Effect of dietary fat on pancreatic lipase levels in the rat

Lewis I. Gidez

Departments of Biochemistry and Medicine, Albert Einstein College of Medicine, Yeshiva University, Bronx, New York 10461

Abstract The effect of dietary fat on levels of lipase and other enzymes in rat pancreas has been studied. It was possible to raise levels of lipase in animals by supplementing their commercial chow diet with added fat or by raising the level of fat in semipurified diets from 4% to 22%. Pancreatic amylase levels decreased in rats fed the high fat diets, whereas levels of chymotrypsinogen and trypsinogen were unaffected. The type of carbohydrate in the semipurified diets made no difference. Thus, the levels of enzymes in rats fed dextrose-containing diets or cornstarch-containing diets were similar. On the basis of the present data, and results of others, it would appear that levels of pancreatic lipase are increased when the fat content of the diet is raised from about 5% to 15–22%, but that little or no additional increase in lipase levels can be attained by any further increase in the amount of dietary fat.

Supplementary key words dietary cholesterol · amylase · chymotrypsinogen · trypsinogen

Previous investigators have shown that the levels of certain enzymes in the pancreas are influenced by diet. Grossman, Greengard, and Ivy (1) demonstrated that rats fed diets rich in starch or protein for 3 wk had elevated levels of amylase or proteolytic enzymes, respectively. Desnuelle and his colleagues showed that levels and rates of biosynthesis of rat pancreatic amylase were increased in animals fed a diet rich in starch (2, 3), while increases in chymotrypsinogen were observed in rats fed a high protein diet (2, 4). These studies clearly demonstrated that the relative proportions of these enzymes could be modified according to the predominant constituent in the diet. Lipase levels, on the other hand, were shown by Grossman et al. (1) and by Reboud, Ben Abdeljlil, and Desnuelle (2) to be unaffected by a high content of fat in the diet. Bučko and Kopec (5) showed that the levels of pancreatic lipase in rats maintained for 28 days on a diet containing 58% corn oil (by weight) was 33% higher than in rats fed a diet containing 9% corn oil (by weight). More recently, Snook (6) fed isonitrogenous, isocaloric diets containing graded amounts of fat to rats and demonstrated increased levels of pancreatic lipase in animals fed diets providing 55% or more of the calories as fat. Deschodt-Lanckman et al. (7) also showed a significant elevation of lipase levels in the pancreas when rats were changed from a diet containing 4% corn oil (by weight) to one with 50% corn oil.

The present work is concerned with studies on the effect of dietary fat on pancreatic lipase levels in rats fed one of three types of diets with or without added fat. One diet was rat chow diet with added olive oil and/or cholesterol; the other diets were commercial semipurified formulations that contained corn oil. The high fat diets in all of these studies contained 15–22% fat (by weight) as compared with 41–58% fat in the studies cited above (1, 2, 5–7).

MATERIALS AND METHODS

Animals and diets

Two types of dietary studies were carried out. In one series of experiments (A and B), rats were fed a chow diet with or without added fat and/or cholesterol. In the second group of experiments (D and S), the rats were maintained on semipurified diets with or without added fat. Rats in all experiments were given food and water ad lib. The animals used in experiments A and B were caged in groups of four or six; for experiments D and S the rats were caged individually.

TABLE 1. Compositions of semipurified diets[a]

	D-1[b]	D-2[c]	D-3[d]	S-1[b]	S-2[c]
			%		
Casein, vitamin-free test, GBI	21.1	21.1	21.1	21.1	21.1
Cellulose	9.0	9.0	9.0	9.0	9.0
Salt mix, U.S.P. XIV	4.0	4.0	4.0	4.0	4.0
Vitamin fortification mix, GBI	1.0	1.0	1.0	1.0	1.0
Corn oil	4.0	22.0		4.0	22.0
Linoleic acid (98% pure)			0.5		
Dextrose	60.8	42.9	64.4		
Corn starch				60.8	60.8

[a] All the diets except S-1 were pelleted (1/2 inch). Diet S-1 was a powder.
[b] In diets D-1 and S-1, 23%, 10%, and 67% of calories were derived from protein, fat, and carbohydrate, respectively (3.6 kcal/g of diet).
[c] In diets D-2 and S-2, 18.5%, 43.5%, and 38% of calories were derived from protein, fat, and carbohydrate, respectively (4.5 kcal/g of diet).
[d] In diet D-3, 24.5%, 1%, and 74.5% of calories were derived from protein, fat, and carbohydrate, respectively (3.5 kcal/g of diet).

