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Glycine acyltransferase activity in developing rat liver

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The detoxication of benzoic acid and similar substances in mammalian tissues is brought about in three stages.[1] Benzoyl adenylate is formed by pyrophosphate exchange with ATP. The adenylate moiety is then exchanged for coenzyme A to produce benzoyl-CoA. Finally, the latter reacts with glycine to give hippuric acid with the regeneration of free CoA. The first two reactions are catalyzed by a thiokinase, the last by glycine acyltransferase (E.C. 2.3.1.13[2]), the specificity of which is indicated by its systematic name: acyl-CoA: glycine N-acyltransferase. The three activities are found only in the mitochondrial fraction of liver and kidney.[3]

Hippuric acid synthesis by liver homogenates has been previously shown to vary with the age of the animal.[4] The present study demonstrates that the activity of the glycine acyltransferase component of the system also varies with the age of the animal and in a similar pattern.

METHODS

Rats of the Sprague-Dawley strain were fed a standard laboratory diet (D & G, The Price-Wilhoite Co.). The approximate time of conception was determined by vaginal smear the morning after an overnight period of mating. Fetal rats were obtained by hysterotomy under ether anesthesia. Older animals were killed by decapitation without anesthesia.

All animals 20 days of age or older were virgin females. In assays involving fetal and newborn animals the livers of one to three litters were used for each experiment. Freshly excised livers were

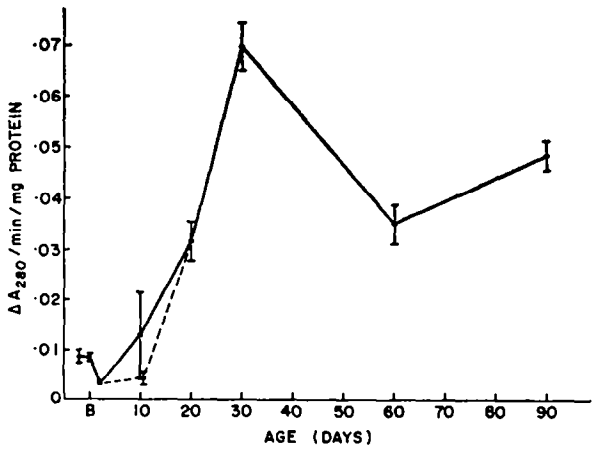

Fig. 1. The variation of glycine acyltransferase activity of rat liver with age. "B" indicates time of birth. Means ± S.E. are given. The broken line indicates value calculated when one markedly deviant result was omitted from the data on 10-day old animals.

homogenized in 9 vol. of cold 0·25 M sucrose and the mitochondria isolated by differential centrifugation, washed four times, and made into acetone powders. Extracts of the latter were made and were assayed for glycine acyltransferase as described by Schachter and Taggart;[1] this consisted of measuring the decrease in absorbance at 280 mμ as the thioester bond of benzoyl-SCoA is cleaved. Protein was determined by the biuret reaction.

RESULTS

The enzyme activities of material prepared from animals of different ages are shown in Fig. 1. All points represent the results of six experiments except that five experiments were performed on 30-day-old. animals, and four experiments were performed on 19-day fetuses and on 90-day-old animals. There is relatively little activity in fetal and early newborn life, and by the time of weaning (20 days) an appreciable increase has taken place. This proceeds to a maximum at 30 days of age then decreases to a small extent. This development pattern is similar to that described for hippuric acid synthesis by liver homogenates,[4] suggesting that glycine acyltransferase may be the rate-limiting step in the overall reaction. These experiments further define the probable biochemical basis for the reduced ability of newborn infants to metabolize *p*-aminobenzoic acid.[5]

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The effect of carbaryl (1-naphthyl-N-methyl carbamate) on blood glucose, and liver and muscle glycogen in fasted and nonfasted rats

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WEISS *et al.*[1] have reported that intraperitoneal administration of toxic doses of carbaryl (1-naphthyl-N-methyl carbamate) produced a pronounced hyperglycemic response in nonfasted rats and that this response was absent in adrenalectomized rats.[2] This suggests that the hyperglycemia induced by carbaryl may be due to the release of epinephrine from the adrenal gland.

Fleming and Kenny[3] reported that subcutaneous injections of several catechol amines produced hyperglycemia and glycogenolysis in skeletal muscle of both fasted and nonfasted rats. Although the blood glucose rise was lower in fasted rats, the glycogenolytic response in the muscle was independent of the prandial state of the animals.

The purpose of the present study was to determine what effect the release of endogenous catecholamines by carbaryl would have on blood glucose and liver and muscle glycogen of fasted and nonfasted rats. The two doses of carbaryl chosen were 5 mg/kg, which produced minimal, if any, cholinergic effects, and 25 mg/kg, which caused pronounced signs of cholinergic stimulation.