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Lack of Cross-Tolerance for Hypophagia Induced by DOI versus m-CPP Suggests Separate Mediation by 5-HT_{2A} and 5-HT_{2C} Receptors, Respectively

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Intraperitoneal administration of 1-(2,5-dimethoxy-4-iodophenyl)-2-aminopropane (DOI) produced significant decreases in the first-hour food intake on day 1 and on day 2 relative to saline-treated animals. Complete tolerance developed to DOI-induced hypophagia by day 3. However, there was no cross-tolerance to m-chlorophenylpiperazine (m-CPP)-induced hypophagia. Similarly, complete tolerance developed to m-CPP-induced hypophagia by day 3, but again there was no cross-tolerance to DOI-induced hypophagia. These findings suggest that DOI and m-CPP-induced hypophagia are mediated by different mechanisms, most likely by selective stimulation of 5-HT_{2A} receptors by DOI and 5-HT_{2C} receptors by m-CPP. The functional sensitivity changes did not parallel changes in hypothalamic [^{3}H]-mesulergine-labeled 5-HT_{2C} receptors or [^{3}H]-ketanserin-labeled 5-HT_{2A} receptors following chronic m-CPP or DOI treatment, although both treatments significantly reduced 5-HT_{2A} and 5-HT_{2C} receptors in cortex. Thus, future studies investigating the effects of daily m-CPP and DOI administration on phosphoinositide hydrolysis or mRNA for 5-HT_{2C} and 5-HT_{2A} receptors in the hypothalamus might help explain the functional sensitivity changes observed in the present study.
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KEY WORDS: *Frontal cortex; Food intake; Tolerance; [^{3}H]-Mesulergine; [^{3}H]Ketanserin; Hypothalamus; Striatum*

There is a variety of evidence from studies in rats indicating that the serotonergic neurotransmitter system is involved in the regulation of food intake. This evidence is based on the peripheral or central administration of serotonin (5-HT) precursors, agonists, releasers or antagonists (Garattini et al. 1989; Samanin and Garattini 1990).

m-Chlorophenylpiperazine (m-CPP), a metabolite of the antidepressant drug trazodone, has mixed agonist and antagonist action at several 5-HT receptor subtypes. In radioligand binding studies, m-CPP possesses an approximately 10-fold higher affinity for 5-HT_{2C} versus 5-HT_{1A}, 5-HT_{1B}, and 5-HT_{2A} sites (Hoyer 1988). In functional studies, various investigations have led to the conclusion that most of m-CPP's effects are predominantly mediated by its 5-HT_{2C} agonist effects (Aulakh et al. 1992a; Berendsen et al. 1990; Kennett and Curzon 1991; Murphy et al. 1991).

1-(2,5-dimethoxy-4-iodophenyl)-2-aminopropane (DOI) and other phenylisopropylamines such as 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane (DOM) and 1-(2,5-dimethoxy-4-bromo-phenyl)-2-aminopro-

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pane (DOB) have been identified as potent 5-HT_{2A} agonists (Glennon 1986). In recent years DOI and DOB have been radioactively tagged and used to biochemically characterize 5-HT_{2A} receptors (Appel et al. 1990; Glennon et al. 1988). However, these phenylisopropylamine hallucinogens also compete strongly for [^{3}H]mesulergine-labeled 5-HT_{2C} receptors in homogenate binding assays, with a similar rank order of potency to that found at [^{3}H]-DOB labeled 5-HT_{2A} receptors (Titeler et al. 1988). Both 5-HT_{2A} and 5-HT_{2C} receptors have been shown to possess similar molecular structures (Julius et al. 1988; Pritchett et al. 1988); both are coupled to guanine nucleotide regulatory proteins (Hartig 1989), and stimulation of both of these receptors increase phosphotidylinositol turnover (Hoyer et al. 1989). These similarities led to the recent nomenclature change in which the former 5-HT_{1C} receptor was renamed 5-HT_{2C} and the former 5-HT_2 receptor designated 5-HT_{2A} (Humphrey et al. 1993).

Systemic administration of both DOI (Aulakh et al. 1992b) and m-CPP (Samanin et al. 1980) produce dose-related decreases in food intake in rats studied in a food-restricted paradigm. By using various 5-HT receptor subtype-selective antagonists, m-CPP-induced hypophagia has been suggested to be mediated by stimulation of 5-HT_{2C} receptors (Kennett and Curzon 1991). Both 5-HT_{2A} and 5-HT_{2C} receptors have been implicated in mediating DOI-induced hypophagia (Aulakh et al. 1992b). In an attempt to further clarify whether m-CPP and DOI produce hypophagia by the same or different mechanisms, we studied the time course of development of tolerance to the hypophagic effects of m-CPP and DOI and also checked for cross-tolerance between m-CPP and DOI. In addition, we studied the effects of chronic m-CPP and DOI administration on mesulergine-labeled 5-HT_{2C} and ketanserin-labeled 5-HT_{2A} receptors in different brain areas.

MATERIALS AND METHODS

Male Wistar rats obtained from Charles River (Kingston, NY) and weighing approximately 250 g at the beginning of the study were used. The animals were housed in a temperature-controlled (22 ± 1°C) room with a 12-hour light-dark cycle (lights on at 6:00 A.M.). Separate groups of animals were used for food intake and receptor binding studies.

