The Effect of Dietary Carbohydrate and Fat on the Activities of some Enzymes Responsible for Glycerolipid Synthesis in Rat Liver

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(Received 23 December 1977)

1. Male rats were fed for 14 days on diets containing (by wt.) 53% of starch, or on diets in which 20% of the starch was replaced by sucrose, corn oil or lard. 2. The hepatic activities of the microsomal glycerol phosphate acyltransferase, dihydroxyacetone phosphate acyltransferase, phosphatidate cytidyltransferase, diacylglycerol acyltransferase and choline phosphotransferase, and of the soluble phosphatidate phosphohydrolase, were measured. 3. The soluble phosphatidate phosphohydrolase activity was higher in those rats fed on lard than in those fed on the starch diet. Choline phosphotransferase activity was higher in the rats fed on corn oil than in those fed on the starch diet. 4. The rate of hepatic glycerolipid synthesis was measured *in vivo* 1 min after injection of [1,3-³H]glycerol and [1-¹⁴C]palmitate into the portal veins. 5. The relative rate of phosphatidylcholine synthesis *in vivo* was increased after feeding with corn oil and the higher specific activity of choline phosphotransferase may contribute to this result. The equivalent rate of triacylglycerol synthesis was increased by feeding with lard rather than corn oil, and the increased activity of phosphatidate phosphohydrolase may partly explain this. The latter changes probably contribute to the increased concentration of triacylglycerol which other authors have observed in the livers and sera of animals fed on saturated and monounsaturated fats.

It is well known that the concentrations of various lipids in the plasma and livers of animals can be altered by dietary manipulation, particularly by altering the amounts of dietary fructose and of saturated fat. These findings are important, since a relationship is thought to exist between the incidence of ischaemic heart disease and the concentrations of circulating lipids.

The purpose of the present investigation was to study the effect of dietary composition on the rate of hepatic glycerolipid synthesis observed *in vivo* and to relate this to the activity *in vitro* of the enzymes responsible for this synthesis. The basic diet consisted of 53% (by wt.) of starch, and in the three other diets 20% of the starch was replaced by sucrose, lard or corn oil.

Materials and Methods

Materials

The source of these has been described (Brindley *et al.*, 1976; Dodds *et al.*, 1976; Whiting *et al.*, 1977).

Animals

Male Wistar rats (160–210g) which had been fed on a 41B diet were purchased and maintained as

described previously (Brindley *et al.*, 1976). They were then fed *ad libitum* for 14 days on synthetic powdered diets before the experiments. The starch diet consisted of 53% (by wt.) of starch, 30% of casein, 10% of cellulose, 5% of corn oil and 2% of the vitamin-salt mixture (Brindley *et al.*, 1976). In the other three diets 20% of the starch was replaced by sucrose, corn oil or lard.

Preparation of subcellular fractions

The rats were killed by cervical dislocation and their livers were perfused with 0.15M-NaCl before preparation of the microsomal fraction and the particle-free supernatant (Mangiapane *et al.*, 1973; Whiting *et al.*, 1977). The soluble fraction was dialysed before being used to assay phosphatidate phosphohydrolase activity (EC 3.1.3.4) (Whiting *et al.*, 1977).

Measurement of enzyme activities

These were measured and were expressed relative to DNA P by using arylesterase (EC 3.1.1.2) as a marker for the recovery of endoplasmic-reticulum membranes (Whiting *et al.*, 1977).

Assay of dihydroxyacetone phosphate acyltransferase activity (EC 2.3.1.42). Each assay contained, in a volume of 0.25ml: 25mM-Tris buffer, adjusted

