SHORT COMMUNICATIONS

BBA 73048

Azotemic inhibition of organic acid transport in the liver

Evidence has accumulated that both liver and kidney possess transport mechanisms for the secretion of organic acids[1–5]. Further, SPERBER[1] has pointed out structural similarities of the compounds transported by the kidney and liver and suggested that the mechanisms in the two organs are similar.

The organic acids transferred by the excretory system in the liver are handled *via* the hippurate transport mechanism in the kidney and include phenolsulphonphthalein, indigo carmine, penicillin and hydrochlorothiazide[2]. Studies by MARTENSSON have indicated that *in vivo* net uptake of citrate occurs principally in the liver and kidney[6]. The uptake of α-ketoglutarate by the liver and kidney appears as another example of organ substrate specificity since membranes of tissues other than the liver and kidney were relatively impermeable to this substance *in vivo*[3]. SELLECK AND COHEN[3] have hypothesized that the uptake of hippurate, α-ketoglutarate and possibly citrate and free fatty acids reflect the activity of a common membrane transport mechanism present primarily in kidney and liver.

If the above observations are correct, earlier studies of hippurate uptake may serve to identify and characterize this system. In azotemia hippurate transport by the kidney was decreased to a greater degree than the glomerular filtration rate[7]. Using studies *in vitro*, it was found that incubation of isolated rabbit renal tubules with human azotemic sera markedly depressed hippurate accumulation when compared to hippurate accumulation in the presence of normal human sera[7]. This decrease in hippurate accumulation could be overcome by removal of the tubules from the azotemic sera or by dialyzing the azotemic sera prior to incubation with the tubules.

Since the organic acid transport system in the liver resembles that in the kidney, it became apparent that azotemia may also depress organic acid transport in liver.

DESPOPOULOS[2] has confirmed previous reports[8] that liver slices do not accumulate hippurate like renal slices. He suggested that the inability to demonstrate high slice to medium ratios in the liver was due to the fact that the fraction of fluid in biliary spaces is small compared to the total space occupied by excretory fluid in the large lumina of the renal tubules. In the study herein described another method was used to show hippurate accumulation by the liver.

At the start of the experiments control or azotemic rats were anesthetized with 10–15 mg sodium pentobarbital, the peritoneal cavities were entered, and the ureters were obstructed bilaterally. After closing the incision with a continuous suture secured with collodion, 5 mg of ^{131}I-labeled sodium iodohippurate (Hippuran, Abbott Laboratories) were injected intraperitoneally. 3 h after the intraperitoneal injection of hippurate, blood was drawn by cardiac puncture and the liver and kidneys rapidly removed. Liver tissue and renal cortex were weighed. The content of ^{131}I of the liver

and renal cortical tissue and of 1 ml of blood were compared by isotope counting using a well-type γ scintillation counter. The liver to blood ratio and kidney to blood ratio were calculated from the counts/min·g^{-1} tissue/counts/min·ml^{-1} blood. Blood urea concentration was determined by the method of GENTZKOW[9] and expressed as blood urea nitrogen.

Azotemia was induced in two ways. The first method was to incise the bladder to allow the urine to leak into the peritoneal cavity for 24 h prior to study. The second method was to obstruct both ureters 24 h prior to study. 24 h after either procedure the BUN range in the animals was 60–112 mg %. In the control animals, the BUN was under 20 mg %. The liver to blood and kidney to blood ratios were comparable in both azotemic states; accordingly, the results have been grouped together.

In kidney tissue, it has been shown that penicillin inhibits hippurate transport, presumably by competing in the organic acid transport mechanism[10]. Therefore, the effect of penicillin loading was also evaluated in this study. Fig. 1 illustrates the effects of penicillin loading and azotemia on hippurate uptake in the liver and kidney. When aqueous penicillin (750 mg) was given intramuscularly 1 h prior to the injection of sodium hippurate, the hippurate accumulation in the kidneys decreased from a control kidney to blood ratio of 149 $\pm$ 25 (S.E.) to 24 $\pm$ 0.8 (S.E.) ($P < 0.001$). Thus inhibition for the renal organic acid transport system was demonstrable in this experiment. This same phenomena was also seen in the liver where the average liver to blood ratio decreased from 1.51 $\pm$ 0.19 (S.E.) to 0.81 $\pm$ 0.02 (S.E.) ($P < 0.001$) in the presence of penicillin (Fig. 1). Of interest is that studies in uremic animals showed a decrease of the kidney to blood ratio to 21 $\pm$ 4.6 (S.E.) and liver to blood ratio to 0.90 $\pm$ 0.01 (S.E.) ($P < 0.001$ when compared with control). It is apparent that penicillin loading and azotemia have similar effects on renal hippurate accumulation.