Food Intake Study

The animals were housed individually and had free access to water. The animals were trained to take their daily food (Purina food pellets) from 10:00 A.M. to 2:00 P.M. for 10 days before drug treatment was begun. At the end of the first hour of food access, the remaining food was weighed, including the spillage, and the difference from the original amount constituted the measure of food intake.

To study the time course of development of tolerance and also to check for cross-tolerance, separate groups of animals were injected with either DOI (2.5 mg/kg) or m-CPP (2.5 mg/kg) or saline every day at 10 A.M., 10 minutes before placing the food into the cages. Food intake (1 hour) was recorded every day for each animal. On the fifth day, chronic DOI-treated (2.5 mg/kg/day × 4) animals were challenged with m-CPP (2.5 mg/kg), whereas chronic m-CPP-treated (2.5/kg/day × 4) animals were challenged with DOI (2.5 mg/kg) and their 1-hour food intake was recorded. In addition, a separate experiment was performed to check for development of tolerance following daily m-CPP and DOI administration. In this experiment separate groups of animals were injected with either m-CPP (2.5 mg/kg) or DOI (2.5 mg/kg) or saline every day for 5 days at 10 A.M. 10 minutes before placing the food into the cages. Food intake (1 hour) was recorded at baseline (day 0) and on the first and fifth day of m-CPP or DOI or saline injection.

5-HT Receptor Binding Study

As in the food intake study, the animals were housed individually and had free access to water. The animals were trained to take their daily food from 10:00 A.M. to 2:00 P.M. for 10 days before drug treatment was begun. Separate groups of animals were injected with either DOI (2.5 mg/kg/day) or m-CPP (2.5 mg/kg/day) or saline every day at 10:00 A.M. for four days. On the fifth day (24 hours after the last injection of either DOI or m-CPP or saline), the animals were sacrificed by decapitation. The whole brain was rapidly removed, and the frontal cortex, hippocampus, striatum, brainstem and hypothalamus were dissected on ice. Brain regions were immediately frozen on dry ice and stored for 2 weeks at −70°C until assayed.

Binding Assays for [^{3}H]Mesulergine

The binding assay for measuring 5-HT_{2C} receptors using [^{3}H]mesulergine was performed according to the method of Pazos et al. (1985) described in Hulihan-Giblin et al. (1993). In brief, brain tissue was homogenized in 50 mM Tris-HCl buffer, pelleted, and washed again. Between washes, the homogenates were incubated for 10 minutes at 37°C to remove endogenous serotonin. The final resuspension of the homogenate was in 50 volumes of Tris-HCl buffer to which 10 μm pargyline, 4 mM CaCl, 0.1% ascorbate, and 20 nM spiperone had been added. The total assay volume of 1.0 ml consisted of 400 μl of tissue suspension, 100 μl of buffer, or 100 μl of 5-HT as the displacing drug (final

concentration 10 μm), and 500 μl of [^{3}H]mesulergine (0.25–16.0 nM). Immediately following the addition of [^{3}H]mesulergine, the assay tubes were incubated at 37°C for 10 minutes. The reaction was terminated by rapid filtration through Whatman GF/C filters and the addition of 3 × 5 ml of ice-cold Tris-HCl buffer using a Brandel 48-well cell harvester. Specific binding represented 60% to 70% of total binding.

Binding Assay for [^{3}H]Ketanserin

The assay for the binding of [^{3}H]ketanserin to 5-HT_{2A} receptors was performed according to the method of Leysen et al. (1982) as described in Hulihan-Giblin et al. (1992). Samples of brain were prepared as described for the binding assay for [^{3}H]mesulergine, with the omission of the 10-minutes preincubation at 37°C. The assay tubes were incubated for 15 minutes at 37°C in the dark, and the reaction was terminated as before. At each concentration of [^{3}H]ketanserin, 2 μM methysergide was added to half the tubes, to determine nonspecific binding; specific binding represented 60% to 70% of total binding.

Data Analysis

For the food intake studies, the data were analyzed using Student's *t*-test. For the receptor binding studies, the dissociation constant (K_d) and maximum number of bindings sites (B_{max}) were calculated using the LIGAND program, a nonlinear model-fitting program written for the Apple Macintosh computer (Rodbard and Munson, NIH, Bethesda, MD). Comparisons of B_{max} and K_d values between saline-treated and m-CPP-treated or DOI-treated animals were made using Student's *t*-test.

RESULTS

The time course of development of tolerance to the hypophagic effect of DOI is shown in Figure 1. Daily administration of DOI (2.5 mg/kg) produced essentially complete tolerance to its hypophagic effect by day 3. However, there was no cross-tolerance to m-CPP since m-CPP (2.5 mg/kg) administration produced significant decreases in 1-hour food intake in DOI-treated (2.5 mg/kg/day × 4) animals relative to saline-treated animals (Figure 1), with an effect of similar magnitude to that found after a single 2.5-mg/kg dose of m-CPP given to naive rats (Figure 2). On the other hand, DOI administration failed to produce a significant decrease in 1-hour food intake in DOI-treated (2.5 mg/kg/day × 4) animals relative to saline-treated animals (Figure 3).