to pH 7.4 with HCl, 5 mM-dithiothreitol, 4.2 mM-MgCl₂, 8 mM-ATP, 66 μ M-CoA, 0.8 mM-potassium [¹⁴C]palmitate (1 μ Ci/ μ mol), 8 mM-dihydroxyacetone phosphate, 17 mM-NaF and 6 mg of fatty acid-poor bovine serum albumin/ml. The reactions were started with 50–200 μ g of microsomal protein and incubations containing no dihydroxyacetone phosphate were used as controls. Reactions were for 30 min at 37°C and were stopped with 1.88 ml of chloroform/methanol (1:2, v/v) and lipids were extracted by the method of Hajra *et al.* (1968). Samples of the bottom phase were immediately applied to t.l.c. plates of Kieselgel 60/Kieselguhr F₂₅₄ (Merck, Darmstadt, Germany) and the plates developed in chloroform/methanol/acetic acid/water (10:2:2:4:1, by vol.) for 60% of their lengths. The plates were then dried and developed for their total lengths with n-hexane/diethyl ether/acetic acid (60:40:1, by vol.) to ensure removal of all the unesterified [¹⁴C]-palmitate from the area of the plate containing the phospholipids. The position of [¹⁴C]palmitoyldihydroxyacetone phosphate was detected by using a spark chamber (Birchover Instruments, Hitchin, Herts., U.K.) and this area (*R_F* about 0.32 in the first solvent system) was scraped from the plate and its radioactivity determined by liquid-scintillation counting (Brindley *et al.*, 1976).

Measurement of glycerolipid synthesis *in vivo*

The method is described by Brindley *et al.* (1976) and the specific radioactivities of the [1-¹⁴C]palmitate and [1,3-³H]glycerol were 58 and 1638 μ Ci/ μ mol respectively.

Results

Effects of diet on body weight and liver composition

The growth of the rats on the four diets was not markedly different, although the rats fed on the lard

and corn-oil diets gained 4–6% more weight than did those fed on the starch or sucrose diets (Table 1). This weight was gained over the 14-day period and the weight-gain profiles were similar for the four groups. The mean liver weight of the lard-fed rats was greater than that of the rats fed on the starch or corn-oil diets (Table 1). However, when expressed relative to body weight the liver weights of the lard-fed rats were only greater than those fed on the starch diet. Since the livers of the sucrose-fed rats were perfused before measurement of enzyme activities, no liver weights were obtained. The rats described in Table 1 were used to investigate whether the composition of the diet altered the rate of glycerolipid metabolism in the liver.

Effect of diet on the activity of some enzymes concerned with glycerolipid synthesis

The effects of dietary modification on the specific activities of these enzymes are summarized in Table 2, and only two enzymes show significant changes. Feeding lard significantly increased the specific activity of the soluble phosphatidate phosphohydrolase relative to those rats fed on the starch diet. The specific activity of choline phosphotransferase (EC 2.7.8.2) was greater in those rats fed on corn oil than in those given the starch diet.

Effects of feeding corn oil or lard on the hepatic synthesis of glycerolipids *in vivo*

These experiments were performed to try to relate the enzymic changes observed in Table 2 to the hepatic synthesis of glycerolipid *in vivo*. This type of approach has been used to assess the enzymic changes caused by drug administration and by ethanol ingestion (Brindley *et al.*, 1976; Savolainen, 1977; Pritchard & Brindley, 1977; Pritchard *et al.*, 1977).

Table 1. *Effects of diet on body-weight change and on the composition of the liver*

Rats were fed on four different diets for 14 days as described in the Materials and Methods section. All results are expressed as mean $\pm$ S.D. and the number of rats is shown in parentheses. Significant differences between the groups were calculated by using a Student's *t* test and are indicated by: * *P* < 0.05; ** *P* < 0.02; *** *P* < 0.01; † *P* < 0.005; †† *P* < 0.001.

	(I) Starch diet	(II) Sucrose diet	(III) Lard diet	(IV) Corn-oil diet
Body-wt. change (g/100 g)	1 $\pm$ 5 (45) (I) versus (III)†† (I) versus (IV)††	1 $\pm$ 4 (10) (II) versus (III)† (II) versus (IV)**	7 $\pm$ 6 (48)	5 $\pm$ 4 (26)
Liver wet wt. (g)	6.6 $\pm$ 0.5 (19) (I) versus (III)†	Not measured	7.2 $\pm$ 0.7 (17) (III) versus (IV)***	6.6 $\pm$ 0.5 (17)
$\frac{\text{Liver wet wt.}}{\text{Body wt.}} \times 100$	3.5 $\pm$ 0.1 (19) (I) versus (III)††	Not measured	3.8 $\pm$ 0.2 (17) (III) versus (IV)*	3.6 $\pm$ 0.3 (17)
Total protein (mg/liver)	1046 $\pm$ 212 (9)	1115 $\pm$ 64 (9)	1210 $\pm$ 175 (10)	1134 $\pm$ 125 (9)
Microsomal protein (mg/liver)	392 $\pm$ 43 (9)	426 $\pm$ 87 (9)	411 $\pm$ 93 (10)	440 $\pm$ 96 (9)
DNA P (μ mol/liver)	86 $\pm$ 8 (9)	88 $\pm$ 12 (9)	84 $\pm$ 18 (10)	82 $\pm$ 6 (9)
Protein (g/mmol of DNA P)	12.0 $\pm$ 2.4 (9) (I) versus (III)*	12.8 $\pm$ 2.0 (9)	15.0 $\pm$ 3.3 (10)	14.0 $\pm$ 2.2 (9)