Previous studies in slices have shown that neomycin feeding in azotemia decreases the azotemic inhibition of renal hippurate transport[11]. In the present investigation, azotemic rats who had received neomycin showed some ability to raise the kidney

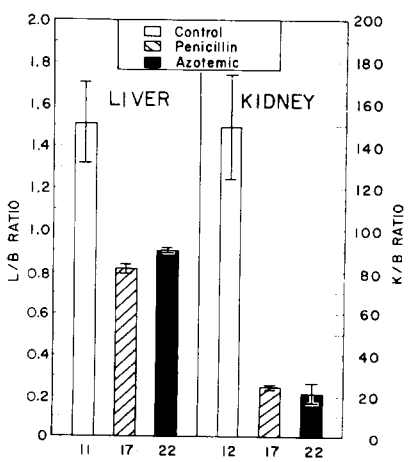

Fig. 1. Liver to blood ratios (L/B) are shown on the left and kidney to blood ratios (K/B) on the right. Three states are characterized: control, after penicillin loading (750 mg intramuscularly) and azotemia. The number of experiments performed is designated under each bar. The S.E. is indicated within each bar.

to blood ratio for hippurate, 45 ± 5.4 (S.E.) ($P < 0.001$), but no effect on the liver to blood ratio, 0.84 ± 0.01 (S.E.). Neomycin had no effect on the kidney to blood or liver to blood ratio in normal control rats.

These results are interpreted to indicate that in azotemia, organic acid accumulation in the liver as well as in the kidney is depressed. Again, this shows similarities between the organic acid transport system in both organs and raises the possibility that other compounds transported by these two systems are affected in azotemia. Since some of these substances are important in intermediary metabolic processes, impaired access of these metabolites into the hepatic cells may cause an adverse effect on hepatic metabolism during azotemia.

Supported in part by a grant from the Health Research and Services Foundation J-50, the American Heart Association Grant 67-875 and National Institutes of Health Grant AM 10417. J.R.C. and S.R.B. were supported by the U.S. Public Health Service Training Grant 1 T1 HE 5706 from the National Heart Institute.

Department of Medicine and the Renal and Electrolyte Division, University of Pittsburgh School of Medicine, Pittsburgh, Pa. (U.S.A.)

J. R. CICCONE
A. I. KELLER
S. R. BRAUN
H. V. MURDAUGH
H. G. PREUSS*

1 I. SPERBER, *Pharmacol. Rev.*, 11 (1959) 109.
2 A. DESPOPOULOS, *Am. J. Physiol.*, 210 (1966) 760.
3 B. H. SELLECK AND J. J. COHEN, *Am. J. Physiol.*, 208 (1965) 24.
4 J. J. COHEN AND E. W. RANDALL, *Am. J. Physiol.*, 206 (1964) 383.
5 T. H. WOO AND S. K. HONG, *Am. J. Physiol.*, 204 (1963) 776.
6 J. MARTENSSON, *Acta Physiol. Scand.*, 1, Suppl. II (1940) 1.
7 H. G. PREUSS, S. G. MASSRY, J. F. MAHER, M. GILLIECE AND G. E. SCHREINER, *Nephron*, 3 (1966) 265.
8 R. J. CROSS AND J. V. TAGGART, *Am. J. Physiol.*, 161 (1950) 181.
9 C. J. GENTZKOW, *J. Biol. Chem.*, 143 (1942) 531.
10 K. H. BEYER, *J. Pharmacol.*, 82 (1944) 310.
11 E. BOURKE, H. G. PREUSS, E. ROSE, M. E. WEKSLER AND G. E. SCHREINER, *Federation Proc.*, 26 (1967) 265.

Received April 12th, 1968

* Performed during tenure as an Established Investigator of the American Heart Association