The time course of development of tolerance to the hypophagic effect of m-CPP is shown in Figure 2. Daily administration of m-CPP (2.5 mg/kg) produced essentially complete tolerance to its hypophagic effect by day 3. However, there was no cross-tolerance to DOI since DOI (2.5 mg/kg) administration produced significant decreases in 1-hour food intake in m-CPP-treated (2.5 mg/kg/day × 4) animals relative to saline-treated

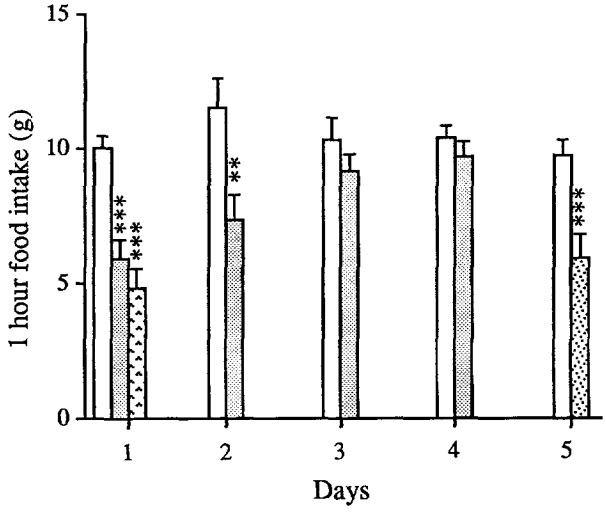

Figure 1. Effects of daily administration of saline or DOI (2.5 mg/kg/day) on 1-hour food intake changes and m-CPP-induced hypophagia in Wistar rats. Values are expressed as means ± SEM from six animals. Values of DOI-treated or m-CPP-treated animals significantly different from saline-treated animals are represented by $^{**}p < .01$; $^{***}p < .001$. *White bars*: saline; *grey bars*: DOI; *arrowheads*: m-CPP (2.5 mg/kg); *dots*: DOI + m-CPP.

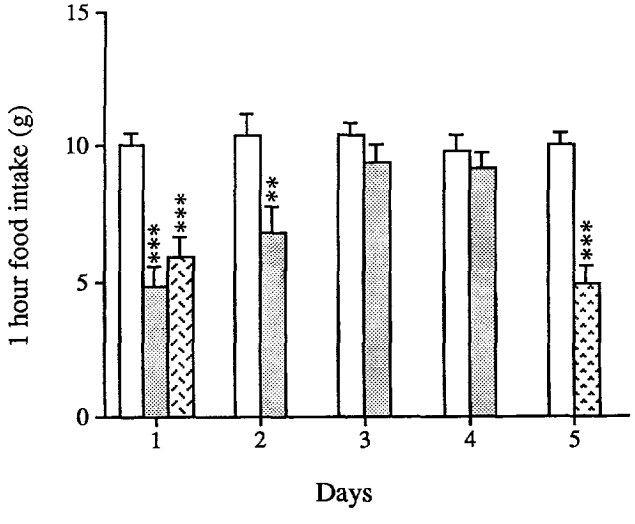

Figure 2. Effects of daily administration of saline or m-CPP (2.5 mg/kg/day) on 1-hour food intake changes and DOI-induced hypophagia in Wistar rats. Values are expressed as means ± SEM from six animals. Values of DOI-treated or m-CPP-treated animals significantly different from saline-treated animals are represented by $^{**}p < .01$; $^{***}p < .001$. *White bars*: saline; *gray bars*: m-CPP; *cross-hatching*: DOI (2.5 mg/kg); *arrowheads*: m-CPP + DOI.

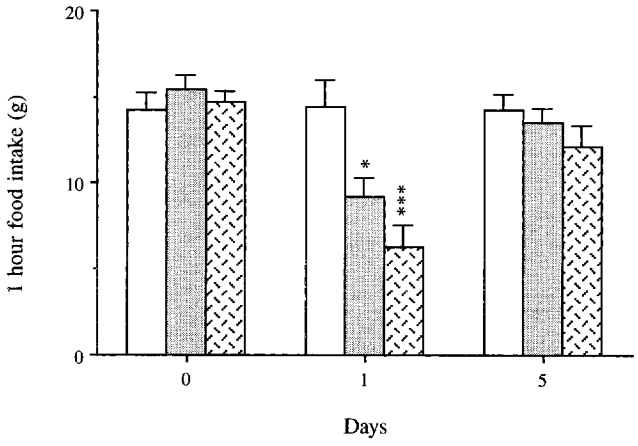

Figure 3. Effects of daily administration of saline or m-CPP (2.5 mg/kg/day) or DOI (2.5 mg/kg/day) on 1-hour food intake changes in Wistar rats. Values are expressed as means ± SEM from six animals. Values of m-CPP-treated or DOI-treated animals significantly different from saline-treated animals are represented by $^{*}p < .05$; $^{***}p < .001$. *White bars*: saline; *gray bars*: m-CPP; *cross-hatching*; DOI.

animals (Figure 2), with an effect of similar magnitude to that found after a single 2.5 mg/kg dose of DOI given to naive rats (Figure 1). On the other hand, m-CPP administration failed to produce a significant decrease in 1-hour food intake in m-CPP-treated (2.5 mg/kg/day × 4) animals relative to saline-treated animals (Figure 3).