Table 2. *Effects of diet on the activities of some enzymes of hepatic glycerolipid biosynthesis*

Rats were fed on four different diets for 14 days as described in the Materials and Methods section. The number of animals in each group is shown in parentheses. Enzyme activities (mean $\pm$ s.d.) were measured in the microsomal fraction, except for the activity of the soluble phosphatidate phosphohydrolase, which was determined in the particle-free supernatant. The significant differences between the groups are expressed as in Table 1.

Enzyme activity	Activity (nmol of substrate converted/min per μ mol of DNA P)			
	(I) Starch diet	(II) Sucrose diet	(III) Lard diet	(IV) Corn-oil diet
Glycerol phosphate acyltransferase	63 $\pm$ 28 (10)	51 $\pm$ 24 (8)	54 $\pm$ 16 (10)	65 $\pm$ 21 (9)
Dihydroxyacetone phosphate acyltransferase	7.8 $\pm$ 1.2 (10)	7.3 $\pm$ 2.4 (8)	8.8 $\pm$ 2.7 (10)	9.2 $\pm$ 2.5 (9)
Soluble phosphatidate phosphohydrolase	5.17 $\pm$ 0.55 (8)	6.43 $\pm$ 1.95 (7)	6.90 $\pm$ 2.17 (8)	5.49 $\pm$ 1.56 (8)
Phosphatidate cytidyltransferase	(I) versus (III)*			
Diacylglycerol acyltransferase	7.2 $\pm$ 1.6 (8)	6.8 $\pm$ 1.6 (7)	7.5 $\pm$ 2.5 (8)	7.1 $\pm$ 1.6 (7)
Choline phosphotransferase	47 $\pm$ 10 (8)	51 $\pm$ 9 (7)	59 $\pm$ 24 (8)	54 $\pm$ 11 (7)
	32 $\pm$ 5 (8)	36 $\pm$ 6 (7)	41 $\pm$ 17 (8)	41 $\pm$ 9 (7)
	(I) versus (IV)*			

Table 3. *Effect of diet on the incorporation of [14 C]palmitate into hepatic lipids in vivo*

The uptake and distribution of [14 C]palmitate in the livers of rats was measured as described by Brindley *et al.* (1976) and the results are expressed as mean $\pm$ s.d. There were 16 rats fed on corn oil, 17 fed on lard and 19 fed on the starch diet. The significance of the difference between the dietary groups is indicated as described in Table 1.

