Pharmacology Biochemistry and Behavior, Vol. 65, No. 2, pp. 275–279, 2000

0091-3057/00/$–see front matter

ELSEVIER

PII S0091-3057(99)00198-7

Effects of Adrenoceptor Agents on Apomorphine-Induced Licking Behavior in Rats

MOHAMMAD-REZA ZARRINDAST,* SOHEILA FAZLI-TABAEI,†
SAEED SEMNANIAN,‡ YAGHOUB FATHOLLAHI§ AND SEYED HOSSEIN YAHYAVI†

*Department of Pharmacology, School of Medicine, Tehran University of Medical Sciences, Tehran, Iran;
†Department of Physiology, Azad University of Tehran;
‡Institute of Biochemistry and Biophysics, Tehran University; and
§Department of Physiology, School of Medical Sciences, Tarbiat-Modares University, Tehran, Iran

Received 10 July 1998; Revised 9 July 1999; Accepted 6 August 1999

ZARRINDAST, M. R., S. FAZLI-TABAEI, S. SEMNANIAN, Y. FATHOLLAHI AND S. H. YAHYAVI. *Effects of adrenoceptor agents on apomorphine-induced licking behavior in rats.* PHARMACOL BIOCHEM BEHAV **65**(2) 275–279, 2000.—In the present study, intraperitoneal (IP) administration of the dopaminergic receptor agonist apomorphine (0.1, 0.25, and 0.5 mg/kg) induced a dose-dependent licking in rats. The intraperitoneal injection of the α_1″adrenoceptor agonist phenylephrine (1–8 mg/kg) but not the α_2-adrenoceptor agonist clonidine (0.025–0.05 mg/kg) decreased licking induced by apomorphine. The α-adrenoceptor antagonists prazosin, phenoxybenzamine, and yohimbine also reduced the apomorphine response significantly. The response induced by phenylephrine was decreased by a dose of prazosin. The β_1-adrenenocepor agonist dobutamine and β_2-adrenenocepor agonist salbutamol did not alter the apomorphine response. However, β_2-adrenenocepor antagonists atenolol and propranolol reduced the apomorphine effect. It may be concluded that α_1- and possibly β_1-adrenoceptor mechanisms may be involved in modulation of licking behavior. © 2000 Elsevier Science Inc.

Adrenergic agents Apomorphine Licking Rats

APOMORPHINE directly activates dopamine receptors in the brain (17,19), and larger doses of the drug induced stereotyped behaviors (sniffing, licking, and gnawing) (1,5). The drug also elicits stereotyped behaviors such as locomotion (23) in mice, yawning (27), chewing (26), and penile erection (29) in rats, while it produces pecking in pigeons (24) and chicks (22). The stimulant effect of high doses of apomorphine is attributed to activation of postsynaptic receptors in the central nervous system (1). The behavioral responses observed in animals after administration of the dopamine agonist apomorphine are attributed to activation of D_1 and D_2 receptors (17,19).

Mesolimbic and nigrostriatal dopaminergic pathways may be important in the mediation of locomotor activity and stereotyped behaviors. Locomotion has been related to nucleus accumbens (9), whereas stereotyped behaviors (e.g., head movement, licking, and localized sniffing) are more closely associated with the caudate striatum (12).

There is a report indicating that adrenoceptor mechanisms may influence locomotion and stereotyped behaviors in the rat (4). The purpose of the present study was to determine the possible interaction of adrenoceptor mechanisms with the apomorphine-induced licking in rats.

METHOD

Animals

Male albino Wistar rats (150–250 g) were used in these experiments. They were housed in plastic cages in an animal room maintained at 22 ± 2°C on a 12 L:12 D cycle. Food and water were available at all times except during the time of the experiments.

Requests for reprints should be addressed to M. R. Zarrindast, Department of Pharmacology, School of Medicine, Tehran University of Medical Sciences, P. O. Box 13145-784, Tehran, Iran.

Behavioral Observation

The rats were placed individually in a glass cylinder (25 cm wide, 25 cm high) and a mirror was arranged in an oblique position under the cylinder to make recording of the licking possible. Animals were allowed to adapt for 30 min before the first drug injection. No more than two rats were observed simultaneously. Immediately after drug administration, the animals placed into the cylinder and the behavior was scored (protrusion of the tongue against the cylinder wall or floor) were recorded by a direct inspection. The subject's behavior was sampled at the end of each 60 s for a period of 60 min, and the licking was scored as "present" or "absent." The results are expressed as the mean ± SEM.

