Hepatic Collagen Metabolism: Effect of Alcohol Consumption in Rats and Baboons

Abstract. *Long-term ethanol feeding causes collagen accumulation in livers of rats and baboons. Activity of collagen proline hydroxylase in the liver is also stimulated, and incorporation of proline into collagen hydroxyproline in rat liver slices is significantly enhanced, a result indicating that increased synthesis is responsible, in part, for the collagen accumulation.*

Cirrhosis is linked epidemiologically to chronic, heavy consumption of alcohol (*1*). Fibrous tissue is characteristically more abundant in the liver when cirrhosis is present (*2*). Since collagen is a chief constituent of fibrous tissue (*3*) it is important in understanding alcohol's role in the etiology of cirrhosis to know whether chronic alcohol consumption has a direct effect on hepatic collagen metabolism.

To learn if prolonged consumption of ethanol by itself leads to an accumulation of hepatic collagen we pair-fed 16 baboons an adequate diet containing 20 percent of calories as protein, 14 percent as fat, 66 percent as carbohydrate, and sufficient amounts of vitamins and minerals. The control animal of each pair received the full carbohydrate allowance while the other consumed ethanol (up to 36 percent of total calories) isocalorically substituted for carbohydrate. After animals were kept on these diets an average of 7 months, liver biopsies were analyzed for hydroxyproline content in protein as a measure of hepatic collagen (*3*, *4*). Livers of animals fed alcohol contained 204 percent as much collagen as did livers of controls ($P < .01$).

Similar studies were then done on rats, which are more suitable for studying the mechanism involved. Rat littermates were pair-fed nutritionally adequate liquid diets; the control animals received 18 percent of calories as protein, 35 percent as lipid, and 47 percent as carbohydrate, and the animals fed alcohol received the identical diet except that ethanol (36 percent of total calories) isocalorically replaced carbohydrate (*5*). After 7 months on these diets, the animals fed alcohol had 72 percent more hydroxyproline in hepatic protein than did pair-fed controls (435 compared to 258 μg per gram of liver, eight pairs, $P < .001$). After 14 months of alcohol consumption the increase was 107 percent (544 compared to 267 μg per gram of liver, nine pairs, $P < .001$).

In previous studies when similar diets were fed to animals and humans, fatty liver and alterations in hepatocellular organelles developed, but no fibrosis or increase in collagen was detected by light or electron microscopy (*6*). Histological techniques, however, are not as sensitive as the biochemical one we used here to measure collagen and to detect its early accumulation.

To determine whether hepatic synthesis of collagen is enhanced in rats fed alcohol, we incubated liver slices from pair-fed animals with [^{14}C]proline in a system modified from that of Huberman *et al.* (*7*) and measured the incorporation of radioactivity into liver collagen hydroxyproline (*7*, *8*). [^{14}C]-Proline incorporation into collagen [^{14}C]-hydroxyproline was 50 percent greater in animals fed alcohol than in controls after 7 months on the diets (seven pairs, $P < .05$) and 184 percent greater after 14 months on the diets (five pairs, $P < .05$).

The mechanism of increased collagen formation was studied. We assayed the livers for the activity of collagen proline hydroxylase (*9*). This enzyme is necessary for collagen synthesis, and a rise in its activity is an early indication of hepatic fibrosis in hepatic injury induced in laboratory animals (*10*). Activity of collagen proline hydroxylase was 40 percent greater in the livers of baboons fed alcohol than in those of control animals after 7 months on the diets (11 pairs, $P < .05$). Enzyme activity was also significantly greater in the livers of rats fed alcohol for 1 month than in those of control rats, and the difference increased after 7 and 14 months on the diets (Fig. 1).

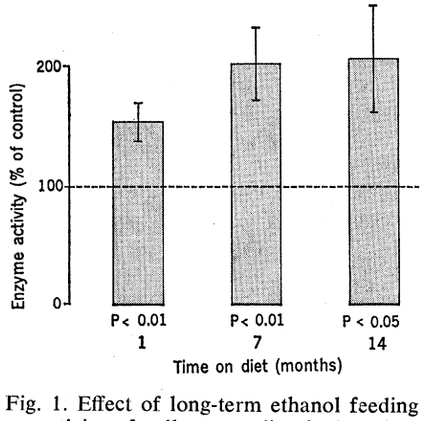

Fig. 1. Effect of long-term ethanol feeding on activity of collagen proline hydroxylase in rat liver. Rats were pair-fed nutritionally adequate diets containing alcohol or dexin. Enzyme activity of control animals is indicated by the broken line.

A possible link between ethanol and collagen metabolism is the increased concentration of lactate in the liver and blood; this increase is favored by the reduced diphosphopyridine nucleotide generated when ethanol is oxidized in the liver (*11*). Elevated lactate concentrations are associated with enhanced activity of collagen proline hydroxylase in vitro (*12*) and in vivo (*13*). In any event the increased synthesis and accumulation of hepatic collagen promoted by ethanol consumption may contribute, along with other factors of hepatic injury, to the initial steps of a complex sequence of events leading to cirrhosis.

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References and Notes

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