(3) Pathogenesis of Hepatic Porphyria

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Introduction and Methods

Main problems concerning the pathophysiology in hepatic porphyria are the mechanisms through which the disorders of porphyrin metabolism and nervous system arise. As reported previously¹⁾, in a biopsy specimen of the liver of a patient with acute intermittent porphyria, δ -aminolevulinic acid (ALA) synthetase activity was markedly increased and ALA dehydrase activity was about twice the control, while porphobilinogen (PBG)-ase and chelatase activities were almost unchanged. In this study inductions of the liver microsomal cytochromes, P-450 and b_5 , and mitochondrial synthetase were attempted in mice by treatment with various porphyria-inducing chemicals and several other drugs known to precipitate acute attacks in human hepatic porphyria. The former chemicals studied include 3, 5-dicarbethoxy-1, 4-dihydrocollidin (DDC) (500 mg./kg.), AIA (100 mg./kg.), griseofulvin (5% in diet) and C_6Cl_6 (5% in diet) and the latter drugs include phenobarbital (80 mg./kg.), tolbutamide (5% in diet) progesterone (50 mg./kg.) and estradiol (0.1 mg./kg.). All drugs were administered every 12 hours. Activity of ALA synthetase was assayed by the method of Granick and Urata²⁾, cytochrome P-450 and b_5 contents by the method of Ohmura and Sato³⁾, and N-demethylase activity by the method of Terayama⁴⁾.

Results and Discussion

One gram of normal liver synthetizing about 20 m μ moles ($20.0 \pm S.E. 1.2$) ALA per hour contained approximately 24 m μ moles ($24.1 \pm S.E. 2.2$) cytochrome P-450 and 10 m μ moles ($10.3 \pm S.E. 0.92$) cytochrome b_5 .

Changes in P-450 content and ALA synthetase activity were illustrated in Figs. 1 and 2. The levels of b_5 resembled the pattern of P-450, although the phase was shifted about 12 hours probably due to longer turnover rate. Activity of N-demethylase paralleled to the P-450 content.

Accumulation of porphyrin was most prominent in the liver of mice treated with DDC. Moderate increase was observed in the liver of AIA or griseofulvin poisoned mice, whereas scarcely any increase was seen when mice were treated with phenobarbital, tolbutamide or female sex hormones. Administration of DDC before or after the treatment with phenobarbital inhibited the increasing tendency of P-450 content. Catalase activity was decreased in all porphyric mice. DDC showed an inhibitory effect on the incorporation of ^{14}C -glycine into both microsomal and mitochondrial heme. Less incorporation of ^{14}C -glycine into microsomal heme than into mitochondrial heme

Fig. 1. Enhanced rate of the mitochondrial ALA synthetase activity during treatment of mice with various chemicals and drugs.

Fig. 2. Effect of various chemicals and drugs on the microsomal content of cytochrome P-450.

was observed in mice treated with AIA or griseofulvin as compared with phenobarbital or C₆Cl₆.

Calculating from our data, the maximal ability of 1 g. liver for ALA synthesis in normal mice appears to be approximately 480 m μ moles per day (20.0 m μ moles ALA $\times$ 24). As the half life of cytochrome P-450 is less than 12 hours⁵⁾, ALA needed for maintaining the normal level of cytochrome P-450 is at least 193.6 m μ moles per g. liver per day (24.2 m μ moles $\times$ 8), which is approximately 2/5 the level of normal maximum liver production. In phenobarbital treated mice at 48 hours after treatment, for example, elevated level of cytochrome P-450 4 times the normal level requires about 1,060 m μ moles ALA (193.6 $\times$ 3 + 480) for the maintenance of that level per day. As considerable amount of ALA is used for the increased content of cytochrome b₅, rate of ALA synthesis must be at least twice the normal level.

That the assayed activity of ALA synthetase is about twice the control is not only comparable, but also means that ALA formed in mitochondria is smoothly utilized for the synthesis of microsomal cytochromes. On the contrary, though AIA and griseofulvin apparently showed the stimulatory effect in the initial stage, they decreased the level of cytochromes until a considerable amount of ALA was supplied. The decreased level persists in case of DDC, resulting in the most prominent accumulation of protoporphyrin in the liver. These results suggest that porphyria-inducing chemicals might block, partially or almost completely, the pathway anywhere from heme formation to heme utilization for synthesis of microsomal cytochromes, resulting in an absolute or relative insufficiency of available hemes, and a positive feedback seems to act to supply more heme precursor through the mechanism of induction of ALA synthetase, the rate-limiting enzyme in porphyrin synthesis. It was also suggested that the amount of accumulated porphyrin would result from the difference between magnitude of ALA formation and of protoporphyrin utilization (Fig. 3).

Applying these results to the operon concept⁶⁾, heme might be the specific effector

Fig. 3. Hypothetic diagrams showing the disturbed porphyrin and heme metabolism in experimental porphyrias.

for apo-repressor of ALA synthetase. The facts that in human hepatic porphyria ALA synthetase activity was increased and that precipitators could induce acute attacks without showing any disturbance in normal human subjects might well be explained by a hypothesis of the presence of an insufficiency of hepatic microsomal cytochromes or other hemoproteins in the manifest stage due to a deficiency of available hemes compensated by increased ALA supply in a latent stage. Endogenous or exogenous toxic agents which escaped the detoxication by their livers are probably causing signs and symptoms of acute attacks in hepatic porphyria as suggested by many pathologists on the basis of autopsy findings⁷⁾, and by a successful treatment of a patient with extracorporeal hemodialysis⁸⁾.

Summary

1. Overproduction of porphyrin and its precursors due to the increased ALA synthetase activity is the common feature in human and experimental hepatic porphyric livers.

2. Increased ALA synthetase activity is induced probably through the feedback mechanism against the decreased heme content resulting from the increased synthesis of microsomal cytochromes. Lesions in the pathway through heme-pool are probably involved in the pathogenesis of porphyrias.

3. Autointoxication due to the decreased drug-metabolizing ability might cause the disorder in the nervous system.

Reference

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