Drugs

The following drugs were used: apomorphine hydrochloride (MacFarlan Smith, Ltd, UK), phenoxybenzamine HCL (Smith-Kline & French Labs, Philadelphia, PA), propranolol (ICI, UK), phenylephrine hydrochloride, prazosin hydrochloride, clonidine hydrochloride, and yohimbine (Sigma, Poole, UK), dobutamine (Lilly, Germany), salbutamol, and atenolol (Darupakhsh, Iran). The drugs were given intraperitoneally and in a volume of 5 ml/kg, and were prepared immediately before use. The control groups received saline. Apomorphine was injected 15 min after adrenoceptor agonists and antagonists.

Statistical Analysis

One-way analysis of variance (ANOVA) followed by Newman–Keuls test was used for statistical analysis. Differences with $p < 0.05$ were considered statistically significant.

RESULTS

Apomorphine-Induced Licking in Rats

Figure 1 shows effect of apomorphine on licking behavior in rats. Intraperitoneal (IP) injection of different doses of apomorphine produced licking behavior dose dependently, $F(4, 40) = 42.6$, $p < 0.0001$. The maximum response was obtained with 0.5 mg/kg of the drug. The dose of 0.5 mg/kg of apomorphine was chosen for the rest of experiments.

Effect Adrenoceptor Agonists and Antagonists on Apomorphine-Induced Licking

Effects of phenylephrine on apomorphine-induced licking are shown in Fig. 2.

Pretreatment of the animals with different doses of α_1-adrenoceptor agonist, phenylephrine (1, 2, 4, 6, and 8 mg/kg IP), 15 min prior to apomorphine (0.5 mg/kg IP) reduced licking induced by apomorphine, $F(5, 48) = 16.11$, $p < 0.0001$. The α_2-adrenoceptor agonist clonidine (0.025, 0.05, and 0.1 mg/kg IP) did not alter licking induced by apomorphine 0.5 mg/kg, $F(3, 32) = 2.55$, $p > 0.05$, or 0.25 mg/kg, $F(3, 20) = 0.44$, $p > 0.05$ (data not shown).

Effects of α-adrenoceptor antagonists on apomorphine-induced licking are shown in Fig. 3.

ANOVA indicates a significant difference between animals that were treated with apomorphine (0.5 mg/kg IP) alone and those treated (IP) with apomorphine in the presence of prazosin (0.5, 1, and 2 mg/kg, $F(3, 32) = 5.59$, $p < 0.05$, yohimbine (0.25, 0.5, 1, and 2 mg/kg), $F(4, 40) = 6.61$, $p < 0.001$, phenoxybenzamine (2.5, 5, and 10 mg/kg or $F(3, 32) = 18.56$, $p < 0.0001$. Further analysis showed that prazosin,

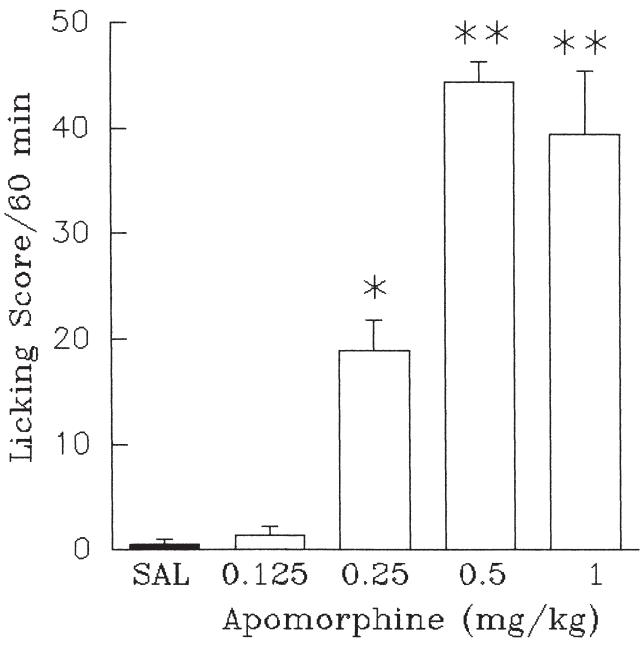

FIG. 1. Rats were treated intraperitoneally (IP) either with saline (5 ml/kg) or with different doses of apomorphine (0.125, 0.25, 0.5, and 1 mg/kg). Licking behavior was scored each 60 s for a period of 60 min after the drug injection. Each point is the mean ± SEM of nine animals. *$p < 0.01$, **$p < 0.001$ different from the saline control group.