The effects of daily administration of m-CPP and DOI on [^{3}H]mesulergine binding in different brain areas are shown in Figure 4. The B_{max} values for [^{3}H]mesulergine binding in the frontal cortex, brainstem, and striatum were found to be significantly lower in both the m-CPP-treated (2.5 mg/kg/day × 4) and DOI-treated (2.5 mg/kg/day × 4) animals relative to saline-treated animals (Figure 4). On the other hand, B_{max} values were significantly higher in the hypothalamus in DOI-treated, but not m-CPP-treated animals relative to saline-treated animals (Figure 4). There were no significant differences in K_d values between saline-treated and m-CPP-treated or DOI-treated animals in any of the brain regions examined (Table 1).

The effects of subchronic administration of m-CPP and DOI on [^{3}H]ketanserin binding in different brain areas are shown in Figure 5. The B_{max} values for [^{3}H]ketanserin binding in the frontal cortex were significantly lower in both the m-CPP-treated (2.5 mg/kg/day × 4) and DOI-treated (2.5 mg/kg/day × 4) animals relative to saline-treated animals (Figure 5). On the other hand, B_{max} values were significantly higher in the hypothalamus in m-CPP-treated, but not DOI-treated animals relative to saline-treated animals. In the striatum, hippocampus, and brainstem, no significant differences were observed in 5-HT$_{2A}$ receptor densities between saline-treated and m-CPP-treated or DOI-treated animals (Figure 5). There were no significant differences in K_d values between saline-treated and m-CPP-treated or DOI-treated animals in any of the brain regions examined (Table 1).

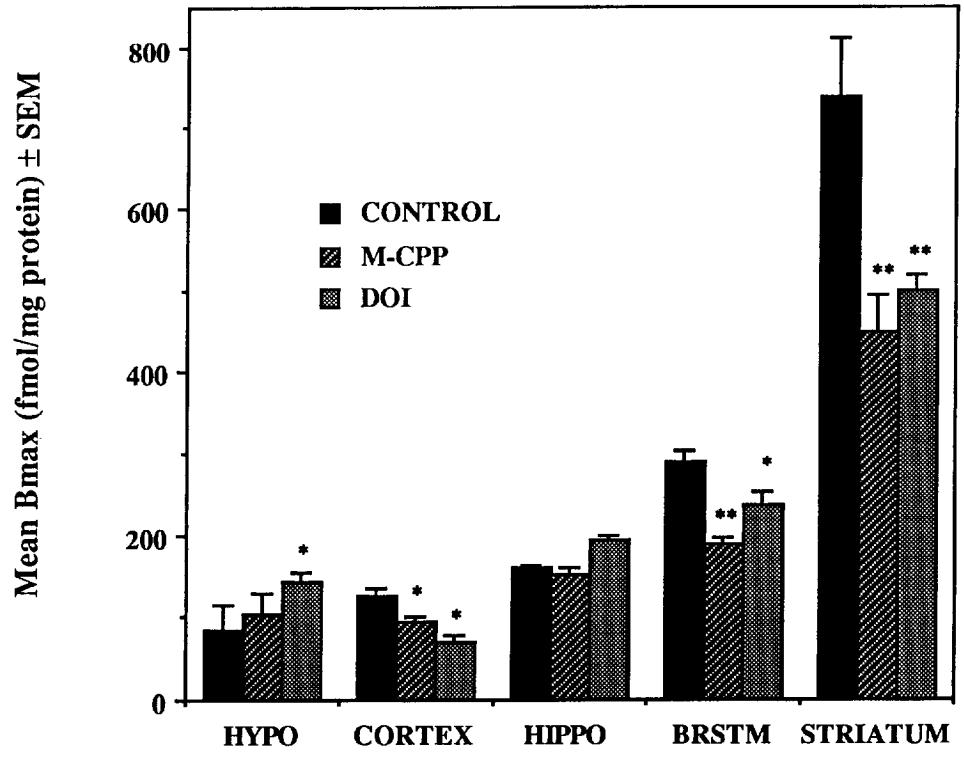

Figure 4. [^{3}H]Mesulergine binding in different brain areas in 4-day m-CPP-treated, 4-day DOI-treated and saline-treated animals. B_{max} values are expressed as means ± SEM from six animals. Value of m-CPP-treated or DOI-treated animals significantly different from saline-treated animals are represented by $^{*}p < .05$; $^{**}p < .01$.

