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BILE ACID TRANSPORT: LESSONS FROM THE INTESTINE

Weinberg SL, Burckhardt G and Wilson FA. Taurocholate transport by rat intestinal basolateral membrane vesicles. Evidence for the presence of an anion exchange transport system. J. Clin. Invest 1986; 78:44–50.

ABSTRACT

The transport of bile acid was studied in basolateral membrane vesicles isolated from rat small intestine. Taurocholate transport into an osmotically reactive intravesicular space was Na^+ independent. The uptake of taurocholate in jejunal and ileal vesicles preloaded with sulfate was stimulated with respect to uptake in unpreloaded vesicles. Glycocholate inhibited the transstimulation of taurocholate uptake by sulfate. Sulfate and taurocholate uptake in ileal vesicles preloaded with bicarbonate was stimulated with respect to uptake in unpreloaded vesicles. Taurocholate inhibited the transstimulation of sulfate uptake by bicarbonate. When ileal vesicles were loaded with p-aminohippurate, an early transstimulation of taurocholate was found that exceeded equilibrium uptake, was insensitive to a K^+ diffusion potential, and was *cis*-inhibited by taurocholate, glycocholate, pyruvate, p-aminohippurate, probenecid, chloride, sulfate, and bicarbonate. These data indicate the presence of an anion exchanger in intestinal basolateral membrane vesicles that may be involved in the exit of bile acids from the enterocyte.

COMMENTS

Much of the exponential increase in our understanding of epithelial transport phenomena over the last decade has been the result of studies utilizing plasma membrane vesicles. This experimental model offers distinct advantages over whole cell or tissue preparations, which are hampered by the effects of metabolic degradation and intracellular organelles, and by the contribution of paracellular pathways to overall solute movement. Perhaps more important, in polar cells such as the hepatocyte, where a clear division exists with respect to structure and function between basolateral (sinusoidal) and apical (canalicular) domains, membrane vesicle preparations represent an ideal system in which events occurring at the respective membrane surfaces can be isolated and characterized. The recent development of several methods to prepare purified basolateral and canalicular liver plasma membrane vesicles (1) has enabled hepatologists to keep apace of their intestinal and renal colleagues. A review of the report of Weinberg et al. may serve to illustrate some of the similarities as well as the differences between bile acid transport in the hepatocyte and other epithelia.

Taurocholate, as a representative conjugated bile acid, is transported across both the ileal brush border membrane (2) and the hepatocyte sinusoidal membrane surface (3) by a secondary active Na^+-dependent transport process. The inwardly directed Na^+ gradient that provides the driving force for the "uphill," intracellular accumulation of the bile acid against its electrochemical gradient arises from the active extrusion of Na^+, mediated by basolateral Na^+,K^+-ATPase. This bile acid transport protein has been putatively identified on the sinusoidal membrane surface of the hepatocyte using photoaffinity labels (4).

Much like the situation in the enterocyte, our understanding of events involved in the exit of bile acids across the contralateral membrane surface, generally regarded as the rate-limiting step in overall hepatic transport, has been comparatively limited. However, recent studies have confirmed that a Na^+-independent, potential-sensitive taurocholate transport system is present at the canaliculus (5), and this bile acid carrier has also been tentatively identified (6).

This report identifies and characterizes transport of bile acids across the basolateral membrane of the rat enterocyte. Initial experiments confirmed that the basolateral membrane vesicles were osmotically sensitive, did not metabolically degrade the taurocholate standard and exhibited the high degree of taurocholate binding that has been previously observed in other membrane vesicle preparations (5). No preferential increase in taurocholate uptake into vesicles was observed with an inwardly directed Na^+ gradient when compared to a similar K^+ gradient, indicating the absence of sodium dependency while indirectly confirming the purity of the membrane vesicle preparation. Sulfate/hydroxyl exchange was first established in jejunal and ileal basolateral membrane vesicles and then sulfate/taurocholate exchange was demonstrated. Bicarbonate could substitute for hydroxyl ions in the former [in contrast to a previous report (7)] and sulfate ions in the latter. Furthermore, the organic anion, p-aminohippurate, could participate in taurocholate exchange. This organic anion exchange was inhibited by a series of representative inorganic and organic anions and appeared to be electroneutral (the uptake of taurocholate in the presence of an outwardly directed p-aminohippurate gradient was not further stimulated by the imposition of a transient intravesicular *positive*, not negative as stated in the report, K^+ diffusion potential).

