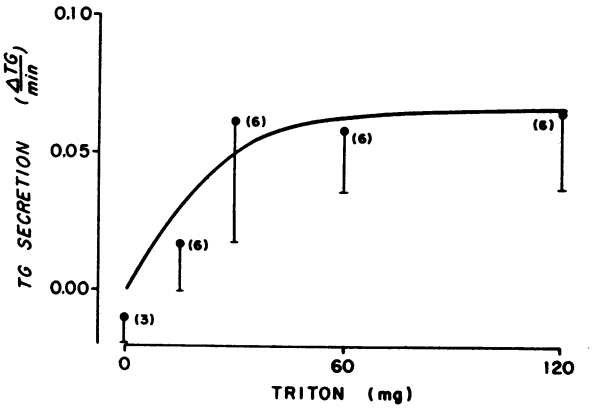

FIGURE 3 Triton dose-triglyceride secretion response curve in lean sand rats (means ±1 SD).

plasma TG concentration (mg/100 ml); TG_1 = first plasma TG concentration after Triton injection; TG_2 = second plasma TG concentration after Triton injection; T_1 = time of first sampling in minutes after Triton injection; T_2 = time of second sampling in minutes after Triton injection; PV_E = estimated plasma volume (cubic centimeters) based upon the linear relationship between body weight and plasma volume (Fig. 2), expressed by the formula: $PV_E = \{0.016 \times \text{weight (grams)}\} + 4.25$.

Experimental studies. (*a*) Determination of TGSR was carried out in 18 animals who were either lean or had varying degrees of adiposity. (*b*) A prospective study was made of a group of seven lean animals in which weight, basal circulating triglyceride concentration, TGSR, and circulating insulin level were determined before and after the induction of obesity through ad lib. feeding for 3 mo. An additional four lean control animals received two Triton injections 2 wk apart to determine whether repeated injections of Triton would influence secretion rates. A second Triton dose-triglyceride secretion response curve using only very obese animals was developed to determine whether the 120 mg dose provided a maximal effect in the obese as well as the lean animals.

Chemical methodology. All blood samples were anticoagulated with ethylenediaminetetraacetic acid (EDTA). Hematocrits were determined using a microhematocrit method. Plasma glucose was determined by an autoanalyzer ferrocyanide method (12). Plasma triglyceride was measured by an autoanalyzer modification (13) of the method of Carlson. Plasma insulin was determined by an ^{125}I radioimmunoassay by double antibody (14) using an albino rat standard. Assay of 1:2, 1:4, 1:8, and 1:16 dilutions of an individual sand rat plasma sample was performed to determine whether there was complete cross-reactivity between sand rat insulin and albino rat standards. In addition, plasma samples from 12 gerbils (used because of their close phylogenetic relationship to the sand rat) were assayed to determine whether these values were comparable with the sand rat values when compared with levels usually observed in man.

RESULTS

Triton dose-triglyceride secretion response. Since a statistically significant correlation ($r = 0.65$, $P < 0.01$) between plasma volume and body weight was found in a group of 21 randomly selected animals who were of varying weight and degrees of obesity (Fig. 2), plasma volumes used in the equation for calculating triglyceride production rates were estimated on the basis of body weight using the equation of the line given in Fig. 2. A

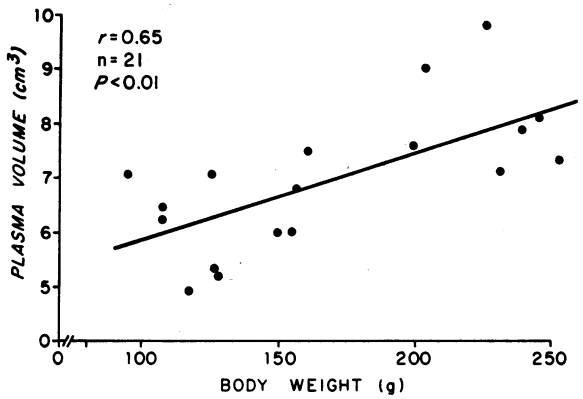

FIGURE 2 Correlation of plasma volume with body weight of sand rats.

