21) Differences between SHR and WKY in Content of Cytochrome P-450 Isozymes Which Contribute Arachidonic Acid Metabolism. Susumu Imaoka, Kazuo Takaori,* Tokihito Yukimura,* Kenjiro Yamamoto,* and Yoshihiko Funae. Laboratory of Chemistry and *Department of Pharmacology, Osaka City University Medical School, Osaka 545.

Renal microsomes readily metabolize arachidonic acid, prostaglandin, and fatty acids. Cytochrome P-450 contributes metabolism of these substrate. Recently, Sacerdoti *et al.* reported that spontaneously hypertensive rats (SHR) produce arachidonic acid metabolites more abundantly than normotensive control rats (Wistar-Kyoto rats, WKY). The renal cytochrome P-450s metabolize arachidonic acid to ω - and (ω -1)-hydroxyarachidonic acid and epoxyeicosatrienoic acids. These metabolites have potent biological effects such as vasoconstriction. Recently, we identified certain cytochrome P-450s that produce epoxyeicosatrienoic acid and ω -, and (ω -1)-hydroxyarachidonic acids in rat renal microsomes. Here, we investigated the differences between SHR and WKY in the amount of these cytochrome P-450s in renal microsomes.

Male SHR and age-matched WKY (12 weeks of age) were used in this study. Systolic blood pressure was measured by the tail cuff method. The mean blood pressures of WKY and SHR were 140 ± 3.6 and 185 ± 6.0 mm Hg, respectively. The renal microsomes were prepared from homogenates of rat kidneys by use of different centrifugation. The amounts of cytochrome P-450 isozymes in renal microsomes were measured by Western blotting with antibody.

The content of total cytochrome P-450, NADPH-cytochrome P-450 reductase, and cytochrome b_5 assayed photometrically were not different in WKY and SHR, but lauric acid ω - and (ω -1)-hydroxylation activities of SHR were two times those of WKY. These results suggested that contents of certain forms of cytochrome P-450 in renal microsomes are different in SHR and WKY. We have purified three different forms of cytochrome P-450, P450 K-2, K-4, and K-5. P450 K-2 metabolized lauric acid at the (ω -1)-position and produced epoxyeicosatrienoic acid from arachidonic acid. P450 K-4 metabolized prostaglandin A_1 , lauric acid, and arachidonic acid at the ω - and (ω -1)-positions. P450 K-5 hydroxylated lauric acid and arachidonic acid at the ω - and (ω -1)-positions. The contents of P450 K-2 and K-4 assayed by Western blotting in SHR were not different from those in WKY. However, the content of P450 K-5 in SHR was significantly higher than that in WKY. Also, P450 K-5 was a major cytochrome P-450 in renal microsomes of WKY and SHR. These results explain the difference of lauric acid hydroxylation activity in SHR and WKY.

P450 K-5 seems to contribute mainly to arachidonic acid ω -hydroxylation in renal microsomes because P450 K-5 was a major cytochrome P-450 in rat renal microsomes and had high arachidonic acid ω -hydroxylation activity. Schwartzman *et al.* reported that ω -hydroxyarachidonic acid is a potent vasoconstrictor and depletion of renal cytochrome P-450 in SHR prevents hypertension. Our findings and these results suggest that renal cytochrome P-450 contributes to the regulation of blood pressure and renal function.