Plasma triglycerides and LCAT activity of mice fed on various diets

Dietary groups	Sex	Triglyceride levels mg/100 ml (mean ± SEM)	p-value**	LCAT activity* µmoles cholesterol/L/h
Pellet	M	60.0 ± 2.5	–	97.7
control	F	57.5 ± 2.0	–	51.8
Basal	M	75.0 ± 2.9	< 0.01	130.9
	F	68.9 ± 3.3	< 0.02	102.1
High	M	128.0 ± 4.5***	< 0.001	154.2
sucrose	F	110.0 ± 5.0	< 0.001	117.3

*Assayed from pooled plasma; each value was mean of 4 readings; **When compared with control; ***Male vs female, $p < 0.05$.

animals were sacrificed under light ether anaesthesia and blood was collected in ice-cold heparinized tubes by cardiac puncture. Plasma was separated and used for various investigations. Cholesterol was estimated by colorimetry[8], and LCAT was assayed essentially by the method of Stokke and Norum[9]. The enzyme was also assayed by the method of Glomset[10]. Though the results of this method were not included in this paper. The enzyme activity in mouse was almost 2 fold to that in humans, and therefore the enzyme concentration and the incubation period were reduced by one-half of that in the original method. Triglycerides were estimated by the method of Van Handel and Zilversmit[11].

Results and discussion. The data presented in the table clearly demonstrate increased triglyceride levels in the basal and high sucrose-fed animals when compared with those fed on control diet. Among the 2 experimental dietary groups, high sucrose-fed animals had higher levels. There was a marked sex difference in plasma triglyceride levels of high sucrose-fed animals. LCAT activity was found to be increased in both basal as well as high sucrose group but more so in case of latter. Males had higher enzyme activity in plasma than female in all the animals including controls. There appeared some association between increase in plasma triglycerides and LCAT activity during this study. Our observations supported the earlier findings[4,5]. The results in the present study were unlike those observed by Goren and Simons[6] because they found a decrease in LCAT activity (when assayed by the method of Stokke and Norum[9]) in hypertriglyceridemic patients, whereas LCAT activity increased (when assayed according to the method of Glomset[10]) in the same patients. We also used both methods and in both cases the activity followed the same pattern. These experiments, therefore, further confirm the view that LCAT activity is influenced in part by the triglyceride concentration of the plasma.

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Dopamine-β-hydroxylase activity in stroke-prone spontaneously hypertensive rats

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Summary. Dopamine-β-hydroxylase (DBH) activity was higher in the serum, the mesenteric artery and the cerebral cortex of 4-week-old stroke-prone spontaneously hypertensive rats (SHRSP), and lower in the nucleus tractus solitarii than it was in spontaneously hypertensive rats (SHR).

Dopamine-β-hydroxylase (DBH) activity was found to be higher in the serum, the mesenteric vessels, of 3-week-old spontaneously hypertensive rats (SHR)[2,3] and lower in the locus coeruleus than it was in the control Wistar-Kyoto rats[4,5]. At 16 weeks of age, when hypertension of SHR was fixed, DBH activity in the serum, the mesenteric vessels, and the locus coeruleus, there were no significant differences between SHR and normotensive Wistar-Kyoto rats. In contrast, DBH and tyrosine hydroxylase (TH) activities in the adrenal glands and the vas deferens were significantly higher in SHR than in Wistar-Kyoto rats[6,7]. These changes suggest that the nervous system is an important regulator of blood pressure, especially in an early phase in the development of hypertension of SHR. Stroke-prone spontaneously hypertensive rats (SHRSP)[8] were isolated as a mutant of SHR by

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Okamoto et al. The characteristics of SHRSP are a very high incidence of cerebrovascular diseases and a very rapid increase in blood pressure. We speculate that the changes of catecholamine-synthesizing enzymes observed in SHR as compared with those in normotensive Wistar-Kyoto rats might be more pronounced in SHRSP. We have therefore compared DBH activity in SHRSP with that in SHR.

Materials and methods. The SHR and SHRSP examined were 3, 4 and 16 weeks of age. They were raised under the same conditions. The blood pressures of SHR and SHRSP at 16 weeks of age were 180 ± 9 ($n = 5$) and 209 ± 9 mmHg (mean ± SEM), respectively ($p < 0.05$). The rats were decapitated, and the mesenteric arteries, the hearts, the adrenal glands, and the brains were quickly removed, weighed, frozen on dry ice and stored at −80 °C. Blood samples were obtained by decapitation and exsanguination into a test-tube kept on ice. Serum was removed after centrifuging at 10,000 × g for 10 min. The brain regions were dissected under a microscope from frozen sections of the brain[9]. DBH activity was determined based on the formation of octopamine from tyramine by dualwavelength spectrophotometry[10], as described previously[6]. TH activity was assayed by measuring the [^{14}C]dopa formed from L-[U-^{14}C]tyrosine[11,12], as described previously[6]. 6-Methyltetrahydropterin was used as a cofactor.