Table 1. k_d Values (nM) for [^{3}H]Ketanserin Binding to 5-HT$_{2A}$ Receptors and [^{3}H]Mesulergine Binding to 5-HT$_{2C}$ Receptors in 4-Day DOI-Treated, 4-Day m-CPP-Treated and Saline-Treated Animals

	Control		DOI		m-CPP	
Brain Region	**[^{3}H]Ketanserin**	**[^{3}H]Mesulergine**	**[^{3}H]Ketanserin**	**[^{3}H]Mesulergine**	**[^{3}H]Ketanserin**	**[^{3}H]Mesulergine**
Striatum	0.86 ± 0.02	0.22 ± 0.02	0.95 ± 0.07	0.21 ± 0.07	0.89 ± 0.04	0.22 ± 0.01
Hypothalamus	0.87 ± 0.03	0.26 ± 0.02	1.00 ± 0.06	0.27 ± 0.01	0.91 ± 0.05	0.28 ± 0.02
Hippocampus	1.05 ± 0.05	0.23 ± 0.02	1.10 ± 0.03	0.17 ± 0.01	1.25 ± 0.05	0.23 ± 0.03
Frontal cortex	0.77 ± 0.02	0.26 ± 0.02	0.75 ± 0.03	0.28 ± 0.02	0.08 ± 0.04	0.28 ± 0.02
Brainstem	1.01 ± 0.03	0.29 ± 0.03	1.24 ± 0.06	0.28 ± 0.02	1.19 ± 0.04	0.30 ± 0.02

Values are expressed as mean ± SEM from six animals. There were no significant differences between saline-treated and m-CPP-treated or DOI-treated in any brain region.

DISCUSSION

The demonstration of reductions in food intake following systemic administration of either DOI or m-CPP in the present study is consistent with several earlier reports (Aulakh et al. 1992b; Kennett and Curzon 1991; Samanin et al. 1979; Schechter and Simansky 1988). DOI-induced hypophagia is mediated by postsynaptic 5-HT receptors since this effect is attenuated by the 5-HT receptor antagonists metergoline, mesulergine, mianserin, and ritanserin (Aulakh et al. 1992b), as well as by ketanserin and 1-(1-naphthyl)-piperazine (1-NP) (Schechter and Simansky 1988). Similarly, m-CPP-induced hypophagia is also mediated by postsynaptic 5-HT receptors since it is attenuated by mesulergine, metergoline, and mianserin (Kennett and Curzon 1991).

The present study further demonstrated that daily administration of DOI produced tolerance to its hypophagic effect but did not produce cross-tolerance to m-CPP-induced hypophagia. Similarly, daily administration of m-CPP also produced tolerance to its hypophagic effect but did not produce cross-tolerance to DOI-induced hypophagia. These findings strongly suggest that DOI and m-CPP administration produce hypophagia by different mechanisms. In radioligand studies, DOI has been shown to have similar high affinity at both 5-HT$_{2A}$ (Appel et al. 1990; Glennon et al. 1988) and 5-HT$_{2C}$ receptor sites (Titeler et al. 1988). In contrast, m-CPP has been reported to possess approximately 10-fold higher affinity for 5-HT$_{2C}$ versus 5-HT$_{1A}$, 5-HT$_{1B}$, and 5-HT$_{2A}$ sites (Hoyer 1988). In addition to its direct interaction with 5-HT receptors, m-CPP has also been reported to exhibit presynaptic actions such as enhancing 5-HT release in hypothalamic

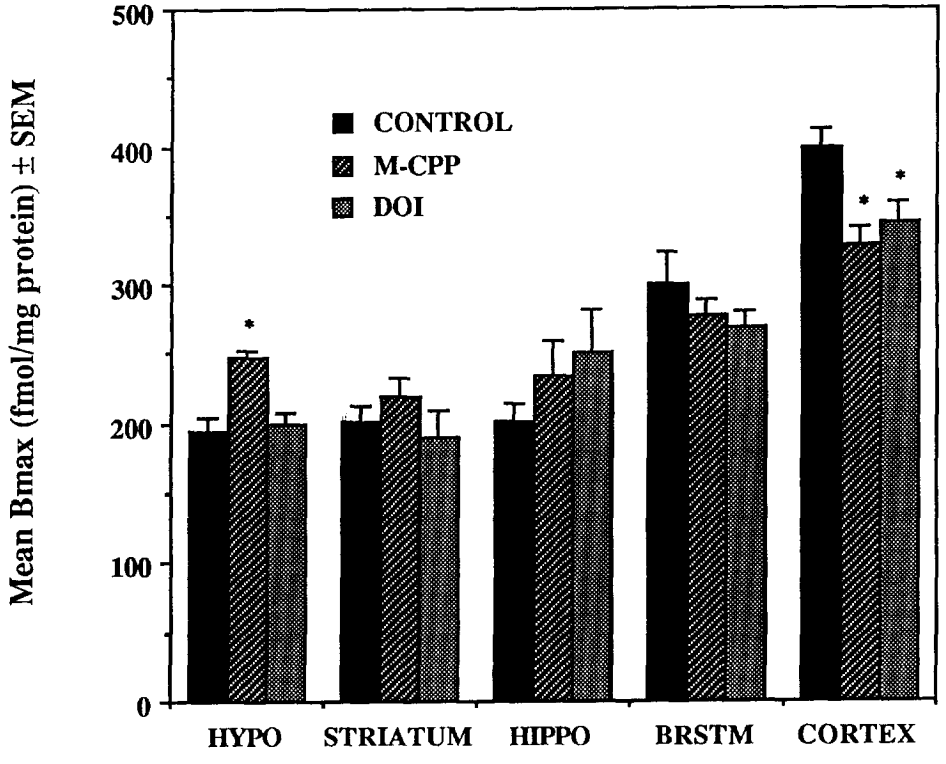

Figure 5. [^{3}H]Ketanserin binding in different brain areas in 4-day DOI-treated, 4-day m-CPP-treated and saline-treated animals. B_{max} values are expressed as mean ± SEM from six animals. Values of DOI-treated or m-CPP-treated animals significantly different from saline-treated animals are represented by $^*p < .05$.