Experiment A[1]

The rat chow used in this experiment (Extralabo M. 25, Paris, France) contained 6.5% fat and 25.4% protein. Male Wistar rats were fed this chow or the chow with added olive oil and cholesterol. The latter diet was prepared by mixing together the cholesterol, olive oil, and powdered chow. The composition of this diet was 1% cholesterol, 9% olive oil, and 90% chow; it contained 14.8% fat and 22.9% protein (calculated).

Experiment B

The rat chow used in this experiment (Rockland Farms, New City, N.Y.) contained 4.5% fat and 24.9% protein. Male Sprague-Dawley rats were fed this chow or the same chow with added olive oil and/or cholesterol. The cholesterol or olive oil, when each was used alone, was mixed thoroughly with powdered chow; when cholesterol and olive oil were used together, the cholesterol was first dissolved in hot olive oil (105–115°C), and this solution was rapidly mixed with the powdered rat chow. The calculated compositions of the final diets were: (*a*) cholesterol diet, 2.3% cholesterol, 4.4% fat, and 24.3% protein; (*b*) olive oil diet, 21.1% fat and 20.5% protein; and (*c*) cholesterol–olive oil diet, 1.9% cholesterol, 20.9% fat, and 20.1% protein.

Semipurified diets

The compositions of the semipurified diets used in experiments D and S are shown in Table 1. These diets were purchased from General Biochemicals Corp., Chagrin Falls, Ohio. The letters D and S refer to the type of carbohydrate in the diet, dextrose and cornstarch, respectively.

Experiment D

23 male Sprague-Dawley rats (average weight, 320 g) that had been maintained on a normal rat chow diet (Rockland Farms) were fed diet D-1, a semipurified diet that contained 4% corn oil, for 21 days. At the end of this time, 7 of the animals were killed (group I, Table 4), and the remaining 16 were placed on the high fat diet (22% corn oil) D-2; 8 of these rats were killed after 14 days (group II, Table 4), and 8 were killed after 41 days (group III, Table 4).

In a second study, 23 additional rats (average weight, 432 g) were divided into three groups. Seven animals in one group (group IV, Table 4) were fed diet D-1 for 36 days, and were then killed. A second group of eight rats (group V, Table 4) was fed diet D-1 for 30 days and then diet D-2 for 42 days before being killed. The third group of eight rats (group VI, Table 4) was fed diet D-1 for 28 days followed by diet D-3 for 41 days. This latter diet contained only 0.5% fat (as linoleic acid). The purpose of the second study (groups IV, V, and VI) was to determine the effect of a very low fat diet on enzyme levels. Diet D-3 was used instead of a completely fat-free diet to avoid an essential fatty acid deficiency.

Experiment S

The diets used in this experiment, S-1 and S-2, were identical with D-1 and D-2, respectively, except that they contained cornstarch instead of dextrose. This experiment was carried out to determine *1*) whether the type of dietary carbohydrate has any effect on enzyme

[1] Experiment A was carried out in the laboratory of Dr. Pierre Desnuelle, Institut de Chimie Biologique, Marseille, France.

levels and *2*) whether changes in enzyme levels are reversible.

33 male Sprague-Dawley rats (average weight, 333 g) were used in this experiment. The lower portion of Fig. 1 indicates the type of diet fed and the duration of each feeding period for the animals in each of the eight groups, A through H. All the rats were fed diet S-1 for 25 days; thereafter they were fed diets S-1 and/or S-2 as indicated. The rats were killed at the end of the last feeding period.

Enzyme and chemical assays

After the indicated periods of time on the respective diets, the animals were killed by decapitation. Food was withdrawn from the rats between 9 and 10 AM on the day of an experiment, and all rats were killed within 1 hr thereafter. The pancreata were quickly removed and freed of adhering fatty and connective tissue. They were then weighed, and frozen on dry ice.