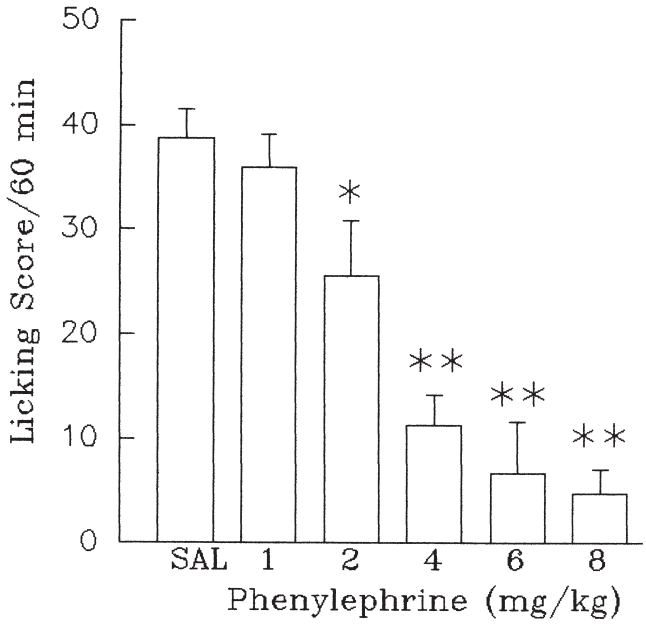

FIG. 2. Animals were treated intraperitoneally (IP) with saline (5 ml/kg) or different doses of phenylephrine (1–8 mg/kg) 15 min before apomorphine (0.5 mg/kg, IP) administration. Licking was scored each 60 s for 60 min after apomorphine injection. Each point is the mean ± SEM of nine animals. *$p < 0.05$, **$p < 0.001$ different from the control group.

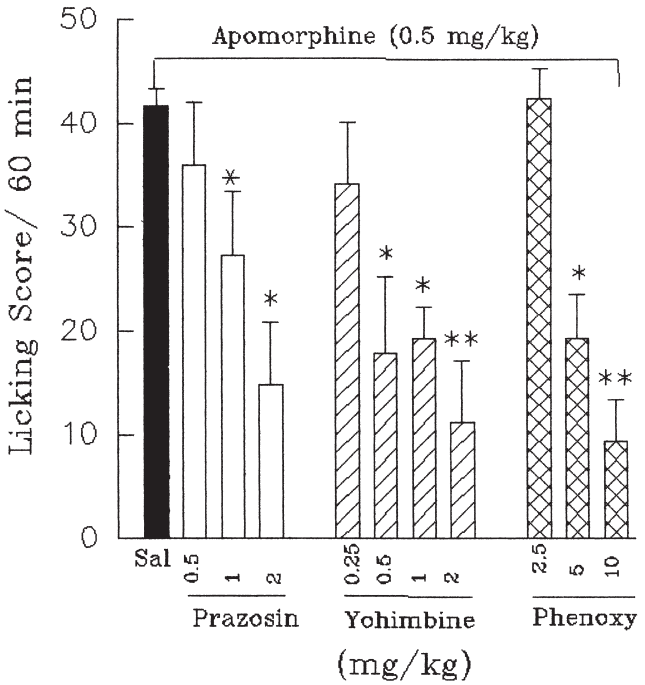

FIG. 3. Rats were treated intraperitoneally (IP) with either saline (5 ml/kg), prazosin (0.125–2 mg/kg), yohimbine (0.25–1 mg/kg), or phenoxybenzamine (Phenoxy; 2.5, 5, and 10 mg/kg) 15 min before apomorphine (0.5 mg/kg, IP) administration. Licking score was recorded for a period of 60 min after apomorphine injection. Each point is mean ± SEM of nine animals. $*p < 0.05$, $**p < 0.01$ different from respective control groups.

