The Involvement of Serotonergic and Dopaminergic Systems in Hypothermia Induced in Mice by Intracerebroventricular Injection of Serotonin

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Abstract—The hypothermia elicited in mice by intracerebroventricular injection (i.c.v.) of serotonin (5-HT) was studied. The 5-HT-induced hypothermia was attenuated by methysergide and pindolol, although ketanserin and p-chlorophenylalanine were without effect. These results suggest that 5-HT-induced hypothermia is produced by the direct activation of the 5-HT1 receptor. In addition, haloperidol, pimozide and α-MT inhibited 5-HT-induced hypothermia, which indicates that the dopaminergic systems are also involved in 5-HT-induced hypothermia in mice.

It is well recognized that serotonin (5-HT) is a neurotransmitter and plays an important role in the sleep-waking cycle, motor activity, feeding behavior, etc. (1). 5-HT is also involved in thermoregulation of the central nervous system. It has been shown that treatment with 5-HT receptor agonists or injection of 5-HT directly into the brain produces hypothermia in rats and mice (2-4). In recent binding studies, it has been suggested that 5-HT receptors can be divided into two main types: 5-HT1 and 5-HT2 receptors; and 5-HT1 receptors can be further subclassified as 5-HT1A, 5-HT1B and 5-HT1C receptors (5). However, the involvement of the 5-HT receptor subtypes in hypothermia induced by 5-HT itself has not been clarified. It has been suggested that dopamine or dopamine receptor agonists produce a fall in the body temperature by activating the dopamine receptors (4, 6). However, the mediation of dopamine in the thermoregulatory effects of 5-HT is not clear. We examined, therefore, the involvement of the 5-HT receptor subtypes and dopaminergic mechanisms in the hypothermia induced in mice by intracerebroventricular injection of 5-HT.

Male Slc:ICR mice (Shizuoka Laboratory Animals Center, Shizuoka, Japan) weighing 19-25 g were used. The mice were maintained at 24±1 °C, at 55±5% humidity and on a 12 hr-12 hr light/dark cycle (starting at 07:00). They were allowed food and water ad libitum, and all experiments were performed between 13:00 and 17:00 hr. Serotonin creatinine sulfate monohydrate (5-HT) was purchased from Merck (Darmstadt, West Germany), ketanserin tartrate from RBI (Natick, U.S.A.), (+)-pindolol from Sigma (St. Louis, U.S.A.), and p-chlorophenylalanine (p-CPA) and α-methyl-p-tyrosine methylester hydrochloride (α-MT) from Nakarai Chemicals Co., Ltd. (Kyoto, Japan). Methysergide hydrogen maleate and pimozide were generously donated by Sandoz (Basel, Switzerland) and Fujisawa Pharmaceutical Co., Ltd. (Osaka, Japan), respectively. Haloperidol was used as the commercially available injectable form, Serenase (Dai-Nippon Pharmaceutical Co., Ltd., Osaka, Japan). Methysergide hydrogen maleate and pimozide were generously donated by Sandoz (Basel, Switzerland) and Fujisawa Pharmaceutical Co., Ltd. (Osaka, Japan), respectively. Haloperidol was used as the commercially available injectable form, Serenase (Dai-Nippon Pharmaceutical Co., Ltd., Osaka, Japan). 5-HT, methysergide, ketanserin and α-MT were dissolved in saline. Pindolol was dissolved in a few drops of 0.1 N HCl and diluted with saline. p-CPA and pimozide were suspended in 1% carboxymethylcellulose-Na. The 5 μl of 5-HT in saline was injected i.c.v. according to the methods of Haley and McCormick (7). Control mice received 5 μl of saline injected in...
Methysergide at 1 mg/kg, ketanserin at 0.1 mg/kg, pindolol at 5 mg/kg and haloperidol at 0.1 mg/kg were injected i.p. 30 min before 5-HT. α-MT at 300 mg/kg and pimozide at 5 mg/kg were injected i.p. at 4 hr and 3 hr before 5-HT, respectively. p-CPA at 150 mg/kg was injected i.p. at 72, 48 and 24 hr before 5-HT. Rectal temperature was monitored using a thermometer (Natsume, Japan). The probe was inserted 2 cm into the rectum. Statistical significance of results was evaluated by a one way analysis of variance (ANOVA) and Dunnett's t-test. The thermal response index (TRI) (8) was calculated for the experiments with 5-HT and dopamine antagonists. A single unit of TRI is equivalent to a 1°C change in temperature lasting 15 min.