	Incorporation of [14 C]palmitate					
	$10^{-3} \times$ d.p.m per liver			d.p.m./100 d.p.m. in lipid		
	(I) Starch diet	(III) Lard diet	(IV) Corn-oil diet	(I) Starch diet	(III) Lard diet	(IV) Corn-oil diet
Phosphatidylinositol+phosphatidylserine	64 $\pm$ 39	77 $\pm$ 39	64 $\pm$ 39	4 $\pm$ 2	4 $\pm$ 2	5 $\pm$ 3
Phosphatidylcholine	219 $\pm$ 77	270 $\pm$ 45	244 $\pm$ 90	16 $\pm$ 3	16 $\pm$ 3	18 $\pm$ 4
	(I) versus (III)*					
Phosphatidylethanolamine	77 $\pm$ 26	90 $\pm$ 26	77 $\pm$ 26	6 $\pm$ 1	5 $\pm$ 1	6 $\pm$ 1
Phosphatidate	244 $\pm$ 129	244 $\pm$ 77	270 $\pm$ 116	16 $\pm$ 3	14 $\pm$ 3	20 $\pm$ 6
				(I) versus (IV)**	(III) versus (IV)††	
Diacylglycerol	476 $\pm$ 167	540 $\pm$ 167	373 $\pm$ 154	31 $\pm$ 3	31 $\pm$ 4	27 $\pm$ 4
		(III) versus (IV)***		(I) versus (IV)†	(III) versus (IV)***	
Triacylglycerol	373 $\pm$ 116	501 $\pm$ 141	347 $\pm$ 167	25 $\pm$ 4	29 $\pm$ 5	24 $\pm$ 6
	(I) versus (III)***	(III) versus (IV)***		(I) versus (III)*	(III) versus (IV)***	
Flux from phosphatidate to diacylglycerol	1131 $\pm$ 308	1414 $\pm$ 321	1028 $\pm$ 411	79 $\pm$ 4	81 $\pm$ 3	74 $\pm$ 6
	(I) versus (III)**	(III) versus (IV)***		(I) versus (IV)***	(III) versus (IV)††	
14 C in whole liver lipid	2313 $\pm$ 874	3316 $\pm$ 964	2892 $\pm$ 926			
	(I) versus (III)†					
Non-lipid 14 C in liver	154 $\pm$ 64	296 $\pm$ 77	180 $\pm$ 116			
	(I) versus (III)††	(III) versus (IV)†				

The effects of fructose feeding have been well demonstrated (Lamb & Fallon, 1974; Fallon *et al.*, 1975, 1977) and therefore the sucrose-fed group was omitted. The present experiments concentrate on the effects of feeding corn oil or lard on hepatic glycerolipid synthesis.

A mixture of [14 C]palmitate and [3 H]glycerol was injected into the portal veins of anaesthetized rats (Brindley *et al.*, 1976) and the incorporation of these precursors into lipid 1 min later is shown in Tables 3 and 4. Under these condi-

Table 4. *Effect of diet on the incorporation of [³H]glycerol into hepatic lipids in vivo*

The uptake and distribution of [³H]glycerol was also measured in the rats described in Table 3 and the results are expressed in the same way. There were 16 rats each in the groups fed on corn oil and lard and 18 rats were fed on the starch diet.

	Incorporation of [³ H]glycerol					
	10 ⁻⁴ × d.p.m./liver			d.p.m./100 d.p.m. in lipid		
	(I) Starch diet	(III) Lard diet	(IV) Corn-oil diet	(I) Starch diet	(III) Lard diet	(IV) Corn-oil diet
Phosphatidylinositol+phosphatidylserine	17 ± 7 (I) versus (III)*	23 ± 10	20 ± 10	4 ± 2	5 ± 2	5 ± 3
Phosphatidylcholine	46 ± 14 (I) versus (III)**	57 ± 10	55 ± 22	11 ± 3 (I) versus (IV)***	12 ± 2 (III) versus (IV)*	14 ± 3
Phosphatidylethanolamine	27 ± 9 (I) versus (III)†	36 ± 8	31 ± 13	7 ± 2	7 ± 2	8 ± 2
Phosphatidate	56 ± 12	62 ± 11	62 ± 24	14 ± 3 (I) versus (IV)**	13 ± 2 (III) versus (IV)***	17 ± 5
Diacylglycerol	141 ± 41	169 ± 41 (III) versus (IV)†	113 ± 51	34 ± 5 (I) versus (IV)††	34 ± 5 (III) versus (IV)††	28 ± 4
Triacylglycerol	109 ± 31	132 ± 35 (III) versus (IV)**	94 ± 44	26 ± 4	26 ± 5	24 ± 6
Flux from phosphatidate to diacylglycerol	324 ± 75 (I) versus (III)***	395 ± 73 (III) versus (IV)*	312 ± 127	80 ± 6 (I) versus (IV)***	79 ± 4 (III) versus (IV)***	74 ± 6
Total lipid ³ H in whole liver	383 ± 86 (I) versus (III)†	487 ± 88	409 ± 125			
Non-lipid ³ H in liver	2760 ± 681 (I) versus (III)**	3422 ± 803	2985 ± 706			

tions the rate of appearance of radioactivity in triacylglycerol, phosphatidylethanolamine and phosphatidylcholine is constant with time.

