PLASMA ARYLESTERASE ACTIVITIES IN INBRED STRAINS OF RABBITS, RATS AND MICE FED LOW- AND HIGH-CHOLESTEROL DIETS

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ABSTRACT

In inbred strains of rabbits and rats it was found that differences in the responsiveness of plasma cholesterol level to dietary cholesterol was often associated with genetically determined differences in plasma esterase pattern. A fast moving zone was present in the plasma zymogram of hyporesponsive strains but not in that of hyperresponsive strains. We have now measured plasma total esterase activities in solution in hypo- and hyperresponsive inbred strains of rabbits and rats on low- and high-cholesterol diets using β-naphthylpropionate as a substrate. Basal plasma esterase activities were significantly higher in a rabbit strain and in a rat strain hyporesponsive to dietary cholesterol than in the corresponding hyperresponsive strains. In all four strains, plasma esterase activity was significantly elevated by cholesterol feeding, but the activities remained higher in the hypo- than in the hyperresponsive strains. In a study with 7 inbred strains of mice we found no association of plasma esterase profile after electrophoresis or plasma total esterase activity with sensitivity to a high-cholesterol diet. In all strains plasma total esterase activity was increased after cholesterol feeding. It appears that arylesterases are associated with cholesterol metabolism and with the response to dietary cholesterol in the selected strains of rabbits and rats, but evidence for such a role in the inbred strains of mice is inconclusive. As plasma contains many different esterases, it would be desirable to study and measure the esterases separately. Such studies are now in progress.

INTRODUCTION

The plasma of vertebrate animals contains enzymes that can hydrolyse artificial fatty acid esters of aromatic alcohols. They are therefore called arylesterases. The enzymes differ in pH optimum and substrate specificity, including the chain length of the esterified fatty acid. Although the physiological function of plasma arylesterases is obscure, there is some evidence that they are involved in cholesterol metabolism. The presence of an isoenzyme of high mobility on starch-gel electrophoresis is often associated with a diminished response of serum cholesterol to a high-cholesterol diet. The cholesterolemic response was low in 6 out of 7 inbred rat strains displaying the zone with high mobility (which in rats is called Es-1) whereas absence of the enzyme was associated with the development of high degrees of hypercholesterolemia after cholesterol feeding in 2 out of 3 inbred strains (1). Similar results were obtained in six inbred strains of rabbits (2). Hyporesponsive rabbit strains displayed a high-mobility band on electrophoresis (called Est-2 here), but the hyperresponders did not. The Est-2 genetic

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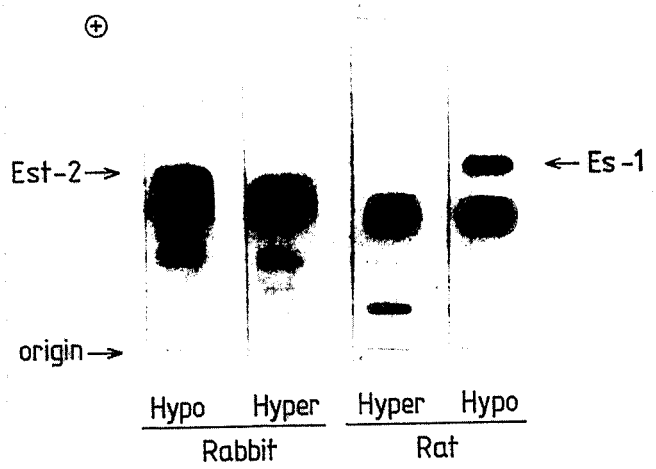

Fig. 1. Electrophoresis on starch gel of arylesterases in plasma from inbred strains of rats and rabbits on a low-cholesterol, commercial diet. β-Naphthyl-propionate was used as substrate for visualizing enzyme activity.

locus of the rabbit is assumed to be homologous with the Es-1 locus in the rat (3). Fig. 1 shows the zymogram of esterases in plasma from hypo- and hyperresponsive rabbit and rat strains.