The frozen glands were homogenized individually for 2 min at 0°C in 9 times their weight of cold-distilled water. Lipase and amylase, and in some studies chymotrypsinogen and trypsinogen, assays were carried out on aliquots from each homogenate.

Lipase activity was determined according to the method of Marchis-Mouren, Sarda, and Desnuelle (8). The rate of addition of 0.1 N NaOH necessary to maintain the pH at 9.0 (37°C) after addition of the enzyme to an olive oil–gum arabic (Fisher) emulsion in the presence of bile salts was measured by means of a Radiometer pH-stat over the initial 3-min period. The assay system consisted of 3.3 ml of emulsion, 0.4 ml of a solution of bile salts (5%), 5.8 ml of water, and 0.5 ml of a 1:200 or 1:500 dilution of the original homogenate.

The bile salts used in the lipase assay were prepared as follows. 75 g of powdered ox bile extract (Nutritional Biochemicals Corp., Cleveland, Ohio) was added to 1500 ml of absolute ethanol, and the mixture was refluxed until most of the powder was in solution. Activated charcoal was added and refluxing was continued for several hours. The hot solution was filtered, and the filtrate was refluxed with fresh charcoal for several hours. This mixture was filtered, and the filtrate was concentrated until a slightly viscous solution was obtained. This solution was then added, with stirring, to 3 l of anhydrous diethyl ether. The bile salts, which precipitated, were collected by filtration, washed several times with anhydrous ether, and finally air dried. The bile salts, thus purified, dissolved readily in water to give a nearly colorless solution.

Amylase was assayed colorimetrically by measurement of the reducing groups (with 3,5-dinitrosalicylic acid) formed as a result of the action of the enzyme on soluble starch (Mallinckrodt) (1, 9).

Chymotrypsinogen and trypsinogen were measured by titrimetry (Radiometer pH-stat), using *N*-acetyl-L-tyrosine ethyl ester and *N*-benzoyl-L-arginine ethyl ester (Mann Research Laboratories, New York), respectively, as substrates (2).

Protein determinations were carried out according to the method of Lowry et al. (10) with crystalline bovine serum albumin as reference standard. DNA was measured according to the method of Schneider (11), using calf thymus DNA (Worthington Biochemical Corp., Freehold, N.J.) as a reference standard for phosphorus analyses.

In some experiments, blood was collected when the animals were killed, and concentrations of serum cholesterol, serum triglycerides, and blood glucose were determined. Cholesterol was measured by the method of Block, Jarrett, and Levine (12) on the AutoAnalyzer (Technicon Instruments Corp., Tarrytown, N.Y.). Serum triglycerides were measured according to Van Handel's modification (13) of the method of Van Handel and Zilversmit (14). Glucose was measured enzymatically with Glucostat (Worthington). All statistical comparisons were based on Student's *t* test.

RESULTS

Experiment A

The data in Table 2 show levels of total protein, lipase, and amylase in the pancreata of rats fed for different periods of time a chow diet containing added olive oil and cholesterol. The control group consisted of 11 rats that were killed just prior to initiating the feeding of the olive oil–cholesterol diet to other rats of similar weight. The enzyme levels in Table 2 are expressed on the basis of DNA content rather than total protein. This obviates apparent changes in specific enzymatic activity that could be due to differences in protein content of pancreata from rats on different diets rather than to differences in enzyme content. Specific enzymatic activities can be calculated by dividing units/μg of DNA P by mg of protein/μg of DNA P. After 6 days on the cholesterol–olive oil diet there was a slight increase in lipase levels, but significantly elevated levels were observed only after 10 days. At the end of 18 days there was a 50% increase. Concomitant with the increased lipase there were significantly lower levels of amylase.