Comparison of the rats fed on the starch and lard diets. The uptake of [¹⁴C]palmitate and [³H]glycerol into the livers was greater in the rats fed on lard (Tables 3 and 4). This increase was seen in both the lipid and the non-lipid fractions. The incorporation of [³H]glycerol (in d.p.m.) into phosphatidylserine plus phosphatidylinositol, phosphatidylcholine and phosphatidylethanolamine was also increased in the lard-fed rats. However, an increased incorporation of [¹⁴C]palmitate was only observed into phosphatidylcholine and triacylglycerol. The flux of both precursors from phosphatidate to diacylglycerol was greater in the rats fed on lard. The only difference between these two groups in terms of the percentage distribution of radioactivity in lipids was the increased proportion of [¹⁴C]palmitate incorporated in triacylglycerol in the rats fed on lard.

Comparison of the rats fed on the starch and corn-oil diets. The relative accumulations of [³H]glycerol and of [¹⁴C]palmitate in diacylglycerol and the flux of phosphatidate to diacylglycerol were lower in the rats fed on the corn-oil diet. Relatively more [³H]-

glycerol and [¹⁴C]palmitate was recovered in phosphatidate (Table 3) and relatively more [³H]glycerol was isolated in the phosphatidylcholine obtained from these rats.

Comparison of the rats fed on the lard and corn-oil diets. The rats fed on the corn-oil diets synthesized less diacylglycerol and triacylglycerol (in d.p.m.) from both [¹⁴C]palmitate and [³H]glycerol (Tables 3 and 4). This group had a smaller proportion of both precursors in diacylglycerol and a larger proportion in phosphatidate. The rats fed on the corn-oil diet showed a relatively greater accumulation of [³H]-glycerol in phosphatidylcholine and relatively less [¹⁴C]palmitate in triacylglycerol than did those rats fed on lard.

Discussion

It is likely that the initial entry of fatty acids into the pathway of glycerolipid synthesis is controlled by a competition for acyl-CoA esters among the enzymes which catalyse the first reactions that commit the fatty acids to a metabolic fate. For instance, the increased specific activity of carnitine palmitoyl-transferase (EC 2.3.1.21) relative to that of glycerol

phosphate acyltransferase may help to direct fatty acids towards β -oxidation rather than to esterification in the livers of starved rats (Aas & Daae, 1971). The initial reactions of fatty acid esterification in liver are catalysed by glycerol phosphate acyltransferase and dihydroxyacetone phosphate acyltransferase, and both of these routes have the potential to synthesize the major glycerolipids (Manning & Brindley, 1972; Pollock *et al.*, 1975). However, the relative importance of these two pathways under different physiological conditions is unknown. The conversion of glucose into triacylglycerol via the dihydroxyacetone phosphate pathway requires NADPH, whereas the conversion via the glycerol phosphate pathway involves NADH. NADPH is normally associated with reactions of reductive synthesis, as opposed to NADH, which is usually involved in degradative pathways. These observations have been used as an argument in favour of the dihydroxyacetone phosphate pathway (Harding *et al.*, 1975; Pollock *et al.*, 1975).

It might also be expected that the activity of the dihydroxyacetone phosphate pathway should be high in conditions of enhanced NADPH production and fatty acid biosynthesis. There is some evidence to support this view. Rao *et al.* (1971) reported that the addition of 15% of safflower oil to a fat-free diet for 7 days, or starvation for 3 days, decreased the rate of phosphatidate synthesis from dihydroxyacetone phosphate in microsomal fractions of rat liver. Lamb & Fallon (1974) showed that feeding with diets containing 75% of fructose or glucose increased the synthesis of total and neutral lipids from dihydroxyacetone phosphate by 2–3-fold. In both of these investigations NADH was used as a reducing cofactor and the results were expressed relative to protein. In Table 2 the results are expressed relative to DNA P, but similar trends in the activity of this enzyme are seen when this activity is related to protein. The specific activity of dihydroxyacetone phosphate acyltransferase does not seem to parallel the expected changes in the rate of fatty acid biosynthesis, since it appears to be slightly increased in the rats fed on the corn-oil and lard diets (Table 2). This increase is not significant ($P > 0.05$) with the numbers of rats used. However, the specific activity of dihydroxyacetone phosphate acyltransferase was increased in the adipose tissue of rats fed on a diet containing 20% (w/w) of beef tallow (Dodds *et al.*, 1976). This increase appeared to be correlated with the energy intake and may not have been caused by a specific effect of the tallow.