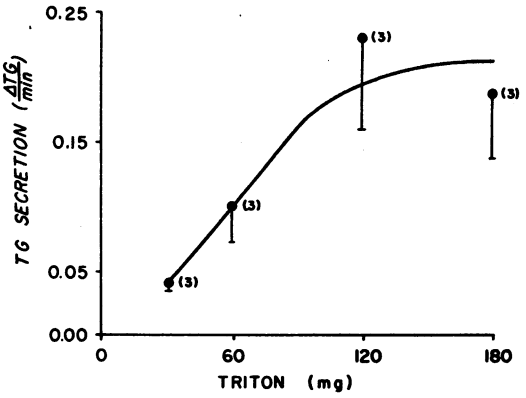

FIGURE 4 Triton dose-triglyceride secretion response curve in obese sand rats (means ±1 SD).

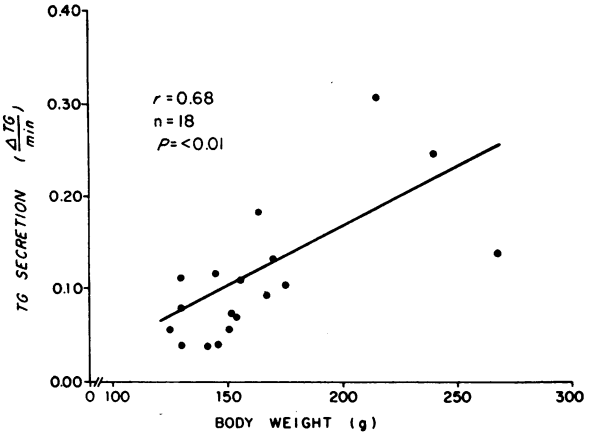

FIGURE 5 Correlation of TGSR with body weights of sand rats.

plateau in secretion rates in lean animals was observed beginning at approximately the 60 mg dose of Triton (Fig. 3). For all subsequent determinations of triglyceride production rates, the 120 mg dose of Triton was chosen to insure maximal effect in obese animals; the validity of this choice was demonstrated in a group of obese animals (body weight = 235±14 g; mean ±SD) in whom the 120 mg dose also provided maximal effect (Fig. 4).

TGSR in animals of varying weight and degrees of adiposity. The range of basal (pre-Triton) plasma insulin values for these 18 sand rats was 12–188 μU/ml (Table I). Assay of the 1:2, 1:4, 1:8, and 1:16 dilutions of a sand rat plasma sample with an insulin value of 140 μU/ml when undiluted yielded results of 72, 40, 21, and 12, respectively. The mean fasting plasma insulin value for 12 gerbils using the same standard was 17±7 μU/ml (mean ±SD).

TGSR correlated significantly with body weight ($r = 0.68$, $n = 18$, $P < 0.01$; Fig. 5). In contrast, the pre-Triton triglyceride level did not correlate significantly with either body weight ($r = 0.30$) or TGSR ($r = 0.15$). Statistically significant correlations were found between basal (pre-Triton) plasma insulin level obtained at time 0 min and body weight ($r = 0.78$, $P < 0.001$; Fig. 6) and between TGSR and the mean plasma insulin level (i.e., mean of the insulin values at 0, 45, and 90 min) during the entire procedure ($r = 0.73$, $P < 0.001$; Fig. 7). The latter correlation was also significant if only the values at 45 and 90 min were used for the mean insulin level ($r = 0.67$, $P < 0.005$). The correlations between basal (pre-Triton) triglyceride level and basal insulin level ($r = -0.41$) and mean insulin level during the Triton procedure ($r = 0.22$) were not significant.

TGSR in animals studied prospectively. All seven animals (Table II) studied prospectively before and after

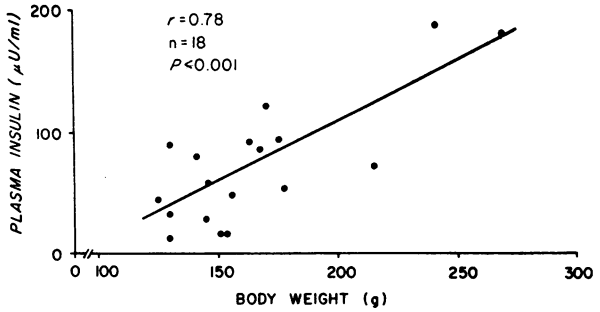

FIGURE 6 Correlation of basal plasma insulin levels with body weights of sand rats.

induction of obesity had significant increases in weight ($P < 0.02$), basal triglyceride level ($P < 0.02$), triglyceride secretion rate ($P < 0.02$), basal insulin level ($P < 0.02$), and mean insulin level during the Triton procedure ($P < 0.02$). No significant increases in secretion rates were observed in the four control animals receiving two Triton injections 2 wk apart (initial rates in mg/min = 0.2006, 0.1118,, 0.1436, and 0.1754; subsequent rates = 0.0958, 0.0989, 0.0885, and 0.1035 respectively); if anything the secretory rates were slightly lower on the second occasion.