The electrophoretic esterase pattern gives only qualitative information. We have therefore measured quantitative plasma esterase activities on low- and high-cholesterol diets in inbred strains of rabbits, rats and mice.

STUDY WITH RABBITS

The qualitative difference in esterase pattern between the hypo- and hyperresponsive inbred rabbits (Fig. 1) corresponds with a quantitative difference in the plasma total esterase activity. The baseline plasma esterase activity was significantly higher in the inbred rabbit strain which is hyporesponsive to dietary cholesterol than in the hyperresponsive strain (Fig. 2). Cholesterol feeding increased plasma total esterase activities in both strains, but the activity in the hyporesponders remained higher than in the hyperresponsive rabbits.

STUDY WITH RATS

Male rats of a hyper- and of a hyporesponsive inbred strain were fed either a low-cholesterol commercial diet or the same commercial diet to which 2% (w/w) cholesterol, 0.5% cholate and 5% olive oil had been added. Fig. 3 shows that, just as in rabbits, plasma esterase activity on the low-cholesterol control diet was significantly higher in the hyporesponsive strain. Feeding the high-cholesterol diet again markedly increased plasma esterase activities in both strains, the activity still being higher in the hyporesponder rats than in the hyperresponders. One interpretation of these data is that a low esterase activity causes an increased susceptibility to dietary cholesterol,

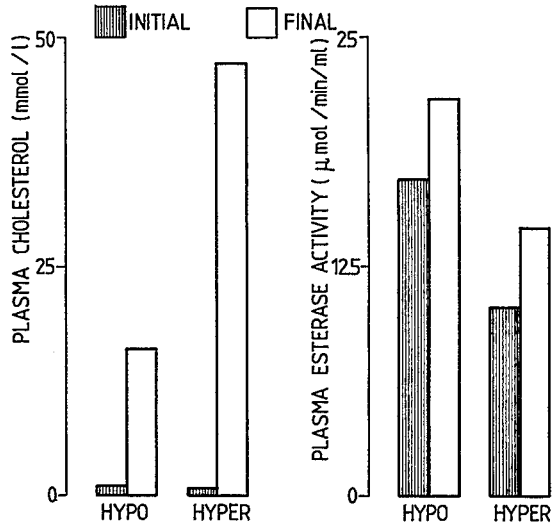

Fig. 2. Plasma cholesterol concentrations and plasma total esterase activities in male rabbits of two inbred strains (IIIVO/Ju and AX/Ju). The animals were sampled before and 28 days after the addition of 0.5% cholesterol (w/w) to the diet. β-Naphthylpropionate was used as substrate to determine esterase activity. Based on ref. 4.

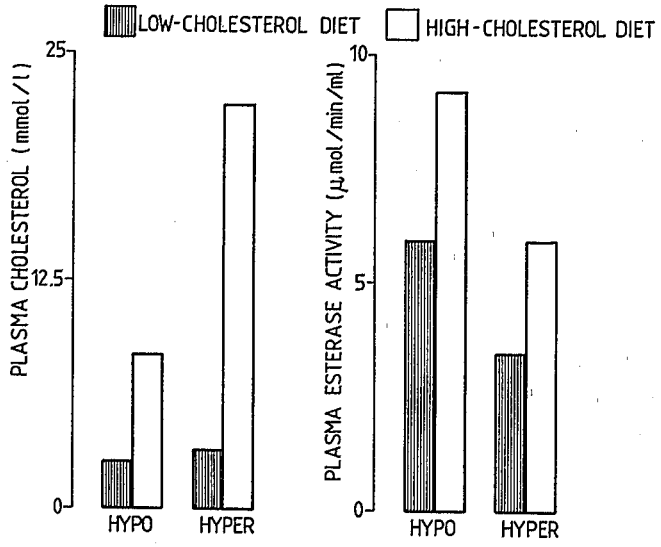

Fig. 3. Plasma cholesterol concentrations and plasma total esterase activities in male rats of two inbred strains (SHR/CpB and SD/CpB). Animals of each strain were either fed a low- or high-cholesterol diet for 23 days. β-Naphthylpropionate was used as substrate to determine esterase activity. Based on ref. 5.

whereas induction of plasma esterase activity is required to compensate for cholesterol loading. Information on the physiological function of aryl-esterases will be required before we can either substantiate or dismiss these speculations.