slices (Pettibone and Williams 1984) and whole-brain synaptosomes (Wolf and Kuhn 1991), as well as increasing extracellular 5-HT in the ventromedial diencephalon in a microdialysis study following systemic administration (Baumann et al. 1993). It has been suggested that m-CPP-induced hypophagia is mediated predominantly by 5-HT_{2C} receptors on the basis of its blockade by relatively selective 5-HT receptor antagonists (Kennett and Curzon 1991). Moreover, m-CPP has been reported to act as an antagonist at 5-HT_{2A} receptors, mediating phosphoinositide hydrolysis in rat brain cortex (Conn and Sanders-Bush 1987). m-CPP also blocks the head twitch response produced by 5-Me-ODMT or 5-HTP in mice and the head shakes produced by quipazine in rats, two other 5-HT_{2A}-mediated responses (Simansky and Schechter 1988). Thus, on the basis of previously published studies using various 5-HT receptor subtype-selective antagonists to block the hypophagia produced by m-CPP (Kennett and Curzon 1991) or DOI (Aulakh et al. 1992b) and on the basis of the lack of cross-tolerance for hypophagia induced by DOI versus m-CPP observed in the present study, it would appear that DOI- and m-CPP-induced hypophagia are mediated by different mechanisms, most likely by stimulation of 5-HT_{2A} and 5-HT_{2C} receptors, respectively.

The present study also demonstrates that daily administration of m-CPP or DOI for 4 days produced significant but small decreases in [^{3}H]ketanserin-labeled 5-HT_{2A} receptors only in the frontal cortex. However, [^{3}H]mesulergine-labeled 5-HT_{2C} receptors were found to be significantly reduced in the brainstem and striatum in addition to the frontal cortex. We did not observe any decrease in either 5-HT_{2A} or 5-HT_{2C} receptors in the hypothalamus following daily administration of either DOI or m-CPP. Actually, 5-HT_{2C} receptor density was found to be significantly increased in the hypothalamus in DOI-treated animals, and 5-HT_{2A} receptor density was significantly increased in the hypothalamus in m-CPP-treated animals. Other investigators have also reported decreases in cortical 5-HT_{2A} sites labeled by [^{3}H]ketanserin but not [^{125}I] DOI following chronic administration of DOI (Himeno et al. 1988). On the other hand, chronic daily treatment with DOI (39 mg/kg) was reported to downregulate 5-HT_{2A} sites in cortex labeled either by [^{3}H]ketanserin or [^{3}H]DOB (Pranzatilli 1991). Recently, chronic treatment with both DOI and m-CPP was shown to decrease [^{3}H]mesulergine-labeled 5-HT_{2C} binding sites in the spinal cord (Pranzatelli et al. 1993). However, it is of note that the hypothalamus is involved in the regulation of food intake, while we found no reductions in either 5-HT_{2A} or 5-HT_{2C} receptor binding in the hypothalamus. All of the above-mentioned studies, including the present one, observed decreases in 5-HT_{2A} and 5-HT_{2C} receptor binding only in other brain areas. However, there are many examples in which serotonin receptor binding changes do not simply reflect concomitant serotonin functional sensitivity changes (Leysen 1984; Samanin et al. 1980). Furthermore, there is growing evidence that subsensitive functional responses to receptor activation during chronic antidepressant drug treatment are not invariably a consequence of desensitization and downregulation of the receptor itself (Hensler et al. 1991).

In the present study the B_{max} values for [3H]mesulergine binding in the control animals were found to be higher in all brain areas as compared to our own two previous studies (Hulihan-Giblin et al. 1993, 1994). However, it is of note that in the present study the animals were individually housed as well as food-deprived, whereas in our previous studies, the animals were group-housed and also had free access to food. Food-deprivation markedly reduces brain 5-HT and 5-hydroxyindoleacetic acid (5-HIAA) concentrations (Squadrito et al. 1994). It is also known that depletion of brain 5-HT concentrations following either parachlorophenylalanine (PCPA) or chemical denervation of serotonergic neurons with 5,7-dihydroxytryptamine (5,7-DHT) significantly increases [^{3}H]mesulergine-labeled 5-HT_{2C} receptor binding sites in various brain areas, including the amygdala, the hippocampus, and the nucleus accumbens (Rocha et al. 1993) and also leads to a supersensitive 5-HT_{2C}-mediated phosphoinositide hydrolysis response in rat choroid plexus (Conn et al. 1987). Thus, it is possible that the higher B_{max} values for [^{3}H]mesulergine binding observed in the present study may be due to the housing and feeding conditions.