The effects on the rectal temperature of 5-HT given i.c.v. are shown in Fig. 1. 5-HT induced significant hypothermia in mice by i.c.v. injection. A maximum hypothermic response of 2.48°C was observed after the injection of 50 nmol of 5-HT. The maximum falls in rectal temperature occurred 10 or 15 min after the injection of 5-HT at all dosages used. Body temperature recovered almost to the control levels 60 min after administration of 5-HT.

The effects of 5-HT and dopamine antagonists on 5-HT-induced hypothermia are summarized in Table 1. As 5-HT induced the maximal drops in body temperature 10 or 15 min after the injection, the effects of the 5-HT and dopamine antagonists were evaluated 10 min after the administration of 5-HT at 10 nmol. The dosages of antagonists which not affect the basal body temperature were used. As shown in Table 1, the 5-HT1 and 5-HT2 receptor antagonist methysergide inhibited 5-HT-induced hypothermia. It has been suggested that the hypothermia induced in mice by 5-HT administered i.c.v. can be inhibited by the non-selective 5-HT receptor antagonist methysergide (3). In this study, the hypothermic effects of 5-HT given i.c.v. were confirmed, and the fact that methysergide inhibited the 5-HT-induced hypothermia indicates the mediation of the 5-HT receptor.

The 5-HT1 receptor antagonist pindolol inhibited the hypothermia induced by 5-HT, although the 5-HT2 receptor antagonist ketanserin did not affect it (Table 1). These results suggest that 5-HT-induced hypo-
thermia is mediated by the 5-HT\textsubscript{1}\, receptor. In addition, the depletion of 5-HT by p-CPA did not affect the hypothermia, which suggests that the hypothermia elicited by 5-HT administered i.c.v. is caused by the direct activation of the 5-HT\textsubscript{1}\, receptor. It has been reported that 5-HT\textsubscript{1A} receptor agonists, 8-hydroxy-2-(di-n-propylamino) tetralin (8-OH-DPAT) or LY 165163 induce hypothermia in mice and rats (9-11). Goodwin et al. (9) suggested that 8-OH-DPAT-induced hypothermia is mediated by the autoreceptor not by the post-synaptic 5-HT\textsubscript{1A} receptor, since 8-OH-DPAT-induced hypothermia is attenuated by p-CPA but not by the 5-HT receptor antagonists. However, recently, it has been reported that 8-OH-DPAT-induced hypothermia is inhibited by the 5-HT\textsubscript{1}\, receptor antagonist pindolol (10), but not by the 5-HT depleter α-MT (11). Taken together with these observations and our results, it is suggested that the hypothermia induced by injection of 5-HT directly into the brain is mediated by the 5-HT\textsubscript{1}\, receptor.

It has been noted that dopamine also plays a physiological role in thermoregulation (4, 12). Intrahypothalamic injection of dopamine produces a fall in the body temperature of rats caused by the activation of the dopamine receptors (4, 12). However, the effect of dopamine in hypothermia elicited by 5-HT is not yet clear. Therefore, we also examined the effects of dopamine antagonists on 5-HT-induced hypothermia. The dopamine receptor antagonists haloperidol and pimozide inhibited the 5-HT-induced hypothermia. It suggests that the dopamine receptors are involved in the hypothermic effects of 5-HT. In addition, the dopamine depleter α-MT attenuated the hypothermia elicited by 5-HT, which indicates the involvement of the endogenous dopamine in 5-HT-induced hypothermia. Cox and Lee (4) suggested that in rats, dopamine-induced hypothermia was blocked by the 5-HT receptor antagonists methysergide and cyproheptadine, although 5-HT-induced hypothermia was not inhibited by the dopamine receptor antagonists haloperidol and pimozide. However, we demonstrated that in mice, both dopamine receptor antagonists and a depleter inhibited 5-HT-induced hypothermia. This indicates that in mice, 5-HT-induced hypothermia is related to the dopaminergic mechanisms. There seem, therefore, to be some species differences between rats and mice in the involvement of dopamine in the hypothermic effects of 5-HT. It has been reported that acetylcholine may mediate the dopamine receptor-mediated hypothermia, since dopamine receptor agonist apomorphine-induced hypothermia in mice can be blocked by the acetylcholine receptor antagonist scopolamine (13). Therefore, the possible mediation of the cholinergic neurons in the hypothermic effects of 5-HT is also of increasing interest.