Experiment B

Pancreatic enzyme and protein levels of animals fed chow diets supplemented with olive oil and/or cholesterol are presented in Table 3. This study was carried out

TABLE 2. Enzyme and protein levels in pancreata of rats fed chow diets supplemented with olive oil and cholesterol (experiment A)

Diet	Number of Rats	Mean Weight of Rats	Protein	Lipase	Amylase
		g	*mg/μg DNA P*	*units/μg DNA P*	
Chow (6.5% fat)	11	274	0.49 ± 0.01	46.8 ± 2.9	39.0 ± 1.3[a]
Chow, olive oil, and cholesterol (14.8% fat)[b]					
2 days	3	297	0.58	46.0	35.6
6 days	5	298	0.40 ± 0.02	58.7 ± 8.0	23.8 ± 2.8[c]
10 days	4	312	0.40 ± 0.04	65.1 ± 5.9[d]	21.7 ± 2.2[c]
18 days	4	330	0.54 ± 0.02	78.8 ± 2.6[e]	26.5 ± 2.2[c]

All results are expressed as means ± SEM.
[a] Mean of four animals. The mean total protein content in these rats was 0.50 mg/μg of DNA P.
[b] 90% chow, 9% olive oil, 1% cholesterol.
[c,d,e] $P < 0.005$, < 0.01, and < 0.001, respectively, compared with control values (chow diet, 6.5% fat). No P value indicates $0.05 < P < 0.5$.

in order to define more precisely the dietary constituent responsible for the rise in lipase levels observed in experiment A. The rats were killed after 14 days on the respective diets. The livers of the animals fed the olive oil–cholesterol diet were fatty; the livers of the other animals appeared normal.

Protein, lipase, and amylase levels for the animals on the control diet (chow) (Table 3) were lower than values for the control animals in the previous study (Table 2). These differences may have been due to the different strain of rat used and/or to the differences in the composition of the basic chow diets. The data in Table 3 show that significant elevation of lipase levels occurred in rats fed the olive oil diets with or without added cholesterol. The addition of cholesterol to the chow diet or to the chow and olive oil diet resulted in an increase in lipase levels (22.8 to 29.1 units/μg DNA P, and 42.2 to 52.6 units/μg DNA P, respectively), but these differences were not statistically significant. As in experiment A, amylase levels decreased appreciably in those animals with the high lipase values. Proteolytic enzyme levels were unaffected in the one group on which these assays were carried out.

Experiment D

Table 4 summarizes the data obtained in experiment D, in which rats were fed low and high fat diets (D-1 and D-2, respectively, and D-3, an almost fat-free diet) containing dextrose as the only carbohydrate.

There were differences in total protein levels among the various groups, and although some of these differences were statistically significant, no conclusions can be drawn with regard to biological significance.

Lipase levels in all animals fed the low fat diets (groups I, IV, and VI) were 49.8–54.9 units/μg of DNA P. The rats fed the high fat diet (groups II, III, and V) had higher levels of lipase (65.6–77.1 units/μg of DNA P). The increases in groups II and III as compared with group I were significant, with P values of < 0.025 and < 0.005, respectively.

TABLE 3. Enzyme levels in pancreata of rats fed chow diets supplemented with olive oil and/or cholesterol for 14 days (experiment B)

Diet[a]	Fat	Mean Weight of Rats	Total Protein	Lipase	Amylase	Chymotrypsinogen	Trypsinogen
	% by wt	*g*	*mg/μg DNA P*		*units/μg DNA P*		
Chow	4.5	454	0.34 ± 0.02	22.8 ± 1.9	17.6 ± 1.9	6.9 ± 0.7	1.1 ± 0.1
+ Cholesterol	4.4	430	0.42 ± 0.01[b]	29.1 ± 3.0	18.2 ± 1.2		
+ Olive oil	21.1	447	0.31 ± 0.01	42.2 ± 2.6[c]	10.6 ± 1.4[d]		
+ Cholesterol and olive oil	20.9	463	0.32 ± 0.01	52.6 ± 7.5[b]	9.0 ± 1.1[b]	7.2 ± 0.6	0.8 ± 0.1

All results are expressed as means ± SEM. There were six rats in each group.
[a] The compositions of these diets are indicated in the text.
[b,c,d] $P < 0.005$, < 0.001, and < 0.025, respectively, compared with appropriate control values (chow-fed rats).