Table 1. Dopamine-β-hydroxylase (DBH) activity of spontaneously hypertensive rats (SHR) and stroke-prone spontaneously hypertensive rats (SHRSP)

DBH activity[a]	Age (weeks)	SHR	SHRSP
Brain regions (pmole/min mg protein)			
Locus coeruleus	4	119±9 (5)	129±3 (4)
Nucleus tractus solitarii	4	84±6 (4)	63±3[b] (4)
Hypothalamus	4	78±4 (5)	77±3 (5)
Cerebral cortex	4	27±2 (5)	33±1[b] (5)
Peripheral tissues (nmole/min g tissue)			
Serum	4	1.22±0.10 (5)	1.54±0.05[b] (4)
Mesenteric artery	4	6.75±0.64 (5)	10.59±0.70[b] (5)
Heart	4	1.44±0.06 (5)	1.29±0.06 (5)
Adrenal glands	4	56.2 ±5.6 (5)	46.2 ±5.7 (5)
Serum	16	0.21±0.02 (5)	0.24±0.03 (4)
Heart	16	1.31±0.09 (5)	1.01±0.05 (5)
Adrenal glands	16	86.0 ±6.8 (5)	102±16 (5)

[a]Values are mean ± SEM; [b]differs from control (SHR), $p < 0.05$.

Table 2. Tyrosine hydroxylase (TH) activity in adrenal glands of spontaneously hypertensive rats (SHR) and stroke-prone spontaneously hypertensive rats (SHRSP)

TH activity[a] (nmole/min g tissue)	SHR	SHRSP
3 weeks of age	37.8 ± 1.9 (6)	32.5 ± 1.7 (6)
16 weeks of age	72.7 ± 7.6 (5)	121.1 ± 18.8[b] (5)

[a]Values are mean ± SEM; [b]differs from control (SHR), $p < 0.05$.

Results and discussion. DBH activities in various brain regions, the serum, the mesenteric arteries, the hearts, and the adrenal glands of SHR and SHRSP at 4 weeks and 16 weeks of age are shown in table 1. DBH activity was higher in the serum and the mesenteric arteries of 4-week-old SHRSP than it was in SHR. These results are similar to the changes between Wistar-Kyoto rats and SHR and support our hypothesis that SHRSP have a higher sympathetic activity especially in blood vessels than SHR, and that serum DBH may be mainly derived from the sympathetic nerve terminals in the blood vessels. Nagaoka and Lovenberg[13] also reported a significantly higher serum DBH activity in SHRSP at 4 weeks of age as compared with that in SHR.

DBH activity in the brain was lower in the nucleus tractus solitarii and higher in the cerebral cortex of 4-week-old SHRSP than it was in SHR. DBH activity in the locus coeruleus of young SHRSP was not significantly different from that of SHR. In our previous report[5], SHR at 3 weeks of age had a lower DBH activity in the locus coeruleus than control Wistar-Kyoto rats, but we did not measure the activity in the nucleus tractus solitarii or cerebral cortex. Since ablation of the nucleus tractus solitarii and its tractus or transection of adjacent areas of the nucleus tractus solitarii was found to cause acute hypertension, probably by removing an inhibitory central control[14,15], low DBH activity in the nucleus tractus solitarii of SHRSP may have some relation with severe hypertension in SHRSP.

DBH activities of SHRSP in the serum, the heart, and the adrenal glands at 16 weeks of age after the establishment of hypertension did not differ significantly from those of SHR. Since we had found that TH activity in the adrenal glands was significantly higher in SHR than in Wistar-Kyoto rats at 14–16 weeks of age[6,7], TH activity in the adrenal glands was assayed in SHRSP and SHR at 3-weeks and 16-weeks of age. As shown in table 2, TH activity was significantly higher only in 16-week-old SHRSP than it was in SHR. These changes in adrenal TH between SHRSP and SHR are similar to those between SHR and normotensive Wistar-Kyoto rats. Higher DBH activity in the serum, the mesenteric artery and the cerebral cortex and lower DBH activity in the nucleus tractus solitarii at 4 weeks of age and higher TH activity in the adrenal glands at 16 weeks of age in SHRSP, as compared with SHR, suggest that the abnormality in the sympathetic adrenergic system may be more pronounced in SHRSP than in SHR.

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