Dopamine- β -hydroxylase and tyrosine hydroxylase activities in brain regions of rolling mouse *Nagoya*

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ABSTRACT

Dopamine- β -hydroxylase and tyrosine hydroxylase activities were specifically high in the cerebellum and brain stem of rolling mouse *Nagoya*, as compared to the control mouse. The activities in the thalamus-hypothalamus, cerebrum and caudate nucleus did not significantly differ between the rolling and control mice.

KEY WORDS Dopamine- β -hydroxylase / tyrosine hydroxylase / rolling mouse *Nagoya* / cerebellum / brain stem

Rolling mouse *Nagoya* is a murine neurological mutant which was developed in the course of mating experiments between male SIII and female C57BL/6 by Oda (9). The mice show a gait disorder recognized as ataxia with creeping or rolling. Genetic studies suggest that the mutation is transmitted by a single autosomal recessive gene (*rol*). Previously reported genetic mutations in the mouse with gait ataxia, i. e. weaver, staggerer and reeler result in severe hypoplastic cerebellum (10), but in the rolling mouse *Nagoya* hypoplasia of the cerebellum is not significant (8).

Recent investigations have revealed abnormalities in catecholaminergic neurons in the cerebellum and brain stem of the hypocerebellar mutant mice. Weaver, reeler and staggerer mice all manifest greatly increased specific catecholamine fluorescence per unit area of cerebellar cortex (5). The activities of tyrosine hydroxylase (TH), the initial enzyme in catecholamine biosynthesis, and dopamine- β -hydroxylase (DBH), which catalyzes the conversion of dopamine to noradrenaline, are increased in the nucleus locus coeruleus in weaver mice (2). On the

other hand, in the cerebellum of the rolling mouse, remarkable increases in numbers and fluorescence intensity of noradrenaline nerve terminals were observed, although no notable hypoplasia of cerebellum was detectable. These were observed in the anterior lobe more than in the posterior lobe (1).

We now report that in the rolling mouse *Nagoya*, the activity of DBH and TH is increased in the cerebellum and brain stem.

The gene of the rolling mouse *Nagoya* was introduced into C3Hf/Nem strain and the C3Hf/Nem-*rol* (M6) mice produced were then used for experiments. The male rolling mice (*rol/rol*) and female normal control (+/+) mice weighing about 20 g and aged about 4 months were examined. The mice were killed by decapitation, and the brain was quickly removed. Brain regions were dissected out and frozen on dry ice. The brain tissues were homogenized in 4 volumes of 0.1 M potassium phosphate buffer, pH 7.5, containing 0.1% Triton X-100. The homogenate was centrifuged at $50,000 \times g$ for 30 min, and the supernatant was used to assay for DBH and TH activities. DBH activity was assayed by the dual-wavelength spectrophotometric method based on the conversion of tyramine to octopamine (3). Tyrosine hydroxylase

Abbreviations: DBH, dopamine- β -hydroxylase; and TH, tyrosine hydroxylase

Table 1 Dopamine- β -Hydroxylase and Tyrosine Hydroxylase Activities in Brain Regions of The Neurological Murine Mutant - Rolling Mouse *Nagoya*

Brain regions	Brain stem	Cerebellum	Thalamus-hypothalamus	Cerebral cortex	Caudate nucleus
Dopamine- β -hydroxylase (nmol/hr/g tissue)					
Control mouse	497 $\pm$ 21 (4)	272 $\pm$ 5 (5)	486 $\pm$ 19 (4)	414 $\pm$ 16 (4)	215 $\pm$ 9 (4)
Rolling mouse	596 $\pm$ 20** (6)	361 $\pm$ 17** (6)	541 $\pm$ 18 (6)	409 $\pm$ 9 (6)	245 $\pm$ 17 (6)
Tyrosine hydroxylase (nmol/hr/g tissue)					
Control mouse	45.8 $\pm$ 5.1 (4)	31.8 $\pm$ 5.1 (8)	41.1 $\pm$ 4.6 (4)	23.9 $\pm$ 1.1 (4)	278 $\pm$ 19 (4)
Rolling mouse	57.4 $\pm$ 5.0 (4)	50.4 $\pm$ 5.4* (8)	46.1 $\pm$ 5.2 (6)	24.7 $\pm$ 5.1 (6)	285 $\pm$ 10 (6)

Values are means $\pm$ S. E. M., with the number of determination in parentheses. Control mouse, C3Hf/Nem, +/+; rolling mouse *Nagoya*, C3Hf/Nem, rol/rol.

* $P < 0.05$ and ** $P < 0.01$

activity was determined by measuring the formation of [14 C]dopa from L-[U- 14 C]tyrosine, using 1 mM 6-methyltetrahydropterin as cofactor (6, 7).

Activities of DBH and TH in brain regions of the rolling (rol/rol) and the control normal (+/+) are shown in Table 1.

DBH activity in the brain stem and cerebellum of the rolling mice was significantly higher than that of the control mice, but the activity in the thalamus-hypothalamus, cerebellum and caudate nucleus did not differ significantly between the rolling and the control mice.

TH activity in the cerebellum and brain stem of the rolling was also higher than that of the control mice. However, activity in the thalamus-hypothalamus, cerebellum and caudate nucleus of the rolling was similar to that of the control mice.

Konagaya *et al.* (4) have recently found a significantly higher TH activity and a significantly lower catechol-*O*-methyltransferase activity in the cerebellum and whole brain of rolling mouse *Nagoya*.

Present results suggest that DBH in the brain stem which contains cell bodies of noradrenergic neurons and in the cerebellum which is innervated by noradrenergic nerve terminals is increased in the rolling mouse *Nagoya*. This finding supports the previously reported large increase in numbers and fluorescence intensity of noradrenaline nerve terminals in the cerebellum of this mutant (1).

It is of interest that DBH and TH activities

in cerebellar projection from locus coeruleus appear to be specifically high in rolling mouse *Nagoya*, as compared with the activities in other noradrenergic projections. This may indicate an increased functional activity of a noradrenergic neuronal group projecting to the cerebellum which is different from those innervating to the rest of the brain. The cause of the increase in DBH and TH activities may be related to a genetic abnormality of the adrenergic neurons from the locus coeruleus to cerebellum in rolling mouse *Nagoya*, and could be causally related to the abnormal gait. However, the increase in DBH activity could also be due to the secondary effects external to the noradrenergic cells innervating from the locus coeruleus to the cerebellum.

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