TABLE 4. Enzyme levels in pancreata of rats fed a semipurified diet containing dextrose (experiment D) and various levels of fat

Group	Diet[a] (Days on Diet)	No. of Rats	Avg Body Wt	Total Protein	Lipase	Amylase	Chymotrypsinogen
			g	*mg/μg DNA P*		*units μg DNA P*	
I	D-1 (21)	7	386	0.42 ± 0.00	49.8 ± 3.0a,b	25.0 ± 1.2c,d	8.1 ± 0.6a
II	D-1 (21)	8	436	0.37 ± 0.01	65.6 ± 4.4a	12.6 ± 1.1c	10.6 ± 0.7a
	D-2 (14)						
III	D-1 (21)	8	475	0.45 ± 0.01	66.4 ± 2.8b	15.3 ± 0.8d	8.7 ± 0.5
	D-2 (41)						
IV	D-1 (36)	7	500	0.55 ± 0.00	54.9 ± 2.8c	29.7 ± 2.1a,e	11.6 ± 0.7
V	D-1 (30)	8	464				
	D-2 (42)		511	0.49 ± 0.01	77.1 ± 4.2c,d	15.0 ± 1.5e,f	8.8 ± 0.6c
VI	D-1 (28)	8	449				
	D-3 (41)[b]		482	0.60 ± 0.12	52.8 ± 2.5d	37.2 ± 1.7a,f	12.7 ± 0.6c

In each column, numbers with the same letter are significantly different from each other at the following levels: a, $P < 0.025$; b, $P < 0.005$; c, d, e, and f, $P < 0.001$.

[a] See Table 1 for compositions of diets.

[b] As linoleic acid.

The highest lipase levels were observed in group V, where the mean value was about 50% greater ($P < 0.001$) than that of each of the groups, IV and VI, fed the diets which contained less fat.

Amylase levels were also affected by diet. In rats fed the high fat diet (D-2), levels of amylase were considerably lower (50–60%) than those in animals fed diet D-1 only ($P < 0.001$ in all cases). Moreover, the level of amylase in the pancreata of the group IV animals fed D-1, a diet containing only 4% fat, was lower ($P < 0.025$) than that of the group VI rats fed diet D-3, which was essentially fat-free (only 0.5% linoleic acid).

Chymotrypsinogen. Although there were some significant differences between the low fat and high fat groups, there was no consistent pattern, and one must conclude that chymotrypsinogen levels were relatively unaffected by changing the fat content of the diet.

Experiment S

In experiment S, cornstarch was used instead of dextrose, and animals from each of the dietary groups were killed on the same day so that there was a control group for each age. Furthermore, studies were carried out to determine whether changes in enzyme levels were reversible.

Data from experiment S are given in Fig. 1. The data are similar to those for experiment D (Table 4). Animals fed the high fat diet for 20 or 60 days after the basal period (groups B and D, respectively) had significantly elevated levels of lipase and reduced levels of amylase compared with the control rats (groups A and C, respectively).

The animals in group E had been fed a high fat diet and then a low fat diet after the initial basal period. This resulted in low levels of lipase and high levels of amylase, and thus the data indicate that enzyme levels are readily reversed by diet (the animals in group E were originally in a larger group B). The group F rats had enzyme levels similar to those of animals in groups B and D.

The two rats in group G, except for the different periods of time on the low and high fat diets, had low