DISCUSSION

Obesity in the sand rat was clearly associated with accelerated rates of triglyceride secretion as calculated from circulating triglyceride increments after Triton. This was demonstrated by the significant positive correlation between TGSR and body weight in the group of 18 animals who were of varying weights and degrees of adiposity. Further evidence of the enhancing effect of obesity upon triglyceride secretion was found in the prospective studies wherein all seven animals demon-

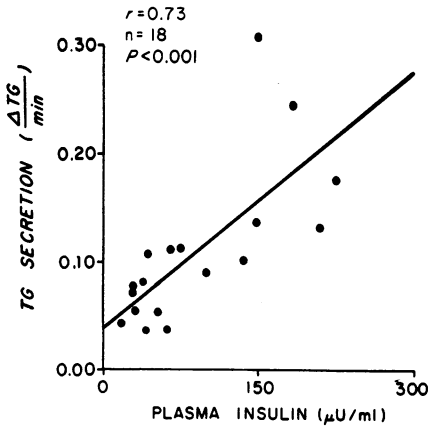

FIGURE 7 Correlation of TGSR with mean plasma insulin levels obtained during the Triton procedure in sand rats.

TABLE II

Values of Body Weight, Fasting Plasma Glucose, Plasma Insulin, TGSR, and Plasma Triglyceride in Sand Rats Studied both in the Lean (LN) and Obese (OB) State

Sand rat	Weight		Glucose		Pre-Triton insulin		Mean insulin*		TG production rate		Pre-Triton TG	
	LN	OB	LN	OB	LN	OB	LN	OB	LN	OB	LN	OB
	g		*mg/100 ml*		*μU/ml*		*μU/ml*		*mg/min*		*mg/100 ml*	
1	124	184	68	107	44	92	29	227	0.0380	0.1893	10	54
2	125	241	101	100	40	1632	24	1680	0.1510	0.2352	34	81
3	125	181	80	78	44	288	53	327	0.0545	0.1553	90	121
4	141	202	80	92	80	220	39	225	0.0369	0.1425	51	149
5	145	207	83	76	28	208	75	203	0.1135	0.2200	63	426
6	146	189	95	86	58	226	61	173	0.0392	0.3627	35	130
7	175	239	94	81	94	1840	135	2880	0.1022	0.1726	35	53
Mean	140	206	86	89	56	644	59	816	0.0765	0.2111	46	145
+SD	17	23	11	11	22	695	35	981	0.0423	0.0691	24	120

* Mean of the three values before and after Triton injection.

strated significant increases in basal triglyceride level and secretion rates after becoming obese. The fact that the prospective study's control group did not have significant increases in basal triglyceride level or secretion implies that repeated administration of Triton did not, of itself, increase these parameters.

The Triton dose-triglyceride secretion response curves for both the lean and obese animals demonstrate that the 120 mg dose caused maximal blockade of triglyceride removal in both groups. Based upon disappearance rates of Triton-incubated ^{14}C-labeled serum lipids obtained from Triton-treated donor rats which were injected in Triton-treated recipient rats, Otway and Robinson (11) estimated that 100 mg of intravenous Triton given to rats comparable in size with ours produced a 90% block in removal. In addition, in the same publication these investigators reported a separate experiment in which preincubation of [^{14}C]chyle with Triton before intravenous injection of the chyle as a bolus in Triton-treated rats produced an 80% blockade of triglyceride removal. It should be noted that in the latter experiment it was necessary to preincubate the [^{14}C]chyle with Triton to obtain the 80% block. However, this maneuver seems warranted since the chyle was presented to the animals' circulation as a large bolus. With no preincubation of this bolus one would expect a lesser degree of removal blockade on the basis of sheer bulk since some of the chyle might be removed before there was adequate time for exposure to the detergent action of circulating Triton. In view of the fact that the dose used in our study provided maximum effect in both the lean and obese animals, it seems reasonable to conclude that removal blockade in our animals was of a comparable order of magnitude, i.e., 80–90%, as was demonstrated by Otway and Robinson (11).