STUDY WITH MICE

In an attempt to further substantiate our observations we performed a similar experiment with male mice of 7 inbred strains. The animals were either fed a low-cholesterol commercial diet or the commercial diet to which cholesterol (2%), cholate (0.5%) and olive oil (5%) were added. In these strains there was no clear association between the plasma esterase pattern after gel electrophoresis and the response of plasma cholesterol to the high-cholesterol diet. Likewise, there was no relation between plasma total esterase activity on the low-cholesterol diet and the plasma cholesterol response to the high--cholesterol diet. However, in all strains plasma total esterase activity was increased upon cholesterol feeding. Fig. 4 shows the plasma cholesterol levels and esterase activities in the most strongly hypo- and hyperresponsive strains.

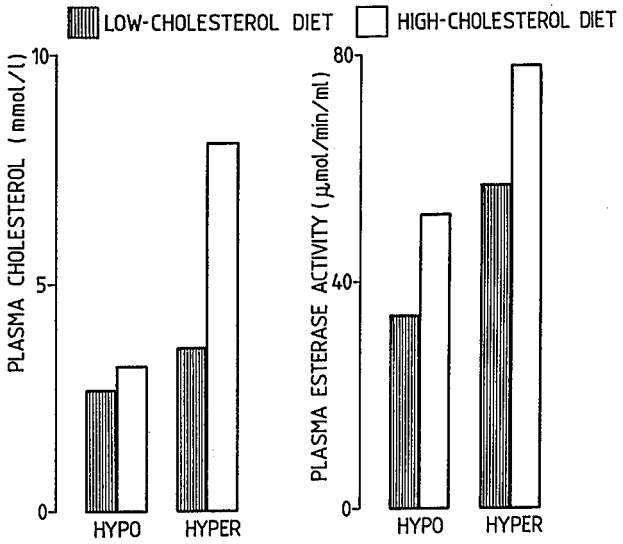

Fig. 4. Plasma cholesterol concentrations and plasma total esterase activities in male mice of two inbred strains (C57BL/U and FT/U). Animals of each strain were either fed a low- of high-cholesterol diet for 28 days. β-Naphthylpropionate was used as substrate to determine esterase activity.

DISCUSSION

We have found that total basal arylesterase activity in plasma was significantly higher in inbred strains of rabbits and rats which are hyporesponsive to dietary cholesterol than in strains which are hyperresponsive. This difference in plasma quantitative esterase activity agrees with the difference

in isoenzyme pattern between the hypo- and hyperresponsive strains. In the study with inbred mice, however, no evidence for an association between plasma esterases and sensitivity to high cholesterol intakes could be obtained. It should be realized that plasma of the laboratory animals used contains at least 10 different arylesterases, most of them probably not related to cholesterol metabolism. It is therefore desirable to study and measure the various plasma esterases separately.

Loading the animals with dietary cholesterol invariably caused an increase in plasma total esterase activity. It is possible that the increase in esterase activity results from a release of esterases from the intestine induced by dietary cholesterol. In rats, Lewis and Hunter (6) found that injection of fat into the stomach caused a marked increase in the activity of esterases of high electrophoretic mobility in the intestinal lymph, and later also in the serum. The possible role of these esterases in cholesterol absorption remains to be elucidated. Alternatively, the increase in plasma esterase activity after cholesterol feeding may be an artefact due to cell damage in the liver. More work is necessary to reveal the functions of plasma arylesterases. The use of inbred strains of animals with defined, but different plasma esterase patterns, may be of great importance in this respect.

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