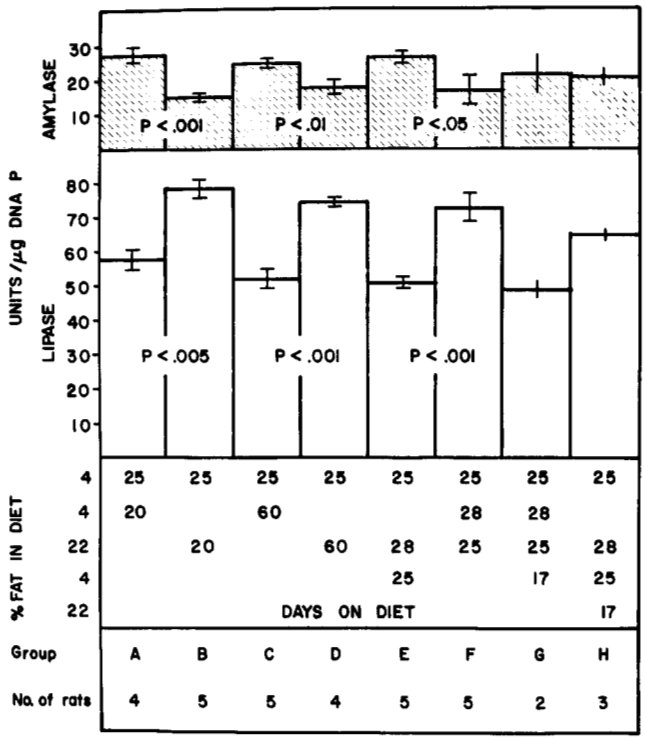

FIG. 1. Enzyme levels in pancreata of rats fed semipurified diets containing starch and various levels of corn oil (experiment S). Groups A and C, low fat; groups B, D, and F, low fat → high fat; groups E and G, low fat → high fat → low fat; group H, low fat → high fat → low fat → high fat.

TABLE 5. Effect of diet on concentrations of serum triglycerides and cholesterol

	Group	% Fat in Final Diet	Triglycerides	Cholesterol
			mg/100 ml serum	
Expt. D	I	4		80 ± 4
	II	22		80 ± 3
	III	22		85 ± 2
	IV	4	91 ± 15a	82 ± 6
	V	22	61 ± 8	96 ± 5b
	VI	0.5	48 ± 5a	69 ± 3b
Expt. S	A	4	79 ± 10	72 ± 2a
	B	22	79 ± 8	91 ± 4a
	C	4[a]	67 ± 11	80 ± 2c
	D	22[b]	95 ± 10	104 ± 5c
	E	4	68 ± 7	92 ± 3
	F	22	78 ± 14	107 ± 8

In each column, numbers with the same letter are significantly different at the following levels: a, $P < 0.025$; b, $P < 0.001$; and c, $P < 0.005$.

[a] Glucose = 102 mg/100 ml blood.

[b] Glucose = 98 mg/100 ml blood.

levels of lipase, although the average amylase value was intermediate between the high and low levels.

Group H consisted of animals whose diet was changed three times. The mean lipase level for this group was about 32% higher than that for group G. This difference was similar to that observed for the other groups in which the animals on the final 22% fat diet had lipase levels 36% (group B), 43% (group D), and 44% (group F) higher than the control values for groups A, C, and E, respectively. There was no difference in amylase levels between groups G and H, but this observation cannot be properly evaluated due to the small number of animals studied and the relatively large spread of values.

Concentrations of serum triglycerides and cholesterol were determined in most animals in experiments D and S; these data are shown in Table 5. Groups I–VI correspond to the same groups in Table 4 (experiment D); groups A–F correspond to the groups in Fig. 1 (experiment S). The only statistically significant difference in triglyceride concentrations was between groups IV (91 mg/100 ml of serum) and VI (48 mg/100 ml of serum). Lipase levels for both of these groups were low, 54.9 and 52.8 units/μg of DNA P, respectively. In general, animals on the high fat diets had cholesterol concentrations that were higher than the rats on the low fat diets; some of these differences were significant. However, the data in Table 5, in conjunction with data in Table 4 and Fig. 1, indicate that there is no direct or consistent relationship between high or low lipase levels and concentrations of lipids in serum sampled when the animals were killed.

DISCUSSION

The results of the present study show that levels of pancreatic lipase in the rat are increased when the fat content of the diet is raised from 4–6% to 15–22%. In two of the experiments (A and B), the animals were fed rat chow to which was added olive oil and/or cholesterol. In experiment A (Table 2), there was a 50% increase in pancreatic lipase levels in rats fed a high fat–cholesterol diet for 18 days; there were no statistically significant changes in rats fed the diet for 2 or 6 days. In experiment B (Table 3), the increase in lipase levels was 130% in rats fed a cholesterol–olive oil-supplemented diet for 14 days and 87% in rats maintained for 14 days on the same diet without the added cholesterol. These latter data suggest that the added olive oil was the dietary factor primarily responsible for the increase in lipase levels. Although the lipase levels in animals fed diets containing cholesterol were higher than those in animals fed the same diet (low or high fat) without the added cholesterol, the effect of cholesterol was not statistically significant.