In conclusion, our results indicate that 5-
HT plays an important role in thermoregulation by the mediation of 5-HT₁ receptors, and the 5-HT-induced hypothermic effects in mice are also associated with dopaminergic systems.

References
1 Green, A.R. and Grahame-Smith, D.G.: 5-Hydroxytryptamine and other indoles in the central nervous system. In Handbook of Psychopharmacology, Vol. 3, Biochemistry of Biogenic Amines, Edited by Iversen, L.L., Iversen, S.D. and Snyder, S.H., p. 169–245, Plenum Press, New York (1975)
2 Yamada, J., Wakita, H., Sugimoto, Y. and Horisaka, K.: Hypothermia induced in mice by intracerebroventricular injection of tryptamine: involvement of the 5-HT₁ receptor. Eur. J. Pharmacol. 139, 117–119 (1987)
3 Ritzmann, R. and Tabakoff, B.: Is serotonin or are its metabolites responsible for induction of hypothermia? Experientia 32, 334–336 (1976)
4 Cox, B. and Lee, T.F.: Possible involvement of 5-hydroxytryptamine in dopamine-receptor-mediated hypothermia in the rat. J. Pharm. Pharmacol. 31, 352–354 (1975)
5 Conn, P.J. and Sanders-Bush, E.: Central serotonin receptors: effector systems, physiological roles and regulation. Psychopharmacology (Berlin) 92, 267–277 (1987)
6 Faunt, J.E. and Crocker, A.D.: The effects of selective dopamine receptor agonists and antagonists on body temperature in rats. Eur. J. Pharmacol. 133, 243–247 (1987)
7 Haley, T.J. and McCormick, W.G.: Pharmacological effects produced by intracerebral injection of drugs in the conscious mouse. J. Pharmacol. 12, 12–15 (1957)
8 Clark, W.G. and Cumby, H.R.: Hyperthermic responses to central and peripheral injections of morphine sulphate in the cat. Br. J. Pharmacol. 63, 65–71 (1978)
9 Goodwin, G.M., De Souza, R.J. and Green, A.R.: The pharmacology of the hypothermic response in mice to 8-hydroxy-2-(di-n-propylamino)-tetralin (8-OH-DPAT). Neuropharmacology 24, 1187–1194 (1985)
10 Gudelsky, G.A., Koenig, J.E. and Meltzer, H.Y.: Thermoregulatory responses to serotonin (5-HT) receptor stimulation in the rat. Neuropharmacology 25, 1307–1313 (1986)
11 Hutson, P.H., Donohoe, T.P. and Curzon, G.: Hypothermia induced by the putative 5-HT₁A agonists LY165163 and 8-OH-DPAT is not prevented by 5-HT depletion. Eur. J. Pharmacol. 143, 221–228 (1987)
12 Cox, B. and Lee, T.F.: Further evidence for a physiological role for hypothalamic dopamine in thermoregulation in the rat. J. Physiol. (Lond.) 300, 7–17 (1980)
13 Glick, S.D. and Marsanico, R.G.: Apomorphine-induced and pilocarpine-induced hypothermia in mice. Drug interactions and changes in drug sensitivity after caudate nucleus lesions. Br. J. Pharmacol. 51, 353–357 (1974)