Caution is advised before extrapolating these findings beyond the enterocyte to the hepatocyte. Whereas sulfate uptake at the basolateral membrane of rat hepatocytes may be mediated by a similar anion exchange mechanism (8) and hydroxyl/cholate exchange has been recently described in basolateral liver plasma membrane vesicles

(9), no evidence was found recently for either sulfate:bile acid exchange (10) or bicarbonate:bile acid exchange (11) at the canalicular membrane surface.

Thus, at present, the movement of bile acid anions out of the hepatocyte appears to be the result of carrier-mediated, facilitated diffusion, favored by the negative intracellular electrical potential. However, based on the Nernst equation, this cannot be the sole process by which bile acids are concentrated into bile (5), and the contribution of exocytosis of vesicle-associated bile acids derived from the endoplasmic reticulum and Golgi apparatus and/or other carrier-mediated transport mechanisms remain to be determined. Perhaps additional studies, currently underway, using both liver plasma membrane vesicles and isolated hepatocyte couplets (12), with a defined canalicular space ideally suited for micropuncture, will clarify hepatic bile acid transport to the same degree as Weinberg and colleagues have for the intestinal transport of these important organic anions.

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DATING GALLSTONES

Mok HYI, Druffel ERM and Rampone WM. Chronology of cholelithiasis. Dating gallstones from atmospheric radiocarbon produced by nuclear bomb explosions. N. Engl. J. Med. 1986; 314:1075–1077.

ABSTRACT

We investigated the natural history of cholelithiasis in 59 samples of stones from the gallbladder or common bile duct in 15 patients, using as a tracer for the timing of stone formation the ^{14}C released into the environment during nuclear weapons testing. The ages of the stones were correlated with the dates of onset of symptoms and with other clinical data.

None of 11 symptomatic patients had symptoms or complications until at least two years (mean ± SD, 8.0±5.1 years) after stone formation began. There was a lag time of 11.7±4.6 years between initial stone formation and cholecystectomy. The growth rates of stones from 11 symptomatic patients and 4 asymptomatic patients were similar (2.6±1.4 and 2.6±1.1 mm per year). Studies of two stones retrieved from the common bile duct showed that one had the same age as a cholecystic stone; the other, removed two years after cholecystectomy, apparently grew in the common bile duct.

The long latency period between the formation of gallstones and the onset of symptoms indicates that interruption of the natural progression of gallstone disease is potentially possible with medical therapy.

COMMENTS

How fast do gallstones grow? Are symptoms related to growth? Such questions are not answered because it has been impractical to measure gallstone growth rates and growth patterns. The scant data that we have come from unusual sources such as the placebo group of the National Cooperative Gallstone Study in which oral cholecystography was repeated serially (1), and from unusual case series (2–4).

Knowledge about gallstone growth is potentially important for understanding some clinical problems. We are still somewhat in the dark about *how* gallstones cause biliary symptoms. Biliary colic probably occurs when a stone becomes caught in the neck of the cystic duct. Perhaps features of bile acid composition and of gallstone growth are related to the development of symptoms. Clinically, it is interesting to recall that Lund suggested in his follow-up study that if biliary symptoms develop they do so soon after the discovery of gallstones (5). Do symptoms tend to occur "sooner" rather than "later"? If we could measure and describe gallstone growth, we might be able to tell. A positive answer would have some clinical use and would raise some fascinating questions for physiologists and physical chemists.