By themselves, experiments A and B must be evaluated with a degree of caution. The unmodified commercial chow is undoubtedly adequate for normal growth and metabolism of rats. However, dilution of the chow with added fat and the consequent alteration of the proportion of the nutrients *could* have resulted in a diet that was only marginally adequate. It is for this reason that experiments were carried out in which semipurified diets of known composition were fed to rats. In these latter experiments, the high fat diets also resulted in elevated levels of pancreatic lipase and decreased levels of amylase. Thus, it is likely that in experiments A and B the changes in enzyme levels were due to the consequences of a high fat diet rather than to nutritional inadequacy of the diet.

There are at least five other studies (1, 2, 5–7) in which investigators have studied pancreatic lipase levels in rats fed high fat diets. In three of these investigations an adaptive response was reported, although the amount of fat in the diets, and in some cases the type of fat, was different than in the present experiments. Thus, Bučko and Kopec (5) fed low fat and high fat diets containing 9% and 58% corn oil, respectively; Snook's diets (6) contained 3.5–30% butter fat (low fat diets)[2] and 41% or 49% butter fat (high fat diets); and the diets used by Deschodt-Lanckman et al. (7) contained

[2] The designation of these diets as low fat diets was not made by Snook, but by the present author. It is based on the lack of significant differences (6) in lipase levels in rats fed these diets as compared to diets with more butter fat.

4% and 50% corn oil (low fat and high fat diets, respectively). The range of fat content in the present study was 4–6% (low fat) to 15–22% (high fat).

Snook (6) observed that significant increases in pancreatic lipase levels occurred only in rats fed diets containing 69% and 77% of the calories as butter fat (41% and 49%, respectively, by weight). Rats fed a diet containing 55% of the calories as butter fat (30% by weight) had levels of lipase that were elevated (about 15%) but not statistically different from levels in animals fed less fat. On the other hand, in the present study when diets containing 43.5% of the calories as corn oil (22% by weight) were fed to rats (experiments D and S), lipase levels were increased significantly.

Deschodt-Lanckman et al. (7) fed rats various types of fat at a level of 50% (by weight) of the diet. The diets also contained 38% (by weight) casein, but no carbohydrate. It was observed that increases in lipase levels in rats fed fats rich in unsaturated fatty acids were twice those in rats fed tricaprylin, tristearin, or lard. These investigators offered no clear-cut explanation of the poorer induction of pancreatic lipase by the saturated fats, lard and tristearin. However, it is possible that the rats did not absorb as much of these fats as the unsaturated fats, and thus the diet that was absorbed might have been relatively richer in protein (amino acids) and poorer in fat than the ration that was actually consumed. The poorer induction of lipase by tricaprylin was attributed to the rapid hepatic oxidation of caprylic acid (7). Perhaps fats which are hydrolyzed to yield fatty acids that are absorbed via the portal vein are relatively poor inducers of pancreatic lipase. This might explain some of the quantitative differences between the results from experiments D and S in the present study and those of Snook (6), who fed butter fat, which is relatively rich in short- and medium-chain fatty acids, to rats.

The failure of others (1, 2) to detect an increase in lipase levels in rats fed high fat diets deserves further comment. In the experiments of Grossman et al. (1), the high fat diet contained 54% lard. Thus, the failure of lipase levels to increase (there was actually a 13% decrease in this study) when the fat content of the diet was raised might have been related to the nature of the fat. In the studies of Reboud et al. (2), the control animals were fed a "régime équilibré" which contained 18% corn oil; the high fat diet in those studies contained 58% corn oil. The specific enzymatic activities of lipase in pancreas homogenates from rats fed each of these diets was essentially the same, 296 and 289 units/mg of protein (control and high fat, respectively). On the other hand, the high carbohydrate diet used by Reboud et al. (2) contained only 4% corn oil; the lipase in pancreas homogenates from rats fed this diet had an activity of 205 units/mg of protein. In the context of the present study, one may consider the high carbohydrate diet of Reboud et al. (2) to be a low fat regimen (4% fat).

TABLE 6. Summary of effect of level of dietary fat on pancreatic lipase

Low Fat	High Fat	Increase	Reference
% fat (by wt)[a]		%	
0.5[b]	4.0	4	Table 4
18	58	2.4	(2)
4	22	32–40	Table 4
4	22	36–44	Fig. 1
4	18	44	(2)
4	50[c]	ca. 200	(7)
9	58	33	(5)

[a] All diets, except as noted, contained the stated percentage of corn oil.
[b] 0.5% linoleic acid (no corn oil).
[c] This was the only diet that contained no carbohydrate.

