Antibody-sensitive renin of adrenal and resistance vessels is markedly elevated in spontaneously hypertensive rats

MITSUHIDE NARUSE AND TADASHI INAGAMI
Department of Biochemistry and Hypertension Center, Vanderbilt University, Nashville, TN, U.S.A.

Summary
1. Three-week-old spontaneously hypertensive rats (SHR) showed markedly elevated specific renin activity in the adrenal and the mesenteric vessels compared with control Wistar-Kyoto (WKY) rats.
2. In 17-week-old SHR with established hypertension, adrenal renin remained markedly elevated over that of WKY rats, whereas no significant difference was observed in renin of the mesenteric vessels.
3. The specific renin activity of the aorta showed no significant difference compared with that of WKY rats at 3 or 17 weeks of age.
4. These results suggest possible involvement of adrenal and vascular renin in the pathogenesis of hypertension in SHR.

Key words: adrenal gland, mesenteric vessels, renin, spontaneously hypertensive rats.

Introduction
The etiology of hypertension in SHR has been of great interest in view of its resemblance to essential hypertension in humans. Although several factors have been implicated in its pathogenesis, no conclusive explanation has yet been obtained. Normal or subnormal levels of plasma renin [1, 2] and the failure to normalize blood pressure by nephrectomy [3] or by angiotensin II antagonists [4] indicate that the renin–angiotensin system may not play a role in spontaneous hypertension. However, recent findings that the angiotensin converting enzyme inhibitor, captopril, normalizes the blood pressure of patients with essential hypertension as well as SHR [4, 5] suggest a possible role for the renin–angiotensin system in extrarenal tissues. Vascular renin has been implicated in some hypertension models [6] and we have recently demonstrated a high renin level in the rat adrenal gland [7]. In the present study, using antirenin antibody, we compared the renin activity of the adrenals, mesenteric vessels and aorta in both young and adult SHR and WKY rats.

Methods
Age-matched male SHR and WKY rats, kept under identical conditions with access ad libitum to regular chow and tap water, were killed by exsanguination under pentobarbital anesthesia 24 h (3-week-old rats) or 36 h (17-week-old rats) after bilateral nephrectomy and perfused with sodium chloride solution (154 mmol/l: saline). Tissues were quickly removed, frozen on solid carbon dioxide, and stored at -80°C. Renin was extracted by a published method [7]. Mesenteric vessels and aorta were frozen and thawed five times before extraction. The renin activity in the extract was determined by a modification of angiotensin I (ANG I) radioimmunoassay [8]. The non-specific renin-like action of cathepsin D was minimized by using as substrate unfractionated plasma of nephrectomized rats (100 µl) in the reaction with tissue extracts. Extracts (25–100 µl) were incubated for 1 h at 37°C in phosphate buffer (0-2 mol/l), pH 7-0, containing EDTA (7 mmol/l) and phenylmethanesulphonyl fluoride (2 mmol/l). The difference in renin activity of the extract before and after pretreatment with monospecific anti-renin antibody [7] was defined as specific renin activity.

Results
The specific renin activity in the adrenal and the mesenteric vessels of 3-week-old SHR with normal blood pressure was markedly elevated to
levels eight times those of the WKY rats. On the other hand, no difference was observed in the aorta. A similar pronounced increase was observed in the adrenal renin levels of adult (17 weeks of age) SHR with established hypertension, whereas no significant difference was observed in renin of the mesenteric vessels or aorta (Table 1).

The distribution of renin between adrenal cortex and medulla was determined in 17-week-old rats. The two regions were separated under a dissecting microscope. The cortex/medulla ratio of the specific activity of renin per mg of protein was 7.8±0.9 in SHR and 2.1±0.2 in WKY rats. The SHR/WKY rat ratio of the specific renin activity was 3.4±0.1 in the cortex and 2.7±1.0 in the medulla.

Discussion

The present study demonstrates markedly elevated levels of tissue renin in the adrenals and mesenteric vessels of SHR, but not in the aorta of SHR, compared with WKY rats. Elevated renin levels could be demonstrated at 3 weeks of age, before hypertension had developed. To date no other tissue has been found to contain an unequivocally elevated level of renin in SHR compared with that of a control. Although an elevated renin activity was reported in the aorta of SHR with established hypertension [9, 10], opposite results have been reported recently [11].

The elevated renin levels of the adrenal and mesenteric vessels may be analogous with the elevated levels of dopamine β-hydroxylase detected in these tissues in SHR [12]; this suggests a possible link with the adrenergic system during the development of hypertension. Although angiotensin II stimulates steroidogenesis in the adrenal cortex [13] and catecholamine release by the medulla [14], the fact that the renin level of adrenal cortex of SHR showed a more pronounced increase than that of the medulla suggests a possible major role for the former. The preventive effect of adrenalectomy on the development of hypertension [15] and the morphological observations suggesting adrenocortical hyperfunction [16] indicated an important role of the adrenal gland in spontaneous hypertension. Although early studies on plasma steroids did not support these observations [17, 18], higher levels of plasma aldosterone and corticosterone [19] as well as a critical role for aldosterone in the development of hypertension in SHR [20] have been demonstrated in recent studies. Since plasma renin is not elevated in SHR, it is likely that the elevated tissue renin—angiotensin system in the adrenal may play a role in the adrenocortical hyperfunction and, probably, in spontaneous hypertension, an intriguing hypothesis open to further investigation.

Acknowledgments

We are indebted for technical assistance to Mr Edward Price Jr. This research was supported by research grants HL-14192 and HL-24112 from the National Institutes of Health.
Adrenal and vascular renin in hypertensive rats


