are apparently correlated with changes in dihydroxyacetone phosphate concentration. In non-diabetic rats they are both decreased in starvation and increased 48 h after re-feeding with glucose, glycerol or sucrose. Further, in diabetes the activity of liver-type pyruvate kinase and dihydroxyacetone phosphate concentration are decreased, and in this state both are raised by treatment with insulin and by re-feeding with sucrose or glycerol but not with glucose. However, no causal relationship has been established.

The authors are indebted to the Science Research Council for a grant to J. M. G.

- Bailey, E., Taylor, C. B. & Bartley, W. (1968) *Nature (London)* **217**, 471
 Sillero, A., Sillero, M. A. G. & Sols, A. (1969) *Eur. J. Biochem.* **10**, 351
 Weber, G., Singhal, R. L., Stamm, N. B., Lea, M. A. & Fisher, E. A. (1966a) *Advan. Enzyme Regul.* **4**, 59
 Weber, G., Lea, M. A., Fisher, E. A. & Stamm, N. B. (1966b) *Enzymol. Biol. Clin.* **7**, 11

The Effect on the Activities of some Hepatic Enzymes of Weaning Rats on to a Diet High in Sucrose Content

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There is a marked increase in hepatic lipogenesis and the activities of enzymes associated with lipogenesis on weaning rats from the high-fat-content milk diet of suckling to the high-carbohydrate (starch)-content laboratory diet (Taylor *et al.*, 1967; Lockwood *et al.*, 1970), Hepatic lipogenesis is also considerably higher in rats fed on sucrose rather than on starch-containing diets (Bailey *et al.*, 1968). To investigate further the involvement of diet in the post-weaning increase in lipogenesis we have studied the effect on the activity of some enzymes involved in carbohydrate conversion into fatty acids of weaning female rats on to a diet high in sucrose content.

Fructokinase (EC 2.7.1.4) activity increased after weaning (at 21 days of age) to a maximum at 40–45 days and then fell to pre-weaning values by the end of the experiment (70 days of age), and feeding with sucrose had little effect on the enzyme activity. Phosphofructokinase (EC 2.7.1.11) and fructose 1,6-diphosphatase (EC 3.1.3.11) activities varied little during the period studied and dietary sucrose had little effect on the activity of either enzyme. Aldolase (EC 4.1.2.13) activity was constant between 21 days and 40 days of age, rose to a maximum at 50 days and then fell to 70 days of age. Feeding with sucrose caused slightly elevated aldolase activities at all ages studied. Pyruvate kinase (EC 2.7.1.40) activity rose steadily during the experiment to a maximum at about 50–60 days of age, and activities in the livers

of sucrose-fed animals were always higher than those of controls. ATP citrate lyase (EC 4.1.3.8) and 'malic' enzyme (EC 1.1.1.40) increased in activity after weaning to a maximum at 35–40 days of age and then fell to the end of the experiment, and feeding with sucrose caused elevated activities at all ages studied. Extramitochondrial aconitate hydratase (EC 4.2.1.3) and NADP-dependent isocitrate dehydrogenase (EC 1.1.1.42) activities fell after weaning until 25–30 days of age and then rose throughout the rest of the experimental period. From 30 days of age the activities of both these enzymes were lowered by feeding with sucrose. Glucose 6-phosphate dehydrogenase (EC 1.1.1.49) activity rose rapidly after weaning to 40 days of age and then remained constant for the remainder of the experimental period. Feeding with sucrose increased this enzyme activity at all ages studied.

We thank the Medical Research Council for a grant to J. R. W.

- Bailey, E., Taylor, C. B. & Bartley, W. (1968) *Nature (London)* **217**, 471–472
 Lockwood, E. A., Bailey, E. & Taylor, C. B. (1970) *Biochem. J.* **118**, 155–162
 Taylor, C. B., Bailey, E. & Bartley, W. (1967) *Biochem. J.* **105**, 717–722

Changes in the Activities of some Kidney Enzymes during Development of the Rat

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Although changes in the activities of many hepatic enzymes during development of the rat are well documented, little is known about the developmental changes that occur in the activity of rat kidney enzymes (for review see Greengard, 1971). We have measured the postnatal changes that occur in the activities of the following kidney enzymes: hexokinase (EC 2.7.1.1), pyruvate kinase (EC 2.7.1.40), 'malic' enzyme (EC 1.1.1.40), glucose 6-phosphate dehydrogenase (EC 1.1.1.49), extramitochondrial NADP-dependent isocitrate dehydrogenase (EC 1.1.1.42), extramitochondrial aconitate hydratase (EC 4.2.1.3), ATP citrate lyase (EC 4.1.3.8) and fructose 1,6-diphosphatase (EC 3.1.3.11). We have also measured developmental changes in the incorporation of [^{14}C]acetate into lipid by kidney homogenates. The experiments were carried out with both male and female rats weaned at 21 days of age, and unless otherwise stated similar results were obtained with both sexes.