On the basis of all the studies discussed and the present results, it would appear that the type and amount of dietary fat are important determinants in bringing about elevated levels of pancreatic lipase. Furthermore, the relative amounts of dietary fat, protein, and carbohydrate may influence the degree of change in lipase levels. Thus, the very large increases (ca. 200% in some cases) in specific activity of lipase (units/mg of total protein) reported by Deschodt-Lanckman et al. (7) might have been due in part to the very high (38%) protein content and lack of carbohydrate in the diet.

It is difficult to compare the increases in the various studies because the design of each investigation was different. However, it may be that above a certain level of a specific dietary fat (e.g., corn oil) no additional increase in lipase levels can be attained by a further increase in the amount of the fat in the diet. Table 6 illustrates this hypothesis. The data in the table are derived from some of the studies cited above (2, 5, 7) and from the results of the present investigation. Although it is not possible on the basis of the data in Table 6 to define exactly the limits of dietary fat content within which adaptation can occur, an approximate range would seem to be 4–20% for animals fed a semipurified diet containing on the order of 20% protein, with a maximal effect perhaps realized with 15–20% fat in the diet.

In the studies in which the rats were fed commercial chow diets, the relative increases in lipase activities were 1.5–4 times those observed for rats maintained on semipurified diets. It is unlikely that these differences were due to the olive oil (as opposed to corn oil used in the other diets). However, it is possible that the added

fat in conjunction with another component of the chow diet and/or the quality of the dietary protein might have been responsible for relatively high levels of lipase. The presence of cholesterol in two of the studies might also have had some effect.

Grossman, Greengard, and Ivy (15) also compared the differences between dietary dextrose and starch with regard to enzyme levels. They observed that animals on control diets (18% lard) in which dextrose was substituted for starch had higher levels of both lipase and amylase than did animals on the starch diet. In the present study there appeared to be no differences between dextrose and starch with regard to levels of these enzymes.

The observed increases in lipase levels pose two fundamental questions. Is the higher level of lipase due to an increase in the rate of its biosynthesis, a decrease in its rate of degradation, or a change in its specific enzymatic activity? On the basis of previous studies of amylase (3) and chymotrypsinogen (4), it is likely that an increase in the rate of biosynthesis is responsible for the increased levels of lipase. Since pancreatic lipase can be isolated in pure form (16), it is possible to test this hypothesis by appropriate radioactive studies.

The second question is concerned with the nature of the information which the pancreas receives concerning the composition of the diet consumed and how this information affects enzyme synthesizing systems or the enzyme itself in bringing about increases (or decreases) in enzyme levels. Ben Abdeljlil and Desnuelle (17) showed that levels of pancreatic amylase in rats fed a diet containing 75% glucose were similar to those in rats fed a diet containing 75% starch. They concluded that the product of hydrolysis (i.e., glucose) and the resulting elevation of plasma glucose concentrations brought about increased amylase biosynthesis rather than just the presence of starch in the diet. Snook (6) also considered the role of glucose in the pancreas in regulation of adaptation and concluded that intracellular rather than total pancreatic glucose may be the more important parameter to measure.

The response to changes in diet is relatively slow, as indicated in Table 2. Thus, a minimum time period for rats fed a given diet may be 6–10 days before an altered level of enzymes is elicited. It would also appear that there is no cause and effect relationship between enzyme levels and serum lipid concentration. Nor does it seem likely that blood glucose levels are related to lipase levels. One must consider, however, that changes in the secretion and/or levels of specific hormones, e.g., insulin or glucagon, that are consequences of a given diet may bring about secondarily changes in enzyme levels. Palla, Ben Abdeljlil, and Desnuelle (18) observed that in alloxan-diabetic rats amylase levels were greatly depressed while lipase activity was slightly elevated. If one assumes that normal values of amylase and lipase levels are those observed in pancreata of rats fed a low fat diet (e.g., 4%), the findings observed in the present study are consistent with the hypothesis that amylase levels might be expected to be lower than normal and lipase levels higher than normal in conditions where a relatively large percentage of calories are derived from the utilization of fat.

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