The demonstration in the present study of functional subsensitivity of 5-HT_{2C}-mediated hypophagia following chronic m-CPP administration is consistent with other reports demonstrating functional subsensitivity of 5-HT_{2C} receptors mediating hypoactivity (Sills et al. 1985) and hyperthermia (Mazzola-Pomietto et al. 1993) following chronic m-CPP. However, m-CPP-induced increases in plasma prolactin were not altered by chronic m-CPP administration (Ulrichsen et al. 1992). m-CPP-induced prolactin release is also mediated by stimulation of 5-HT_{2C} receptors (Aulakh et al. 1992a). It is of note that chronic m-CPP administration decreased hypothalamic dopamine (DA) levels, which may be responsible for the failure to observe an attenuation of m-CPP's effect on prolactin levels in chronic m-CPP-treated animals (Ulrichsen et al. 1992).

In summary, the lack of cross-tolerance for hypophagia induced by DOI versus m-CPP suggests that DOI- and m-CPP-induced hypophagia are mediated by different mechanisms, most likely by selective stimulation of the 5-HT_{2A} and 5-HT_{2C} receptors, respectively.

REFERENCES

Appel NM, Mitchell WM, Garlick RK, Glennon RA, Teitler M, DeSouza EB (1990): Autoradiographic characterization of (±)-1-(2,5-dimethoxy-4-[^{125}I]iodophenyl)-2-aminopropane ([^{125}I] DOI) binding to 5-HT_2 and 5-HT_{1C} receptors in rat brain. J Pharmacol Exp Ther 255:843–857

Aulakh CS, Hill JL, Murphy DL (1992a): Effects of various serotonin receptor subtype selective antagonists alone and on m-CPP-induced neuroendocrine changes in rats. J Pharmacol Exp Ther 263:588–595

Aulakh CS, Hill JL, Yoney HT, Murphy DL (1992b): Evidence for involvement of 5-HT_{2C} and 5-HT_2 receptors in the food intake suppressant effects of 1-(2,5-dimethoxy-4-iodophenyl)-2-aminopropane (DOI). Psychopharmacology 109:444–448

Baumann MH, Rutter JJ, Auerbach SB (1993): Intravenous administration of the serotonin agonist m-chlorophenylpiperazine (mCPP) increases extracellular serotonin in the diencephalon of awake rats. Neuropharmacology 32:1381–1386

Berendsen HHG, Jenck F, Broekkamp CLE (1990): Involvement of 5-HT_{1C} receptors in drug-induced penile erections in rats. Psychopharmacology 101:57–61

Conn PJ, Sanders-Bush E (1987): Relative efficacies of piperazines at the phosphoinositide hydrolysis-linked serotonergic (5-HT_2 and 5-HT_{1C}) receptors. J Pharmacol Exp Ther 242:552–557

Conn PJ, Janowsky A, Sanders-Bush E (1987): Denervation supersensitivity of 5-HT_{1C} receptors in rat choroid plexus. Brain Res 400:396–398

Garattini S, Mennini T, Samanin R (1989): Reduction of food intake by manipulation of central serotonin. Current experimental research. Br J Psychiatry 155:41–51

Glennon RA (1986): Discriminative stimulus properties of the serotonergic agent 1-(2,5-dimethoxy-4-iodophenyl)-2-aminopropane (DOI). Life Sci 39:825–830

Glennon RA, Seggel MR, Soine WH, Herrick-Davis K, Lyon RA, Titeler M (1988): [I^{125}]-1-(2,5-Dimethoxy-4-iodophenyl)-2-aminopropane: An iodinated radioligand that specifically labels the agonist high-affinity state of 5-HT_2 serotonin receptors. J Med Chem 31:5–7

Hartig PR (1989): Molecular biology of 5-HT receptors. Trends Pharmacol Sci 10:64–69

Hensler JG, Kovachich GB, Frazer A (1991): A quantitative autoradiography study of 5-HT_{1A} receptor regulation: Effect of 5,7-dihydroxytryptamine and antidepressant treatments. Neuropsychopharmacology 4:131–144

Himeno A, McKenna DJ, Nazarali AJ, Saavedra JM (1988): (±)DOI, a hallucinogenic phenylalkylamine, down-regulates 5-HT_2 receptors in rat cortex. Soc Neurosci Abstr 14:575

Hoyer D (1988): Functional correlates of serotonin 5-HT_1 recognition sites. J Recept Res 8:59–81

Hoyer D, Waeber C, Schoeffter P, Palacios JM, Dravid A (1989): 5-HT_{1C} receptor-mediated stimulation of inositol phosphate production in pig choroid plexus. A pharmacological characterization. Naunyn-Schmiedeberg's Arch Pharmacol 339:252–258

Hulihan-Giblin BA, Park YD, Aulakh CS, Goldman D (1992): Regional analysis of 5-HT_{1A} and 5-HT_2 receptors in the Fawn-Hooded rat. Neuropharmacology 31:1095–1099

Hulihan-Giblin BA, Park YD, Goldman D, Aulakh CS (1993): Analysis of the 5-HT_{1C} receptor and the serotonin uptake site in Fawn-Hooded rat brain. Eur J Pharmacol 239:99–102

Hulihan-Giblin BA, Park YD, Aulakh CS (1994): Differential effects of chronic antidepressant treatment on 5-HT_{1C} receptor binding sites in Wistar rat brain. Eur J Pharmacol 263:213–216

Humphrey PPA, Hartig P, Hoyer D (1993): A proposed new nomenclature for 5-HT receptors. Trends Pharmacol Sci 14:233–236