Hexokinase activity increases after birth to a maximum at 30 days of age, thereafter falling to reach adult values by 50 days of age. These results extend

the findings of Sydow (1969). The activity of pyruvate kinase, the other glycolytic enzyme studied, changed very little during the period investigated. Glucose 6-phosphate dehydrogenase activity fell slightly after birth to a minimum at 20 days of age and then rose gradually to reach adult values at 50 days of age. Isocitrate dehydrogenase and 'malic' enzyme exhibited similar developmental patterns, the activities of both enzymes rising steadily throughout postnatal development. The activity of kidney 'malic' enzyme was 50% higher in female rats than in male rats.

Extramitochondrial aconitate hydratase activity increased after birth, reaching adult values by 20 days of age. ATP citrate lyase activity, which was very low throughout development, fell after birth to a minimum at 20 days of age and then rose steadily to adult values. [^{14}C]Acetate incorporation into lipid rose after birth to a maximum at 20 days of age and then fell to a minimum at 50 days of age, thereafter rising to adult values. Fructose 1,6-diphosphatase activity rose after birth, reaching adult values at about 30 days of age, thus exhibiting a similar developmental pattern to that of glucose 6-phosphatase and gluconeogenesis reported by Zorzoli *et al.* (1969).

We thank the Medical Research Council for grants to C. A. H. and E. A. L.

Greengard, O. (1971) *Essays Biochem.* 7, 159–205

Sydow, G. (1969) *Hoppe-Seyler's Z. Physiol. Chem.* 350, 263–268

Zorzoli, A., Turkenkopf, I. J. & Mueller, V. L. (1969) *Biochem. J.* 111, 181–185

Effects of Protein Deficiency on Uridine Diphosphate Glucuronyltransferase Activity and Phospholipid Composition of Rat Liver Microsomal Fraction

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Protein deficiency markedly increases the activity of rat liver microsomal UDP-glucuronyltransferase with *o*-aminophenol or *p*-nitrophenol as acceptor (Wood & Woodcock, 1970; Woodcock & Wood, 1971). Since the enzyme's activity is affected by treatment of microsomal preparations with agents perturbing the phospholipids of the microsomal membrane (Graham & Wood, 1969; Attwood *et al.*, 1971; Hänninen & Puukka, 1971), it was suggested (Woodcock & Wood, 1971) that protein deficiency might affect enzyme activity by altering the structure of the microsomal membrane. We have therefore determined the phospholipid composition of liver microsomal preparations from normal and protein-deficient rats.

Male Wistar rats were force-fed on a protein-free purified diet for 5–7 days. In agreement with our previous work, the activity of microsomal UDP-glucuronyltransferase in livers from these animals was substantially higher than in those from control animals fed on the same diet supplemented with 18% casein (protein-deficient, 148.7 ± 13.4 nmol of glucuronide formed/h per mg of microsomal protein; controls, 63.2 ± 6.8).

Although the weights of microsomal protein and phospholipid per g of liver were both 30% lower in protein-deficient animals than in control animals, there was no significant difference in the ratio of phospholipid to protein between the groups (protein-deficient, 1.07 ± 0.03 g/g; controls, 1.00 ± 0.01 g/g).

The microsomal phospholipid compositions were similar in the two groups, but there were significant differences in the content of phosphatidylinositol (protein-deficient, $6.1 \pm 0.5\%$ of total phospholipid; controls $8.7 \pm 0.3\%$) and sphingomyelin (protein-deficient, $7.0 \pm 0.6\%$; controls, $4.2 \pm 0.3\%$). Moreover, lysophosphatidylcholine ($7.4 \pm 0.5\%$) and lysophosphatidylethanolamine ($3.1 \pm 0.9\%$) were found with protein-deficient rats whereas no lysophosphatides were detected with control animals.

Hänninen & Puukka (1971) report that addition of lysophosphatides to rat liver microsomal fractions or their treatment with phospholipase A increases UDP-glucuronyltransferase activity. Treatment of our control microsomal preparations with phospholipase A showed that a degree of phospholipid hydrolysis similar to that observed in preparations from protein-deficient rats increased enzyme activity 2–3-fold.

We therefore suggest that the elevated enzyme activity in microsomal fractions from protein-deficient rats is probably due to the presence of lysophosphatides. The involvement of other factors such as the alterations of phosphatidylinositol and sphingomyelin contents cannot, however, be excluded.

We gratefully acknowledge support for this work from the Medical Research Council and the Science Research Council.

Attwood, D., Graham, A. B. & Wood, G. C. (1971) *Biochem. J.* 123, 875–882

Graham, A. B. & Wood, G. C. (1969) *Biochem. Biophys. Res. Commun.* 37, 567–575

Hänninen, O. & Puukka, R. (1971) *Chem.-Biol. Interact.* 3, 282–284

Wood, G. C. & Woodcock, B. G. (1970) *J. Pharm. Pharmacol.* 22, 60S–63S

Woodcock, B. G. & Wood, G. C. (1971) *Biochem. Pharmacol.* 20, 2703–2713