It has been claimed that the microsomal activities of glycerol phosphate acyltransferase and dihydroxyacetone phosphate acyltransferase of liver are catalysed by the same enzyme (Hajra *et al.*, 1968; Schlossman & Bell, 1977). There was a significant correlation ($r = 0.50$; $P < 0.01$) between these two

activities in the present experiments, which is compatible with this conclusion. However, there appears to be a distinct dihydroxyacetone phosphate acyltransferase in the mitochondrial fraction of rat liver (Hajra, 1968; Bowley *et al.*, 1973), in the microsomal fraction of rabbit harderian gland (Rock *et al.*, 1977) and possibly in the mitochondrial fraction of rat adipose tissue (Dodds *et al.*, 1976).

The results in Tables 3 and 4 indicate that the hepatic uptake and esterification of the exogenously injected [^3H]glycerol and [^{14}C]palmitate are greater in the rats fed on lard than in those fed on the starch diet. If so, this would involve an increased flux through the initial glycerol phosphate acyltransferase and dihydroxyacetone phosphate acyltransferase reactions. The interpretation of the experiments in Tables 3 and 4 has to be limited, since the extent of isotope dilution is not taken into account (Brindley *et al.*, 1976), even though this dilution may be greater in the lard-fed rats. The specific activities of dihydroxyacetone phosphate acyltransferase and of glycerol phosphate acyltransferase were not significantly altered by dietary modification in the present experiments. Glycerol phosphate acyltransferase does not appear to be rate-limiting in hepatic triacylglycerol synthesis *in vitro*, nor does its activity correlate well with the rates of triacylglycerol synthesis observed *in vivo* (Brindley, 1978). The flux of fatty acids to phosphatidate may therefore be largely dependent on substrate availability.

The activity that normally parallels the potential to synthesize triacylglycerols is that of phosphatidate phosphohydrolase (Brindley, 1978). The addition of fructose to the diet is known to increase the hepatic activity of phosphatidate phosphohydrolase (Lamb & Fallon, 1974; Fallon *et al.*, 1977) and to decrease the microsomal content of phosphatidate while increasing that of diacylglycerol (Fallon *et al.*, 1975). The increased specific activity of the phosphatidate phosphohydrolase parallels the increased hepatic synthesis of triacylglycerol and probably contributes to the fructose-induced hypertriglyceridaemia (Lamb & Fallon, 1974; Fallon *et al.*, 1977). In these experiments fructose constituted up to 70% of the diet, whereas in Table 2 it contributed the equivalent of 10% by wt. This concentration of fructose may have been too low to produce significant alterations in the hepatic phosphatidate phosphohydrolase activity. However, when a similar diet was fed to rats it did increase the rate at which phosphatidate was converted into diacylglycerol in homogenates of the adipose tissue (Dodds *et al.*, 1976).

The specific activity of the soluble phosphatidate phosphohydrolase was higher in the livers of rats fed on the lard diet than in those fed on the corn-oil and starch diets, but the difference was only significant in the latter case (Table 2). A greater proportion of the injected [^{14}C]palmitate and [^3H]glycerol passed

through the phosphatidate phosphohydrolase reaction in the rats fed with lard than in those fed on the corn-oil and starch diets (Tables 3 and 4). However, when the flux from phosphatidate to diacylglycerol is expressed as a percentage of the radioactivity in glycerolipid, the results for the rats fed on the starch and lard diets are similar and greater than those for the rats fed on the corn-oil diet. The activity of phosphatidate phosphohydrolase may therefore not be the only factor influencing the rate of hepatic triacylglycerol synthesis in these experiments.