Julius D, MacDermott AB, Azel R, Jessell TM (1988): Molecular characterization of a functional cDNA encoding the serotonin 1C receptor. Science 241:558–564

Kennett GA, Curzon G (1991): Potencies of antagonists indicate that 5-HT_{1C} receptors mediate 1-3(chlorophenyl) piperazine-induced hypophagia. Br J Pharmacol 103: 2016–2020

Leysen J (1984): Problems in *in vitro* receptor binding studies and identification and role of serotonin receptor sites. Neuropharmacology 23:247–254

Leysen JE, Niemegeers CJE, VanNueten JM, Laduron PM (1982): [^{3}H]Ketanserin (R41 468) a selective ^{3}H-ligand for serotonin$_2$ receptor binding sites. Binding properties, brain distribution, and functional role. Mol Pharmacol 21:301–314

Mazzola-Pomietto P, Aulakh CS, Wozniak KM, Johnson D, Murphy Dl (1993): Evidence that m-CPP-induced hyperthermia in rats is mediated by 5-HT_{1C} receptors. Soc Neurosci Abstracts 19:1380

Murphy DL, Lesch KP, Aulakh CS, Pigott TA (1991): Serotonin-selective arylpiperazones with neuroendocrine, behavioral, temperature, and cardiovascular effects in humans. Pharmacol Rev 43:527–552

Pazos A, Hoyer D, Palacios JM (1985): The binding of serotonergic ligands to the porcine choroid plexus: Characterization of a new type of serotonin recognition site. Eur J Pharmacol 106:539

Pettibone DJ, Williams, M (1984): Serotonin-releasing effects of substituted piperazine *in vitro*. Biochem Pharmacol 33:1531–1535

Pranzatelli MR (1991): Regulation of 5-HT_2 receptors in rat cortex: Studies with a putative selective agonist and an antagonist. Biochem Pharmacol 42:1099–1105

Pranzatelli MR, Murthy JN, Tailer PT (1993): Novel regulation of 5-HT_{1C} receptors: Down-regulation induced both by 5-$HT_{1C/2}$ receptor agonists and antagonists. Eur J Pharmacol (Mol Pharmacol Sec) 244:1–5

Pritchett DB, Bach AWJ, Wozny M, Taleb O, DalToso R, Shih JC, Seeburg PH (1988): Structure and functional expression of cloned rat serotonin 5-HT_2 receptor. EMBO J 7:4135–4140

Rocha B, DiScala G, Rigo M, Hoyer D, Sandner G (1993): Effect of 5,7-dihydroxytryptamine lesion on mianserin-induced conditioned place aversion and on 5-hydroxytryptamine$_{1C}$ receptors in the rat brain. Neuroscience 56:687–693

Samanin R, Garattini S (1990): The pharmacology of serotoninergic drugs affecting appetite. In Wurtman RJ, Wurtman JJ (eds) Nutrition and the Brain, New York, Raven, pp 163–192

Samanin R, Caccia S, Bendotti C, Borsini F, Borroni E, Garattini S (1979): m-Chlorophenylpiperazine: A central serotonin agonist causing powerful anorexia in rats. Naunyn Schmidberg Arch Pharmacol 308:159–163

Samanin R, Mennini T, Ferraris A, Bendotti C, Borsini F (1980): Hyper- and hyposensitivity of central serotonin receptors: ^{3}H-serotonin binding and functional studies in the rat. Brain Res 189:449–457

Schechter LE, Simansky KJ (1988): 1-(2,5-Dimethoxy-4-iodophenyl)-2-aminopropane (DOI) exerts an anorexic action that is blocked by 5-HT_2 antagonists in rats. Psychopharmacology 94:342–346

Sills MA, Lucki I, Frazer A (1985): Development of selective tolerance to the serotonin behavioral syndrome and suppression of locomotor activity after repeated administration of either 5-MeODMT or m-CPP. Life Sci 36: 2463–2469

Simansky KJ, Schechter LE (1988): Properties of some l-arylpiperazines as antagonists of stereotyped behaviors mediated by central serotonergic receptors in rodents. J Pharmacol Exp Ther 247:1073–1081

Squadrito F, Calapai G, Altavilla D, Cucinotta D, Zingarelli B, Campo GM, Arcoraci V, Sautebin L, Mazzaglia G, Caputi AP (1994): Food deprivation increases brain nitric oxide synthase and depresses brain serotonin levels in rats. Neuropharmacology 33:83–86

Titeler M, Lyon RA, Glennon RA (1988): Radioligand binding evidence implicates the brain 5-HT_2 receptor as a site of action of LSD and phenylisopropylamine hallucinogens. Psychopharmacology 94:213–216

Ulrichsen J, Partilla JS, Dax EM (1992): Long-term administration of m-chlorophenylpiperazine (m-CPP) to rats induces changes in serotonin receptor binding, dopamine levels and locomotor activity without altering prolactin and corticosterone secretion. Psychopharmacology 107: 229–235

Wolf WA, Kuhn DM (1991): The 5-HT transporter is an additional site of action for the 5-HT agonists RU 24969 and TFMPP. Neurochem Int 19:39–44