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Central cholinergic neurons and receptors are implicated in the regulation of cardiovascular function. It has been shown that central cholinergic activation by cholinergic agonists or cholinesterase inhibitors has a significant influence on the blood pressure in several species including human (Hoffman: CLIN EXP PHARMACOL PHYSIOL 6: 373, 1979). Physiological studies have revealed that a pressor response to the cholinergic agonists was markedly greater in spontaneously hypertensive rats (SHR) than in Wistar Kyoto rats (WKY) (Buccafusco and Spector: J CARDIOVAS PHARMACOL 2: 347, 1980), suggesting an alteration in central cholinergic mechanisms in the hypertension. In the present study we have investigated the alteration of muscarinic and nicotinic cholinergic receptors by comparing the specific binding of radioligands, $[^3H]$quinuclidinyl benzilate (QNB) and $[^3H]$nicotine in brain regions (cerebral cortex, hippocampus, thalamus, hypothalamus, striatum, midbrain, cerebellum, brainstem) of WKY, SHR and stroke-prone SHR (SHRSP). The binding assays were performed using methods described previously (Yamamura and Snyder: PROC NATL ACAD SCI USA 71: 1725, 1974; Marks and Collins: MOL PHARMACOL 22: 554, 1982). In addition, the activity of choline acetyltransferase (ChAT) and acetylcholinesterase (AChE) was simultaneously determined (Ellman et al.: BIOCHEM PHARMACOL 7: 88, 1961; Fonnum: J NEUROCHEM 24: 407, 1975).

Specific binding of $[^3H]$QNB and $[^3H]$nicotine in the rat brain under our assay conditions showed a pharmacological drug profile compatible with muscarinic and nicotinic cholinopeptidases respectively. Hypothalamus and hippocampus but not other brain regions from SHRSP at 24 weeks of age (wks) showed a consistent and significant ($P<0.05$) increase in specific $[^3H]$QNB binding as compared to age-matched WKY. Scatchard analysis demonstrated an elevated density ($B_{max}$, WKY = 415 ± 34, SHR = 559 ± 50, SHRSP = 575 ± 48 fmol/mg protein, $n=6-8$) of hypothalamic muscarinic cholinopeptidases without a change in the affinity ($K_d$, WKY = 68.6 ± 10.6, SHR = 83.8 ± 11.9, SHRSP = 79.0 ± 10.4 pM). The enhancement of muscarinic cholinopeptidase density was observed in the hypothalamus of SHR and SHRSP at every age examined, including prehypertensive age (5 wks). Chronic treatment of young SHRSP with hypotensive drugs (hydralazine, methylothiazide and reserpine) for three months, although preventing the development of hypertension significantly ($P<0.001$), failed to reduce significantly an increased density of hypothalamic muscarinic cholinopeptidases. Neither renal nor DOCA hypertension in rats showed a significant alteration in hypothalamic muscarinic receptors as compared to normotensive control rats. In contrast to the enhancement of these muscarinic receptors, SHRSP at 16 wks demonstrated a significant ($P<0.01$) decrease in specific $[^3H]$nicotine binding in the lower brainstem (pons + medulla) but not in the hypothalamus. There was a 38% loss of nicotinic receptor density ($B_{max}$, WKY = 75.8 ± 8.4, SHRSP = 46.9 ± 2.8 fmol/mg protein, $n=7$) without a change in the affinity ($K_d$, WKY = 13.5 ± 1.0, SHRSP = 11.6 ± 1.3 nM), in the brain region of 24 wks SHRSP. It was found that the hypothalamus of both SHR and SHRSP at 5 and 24 wks had significantly less (18-28%, $P<0.05$) activity of ChAT than that of age-matched WKY (5 wks: WKY = 36.3 ± 2.3, SHR = 26.0 ± 0.3, SHRSP = 29.7 ± 1.7, 24 wks: WKY = 36.5 ± 2.8, SHRSP = 29.4 ± 1.6 nmol acetylcholine formed/g fresh tissue/min, $n=5-11$). On the other hand, the activity of AChE in the brain region was unaltered by spontaneous hypertension.

In conclusion, there was a specific alteration in the density of muscarinic and nicotinic cholinopeptidases in the hypothalamus and brainstem of SHR and SHRSP. Thus the present study provides a biochemical evidence for an important role of these cholinopeptidases in the pathogenesis of spontaneous hypertension.