Feeding with saturated fat rather than polyunsaturated fat can increase the concentration of serum triacylglycerols (MacDonald, 1971; Bruckdorfer *et al.*, 1972; Chait *et al.*, 1974; Waterman *et al.*, 1975). One explanation for this is that the activity of adipose-tissue lipoprotein lipase (EC 3.1.1.34) is increased by feeding with polyunsaturated fat and therefore the circulating triacylglycerol is removed more quickly (Pokrajac & Lossow, 1967; Pawar & Tidwell, 1968; Bagdale *et al.*, 1970; Weisenburg Delorme & Harris, 1975; Cryer *et al.*, 1978). Tables 3 and 4 show that more of the injected [^3H]glycerol and [^{14}C]palmitate was recovered in hepatic triacylglycerol in the rats fed on lard than in those fed on the starch and corn-oil diets and that a greater percentage of the esterified [^{14}C]palmitate was recovered in triacylglycerol. The results in Tables 2, 3 and 4 indicate that the capacity of the liver to synthesize triacylglycerols is increased when rats are fed on diets enriched with triacylglycerols containing predominantly saturated and monounsaturated fatty acids. This increased synthesis could also contribute to: (a) the increased liver weight of the lard-fed rats (Table 1) and the accumulation of lipids observed in the livers of rats fed on saturated and monounsaturated fat (Bruckdorfer *et al.*, 1972; Laitinen *et al.*, 1975), and (b) the increased production of very-low-density lipoproteins and the hyperlipidaemia observed during the feeding with saturated compared with polyunsaturated fats (Chait *et al.*, 1974).

A common feature which appears to accompany an increase in activity of hepatic phosphatidate phosphohydrolase is an increased supply of saturated and monounsaturated fatty acids to the liver. The predominant fatty acids of adipose tissue are normally of this type and when they are mobilized and their flux to the liver is increased, the activity of phosphatidate phosphohydrolase rises. This is seen in starvation (Vavrečka *et al.*, 1969; Mangiapane *et al.*, 1973), stress (Mangiapane *et al.*, 1973) and in diabetes (Whiting *et al.*, 1977). The obese state (Fallon *et al.*, 1977) and the ingestion of diets rich in glucose or fructose (Lamb & Fallon, 1974) is accompanied by an increased synthesis of saturated and monounsaturated fatty acids in the liver and by an increased phosphatidate phosphohydrolase activity. One of the consequences of ethanol ingestion is an

increase in the hepatic uptake of fatty acids from the circulation (Bode & Goebell, 1971; Johnson *et al.*, 1975; Abrams & Cooper, 1976a,b) and to increase the incorporation of dietary fats into triacylglycerols. This is accompanied by a rapid rise in hepatic phosphatidate phosphohydrolase activity (Lamb & Fallon, 1977; Savolainen, 1977; Pritchard *et al.*, 1977). Although the ethanol-induced fatty liver is aggravated by a diet which contains more than 20% of its energy as fat (Chen *et al.*, 1977), the fatty liver is seen when either saturated or polyunsaturated fats are used (Jones & Greene, 1966; Carroll & Williams, 1971).

The type of fat in the diet might also be expected to influence the relative rates of synthesis of the major phospholipids and of triacylglycerol, since unsaturated fatty acids are preferentially incorporated into phosphatidylcholine and particularly into phosphatidylethanolamine (Åkesson *et al.*, 1970; Sundler *et al.*, 1974; Kanoh & Ohno, 1975). Table 4 also shows that the relative rate of synthesis of phosphatidylcholine, measured by using [^3H]glycerol as the marker, is greater in the rats fed on corn-oil diet than in those fed on the starch and lard diets. It occurs despite a decrease in the net flux of [^3H]glycerol from phosphatidate to diacylglycerol in the rats fed on corn oil. This finding is probably explained by a combination of the preferential incorporation of diacylglycerols containing polyunsaturated fatty acids into phosphatidylcholine (Åkesson *et al.*, 1970; Sundler *et al.*, 1974; Sarzala & van Golde, 1976) and the increased specific activity of choline phosphotransferase (EC 2.7.8.2) (Table 2).

The object of the present paper was to document the effects of some dietary modifications on the activities of enzymes involved in glycerolipid synthesis. The changes found could contribute to the alterations in the metabolism of glycerolipids and lipoproteins that are observed *in vivo*.

We thank the Medical and Science Research Councils for financial support.

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