Despite the increased TGSR in the obese animals, no significant correlation was demonstrable between pre-Triton triglyceride concentration and body weight nor with TGSR. However, as shown in the prospective study, weight gain was associated with parallel increases in pre-Triton triglyceride level and TGSR. These data suggest that simple correlation analysis between triglyceride level and either body weight or TGSR may not be the most sensitive statistical method to detect changes induced in triglyceride transport by obesity, although positive correlations between plasma triglyceride and obesity have been previously reported in man (15).

As has been previously reported both in sand rats (16), other types of rats (17), and in man (1), induction of obesity is associated both with increases in circulating insulin level and in in vitro parameters of insulin resistance (18). This phenomenon was demonstrated in this study by the significant positive correlation between the basal circulating insulin level and body weight in the group containing 18 lean or obese animals. Moreover, all seven animals in the prospective study had increases in insulin level as they became obese. In general, the insulin levels observed in both the lean and obese sand rats were noticeably higher than those usually seen in man. Dilution studies indicated that the insulin values were not artifactually elevated as a result of incomplete cross reactivity of the immunoreactive material with the insulin antibody, the linear relationship observed between insulin level and sample dilution suggesting that the cross-reactivity between sand rat insulin and albino rat standards was complete. In addition, plasma samples from gerbils

contained insulin levels within the range observed for man. Thus, it appears that the sand rat, both in the lean and particularly in the obese state, is characterized by high levels of circulating insulin.

TGSR and circulating insulin level were significantly related. The mean of the three insulin values obtained from the Triton procedure was used for this correlation since it seemed probable that this value more accurately reflected the degree of endogenous insulinization during the time that secretion rates were determined. This relationship remained significant if the 45- and 90 min samples only were used for the mean insulin level. In contrast, the pre-Triton insulin value was used for the correlation between basal insulin and body weight since this insulin value more accurately reflects the basal state.

The fact that pre-Triton triglyceride levels did not correlate significantly with plasma insulin in the group of 18 sand rats studied cross-sectionally is of interest in light of the fact that both of these values increased simultaneously in all the 7 animals studied prospectively. Since insulin level correlated well with rates of triglyceride secretion, and triglyceride level represents a balance between secretion and removal, the fact that triglyceride level did not correlate with insulin level implies that rates of triglyceride removal varied considerably among the animals. If marked variable removal efficiency did occur, correlation analysis between triglyceride level and insulin level might not be a sufficiently sensitive statistical method to detect the effects of increased insulin level upon triglyceride metabolism and may account for the weak correlation between plasma triglyceride and insulin in the 18 cross-sectional studies and in studies previously reported in man (7). In contrast to the cross-sectional studies, when each animal was used as its own control, the postulated factor of variable removal efficiency among the animals should have been minimized; this may explain why increases in both plasma triglyceride and insulin level appeared to be associated as the animals became obese in the seven prospective studies.

In conclusion, these studies demonstrate that induction of obesity in the sand rat leads to an increase in rates of triglyceride secretion which can be invoked as one possible mechanism to explain the increased levels of plasma triglyceride associated with obesity. The fact that plasma levels of insulin were increased in the obese animals and correlated well with TGSR is consistent with the concept that hyperinsulinemia might have been the mechanism through which obesity expressed its hypertriglyceridemic effect. Although similar hypotheses have been previously formed (19–21), this appears to be the first demonstration that in vivo TGSR correlate significantly with circulating insulin levels.

