4) Age-Related Change in the Cholesterol 7 α -Hydroxylase Activity in SHRSP.
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The key step in the catabolism of cholesterol to bile acids in the liver is the induction of a hydroxy group in 7 α -position of cholesterol. The enzyme catalyzing this reaction is the microsomal cholesterol 7 α -hydroxylase (7 α -hydroxylase, EC 1.14.13.17). In our previous paper (JPN HEART J 31: 577, 1990), we reported the diurnal variation of 7 α -hydroxylase activity and demonstrated that the activity in SHRSP is significantly lower than that in WKY at 24 weeks of age. In the present work, to investigate the effect of aging on the 7 α -hydroxylase activity and to confirm the difference of the activity between SHRSP and WKY, we have studied age-related change in the 7 α -hydroxylase activity in SHRSP, using Kyo:Wistar rats (WKY) as control.

MATERIALS AND METHODS: Male SHRSP and the age-matched male WKY at various ages until 12 months of age were used. The rats were maintained on a normal stock diet (CE-2, Clea Japan Inc.) with free access to the diet and drinking water (tap water). The animals were killed at 9-11 AM by collecting whole blood from the abdominal aorta after fasting overnight. Their livers were perfused with saline and removed immediately. The hepatic microsomal fraction was obtained by the regular ultracentrifugation procedure. The microsomal fraction was suspended in 0.1M phosphate buffer (pH 7.4) and used for the assay of 7 α -hydroxylase. The microsomal content of protein was determined by a dye-binding assay (Bio-Rad). The enzyme assay was carried out according to the method of Ogishima et al. and slightly modified by us (JPN HEART J 31: 577, 1990). The activity was expressed in terms of pmole of 7 α -hydroxy cholesterol formed per mg microsomal protein per min (pmole/mg protein/min). The total lipids of the liver were extracted by the method of Folch et al. and the contents of cholesterol and phospholipid were measured by commercially available kits.

RESULTS AND DISCUSSION: The liver wet-weight (g/100g B.W.) of SHRSP almost remained no change with age, while that of WKY decreased with age until around 4 months of age, and then remained almost constant. Accordingly, although there was no significant difference in the liver wet-weight between SHRSP and WKY at the young age (until around 2 months of age), the weight of SHRSP became significantly higher than that of WKY from around 4 months of age.

The liver phospholipid contents in both strains were gradually increased with age, and no significant difference was observed between SHRSP and WKY during the experimental period. The liver cholesterol content of WKY markedly increased at around 4 months of age, and then it decreased to the initial level. On the other hand, that of SHRSP gradually decreased with age, but at around 8 months of age it markedly decreased, then returned to the initial level. Consequently, at 4 and 8 months of age among examined ages, the liver cholesterol contents of WKY were significantly higher than those of SHRSP.

The hepatic microsomal 7 α -hydroxylase activity of WKY gradually increased with age from 1 month of age (15.7 ± 2.8) until 12 months of age (24.7 ± 2.5). The 7 α -hydroxylase activity of SHRSP markedly increased at 2 months of age (15.3 ± 1.2). Thereafter the activity returned to the level of 1 month of age (6.2 ± 0.7), then it gradually increased with age until 12 months of age (12.2 ± 1.9). The 7 α -hydroxylase activity of SHRSP was significantly lower than that of WKY throughout the experimental period until 12 months of age. The age-related change in the 7 α -hydroxylase activity showed no similarity to those in serum lipids and apolipoproteins reported previously.

These results indicate that the lower 7 α -hydroxylase activity in SHRSP may be genetically determined and could cause the disorder in lipid metabolism of SHRSP, in other words, the lower 7 α -hydroxylase activity could easily induce the development of hypercholesterolemia and fatty liver in SHRSP fed a high-fat and high-cholesterol diet.