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REFERENCES

1. Sims, E. A. H., R. F. Goldman, C. M. Gluck, E. S. Horton, P. C. Kelleher, and D. W. Rowe. 1968. Experimental obesity in man. *Trans. Assoc. Am. Physicians Phila.* **81**: 153.
2. Levy, R. I., D. S. Fredrickson, R. Shulman, D. W. Bilheimer, J. L. Breslow, N. J. Stone, S. E. Lux, H. R. Sloan, R. M. Krauss, and P. N. Herbert. 1972. Dietary and drug treatment of primary hyperlipoproteinemia. *Ann. Intern. Med.* **77**: 267.
3. Bagdade, J. D., E. L. Bierman, and D. Porte, Jr. 1967. The significance of basal insulin levels in the evaluation of the insulin response to glucose in diabetic and nondiabetic subjects. *J. Clin. Invest.* **46**: 1549.
4. Mayes, P. A. 1970. Studies on the major pathways of hepatic lipid metabolism using the perfused liver. *In* Adipose Tissue. B. Jeanrenaud and D. Hepp, editors. Academic Press, Inc., New York. 186.
5. Letarte, J., and T. R. Fraser. 1969. Stimulation by insulin of the incorporation of U ^{14}C-glucose into lipids released by the liver. *Diabetologia.* **5**: 358.
6. Woodside, W. F., and M. Heimberg. 1972. Hepatic metabolism of free fatty acids in experimental diabetes. *Isr. J. Med. Sci.* **8**: 309.
7. Bierman, E. L., D. Porte, Jr., and J. D. Bagdade. 1970. Hypertriglyceridemia and glucose intolerance in man. *In* Adipose Tissue. B. Jeanrenaud and D. Hepp, editors. Academic Press, Inc., New York. 209.
8. Schmidt-Nielsen, K., H. B. Haines, and D. B. Hackel. 1964. Diabetes mellitus in the sand rat induced by standard laboratory diets. *Science* (*Wash. D. C.*). **143**: 689.
9. Hackel, D. B., H. E. Lebovitz, L. A. Frohman, E. Mikat, and K. Schmidt-Nielsen. 1967. Effect of caloric restriction on the glucose tolerance and plasma insulin of the sand rat. *Metab.* (*Clin. Exp.*). **16**: 1133.
10. Otway, S., and D. S. Robinson. The use of non-ionic detergent (Triton WR 1339) to determine rates of triglyceride entry into the circulation of the rat under different physiological conditions. 1967. *J. Physiol.* (*Lond.*). **190**: 321.
11. Otway, S., and D. S. Robinson. 1967. The effect of a non-ionic detergent (Triton WR 1339) on the removal of triglyceride fatty acids from the blood of the rat. *J. Physiol.* (*Lond.*). **190**: 309.
12. Hoffman, W. S. 1937. Rapid photoelectric method for the determination of glucose in blood and urine. *J. Biol. Chem.* **120**: 51.
13. Bierman, E. L., D. Porte, Jr., D. D. O'Hara, M. L. Schwartz, and F. C. Wood, Jr. 1965. Characterization of fat particles in plasma of hyperlipemic subjects maintained on fat-free high-carbohydrate diets. *J. Clin. Invest.* **44**: 261.

14. Morgan, C. R., and A. Lazarow. 1963. Immunoassay of insulin: two antibody system: plasma insulin levels of normal, subdiabetic, and diabetic rats. *Diabetes.* **12:** 115.
15. Ford, S., Jr., R. C. Bozian, and H. C. Knowles, Jr. 1968. Interactions of obesity and glucose and insulin levels in hypertriglyceridemia. *Am. J. Clin. Nutr.* **21:** 904.
16. Hackel, D. B., L. Frohman, E. Mikat, H. E. Lebovitz, K. Schmidt-Nielsen, and T. D. Kinney. 1966. Effect of diet on the glucose tolerance and plasma insulin levels of the sand rat (Psammomys Obesus). *Diabetes.* **15:** 2, 105.
17. York, D. A., J. Steinke, and G. A. Bray. 1972. Hyperinsulinemia and insulin resistance in genetically obese rats. *Metab. (Clin. Exp.).* **21:** 4, 277.
18. Stern, J., P. R. Johnson, M. R. C. Greenwood, L. M. Zucker, and J. Hirsch. 1972. Insulin resistance and pancreatic insulin release in the genetically obese Zucker rat. *Proc. Soc. Exp. Biol. Med.* **139:** 66.
19. Farquhar, J. W., A. Frank, R. C. Gross, and G. M. Reaven. 1966. Glucose, insulin, and triglyceride responses to high and low carbohydrate diets in man. *J. Clin. Invest.* **45:** 1648.
20. Reaven, G. M., R. L. Lerner, M. P. Stern, and J. W. Farquhar. 1967. Role of insulin in endogenous hypertriglyceridemia. *J. Clin. Invest.* **46:** 11, 1756.
21. Schonfeld, G., and B. Pfleger. 1971. Overproduction of very low-density lipoproteins by livers of genetically obese rats. *Am. J. Physiol.* **220:** 1178.