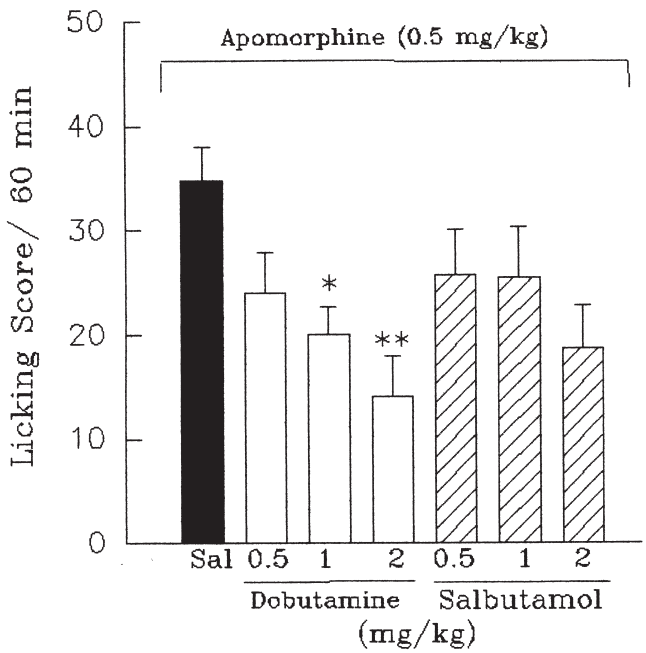

FIG. 4. Animals were treated intraperitoneally (IP) with either saline (5 ml/kg), atenolol (0.5–2 mg/kg), or propranolol (2.5–10 mg/kg) 15 min before apomorphine (0.5 mg/kg, IP) administration. Licking score was recorded for a period of 60 min after apomorphine injection. Each point is mean ± SEM of nine animals. $*p < 0.05$, $**p < 0.01$ different from respective control groups.

yohimbine, and phenoxybenzamine reduced the apomorphine effect on licking behavior.

Effects of prazosin on apomorphine-induced licking in the presence or absence of phenylephrine are shown in Table 1.

ANOVA showed that there is a significant difference between data obtained with apomorphine in the presence or absence of prazosin, $F(3, 32) = 5.9, p < 0.01$. There is also a difference in response to apomorphine plus phenylephrine in the presence or absence of prazosin, $F(3, 32) = 5.01, p < 0.01$.

TABLE 1

EFFECT OF PRAZOSIN ON APOMORPHINE-INDUCED LICKING IN THE PRESENCE OR ABSENCE OF PHENYLEPHRINE

Treatment (mg/kg)	Apomorphine 0.5	Apomorphine + phenylephrine 4
Saline 5 ml/kg	38.4 ± 3.2	11.3 ± 2.9
Prazosin 0.5	36.1 ± 5.9	26.1 ± 5.2
Prazosin 1	17.4 ± 4.7*	40.1 ± 3†
Prazosin 2	14.8 ± 6.0*	18.4 ± 6.8

Animals were treated intraperitoneally (IP) with either saline (5 ml/kg), or prazosin (0.5–2 mg/kg), or prazosin plus phenylephrine (4 mg/kg) 15 min before apomorphine administration. Licking score was recorded for a period of 60 min after apomorphine injection. Each point is mean ± SEM of nine animals.

$*p < 0.05$, $†p < 0.01$ different from respective control groups.

Further analysis indicates that prazosin reduced a response to apomorphine. The antagonist also reduced the inhibitory effect of phenylephrine.

Effect of β*-Adrenoceptor Agonists and Antagonists on Apomorphine-Induced Licking*

Effect of β-adrenoceptor agonists are shown in Fig. 4. ANOVA indicates that pretreatment of the animals with different doses of the β_1-adrenoceptor agonist dobutamine (0.5, 1, and 2 mg/kg, IP) 15 min prior to apomorphine (0.5 mg/kg IP), $F(3, 32) = 6.5, p < 0.01$, but not β-$_2$adrenoceptor salbutamol (0.5, 1, and 2 mg/kg, IP), $F(3, 32) = 2/5, p > 0.05$, reduced licking induced by apomorphine.

Effect of β-adrenoceptor antagonists have been shown in Fig. 5. ANOVA showed a difference between response of apomorphine (0.5 mg/kg IP) and those of apomorphine plus atenolol (0.5, 1, and 2 mg/kg), $F(3, 32) = 7.9, p < 0.001$, or plus propranolol (2.5, 5, and 10 mg/kg), $F(3, 32) = 4.5, p < 0.01$. Further analysis showed that both atenolol and propranolol reduced the apomorphine effect.

DISCUSSION

Dopamine appears to play a role in stereotyped behaviors. There is evidence indicating that dopamine (11) and dopamine agents (3) produce behavioral effects through multiple population of dopamine receptors located in different brain structures. Two types of receptors, D_1 and D_2, were distinguished on a pharmacological and biochemical basis. D_1 stimulates adenylate cyclase, while D_2 inhibits it (11,19). Gene cloning studies have split these into further subgroups (7,18). The D_1 family now includes D_1 and D_5, while the D_2 family

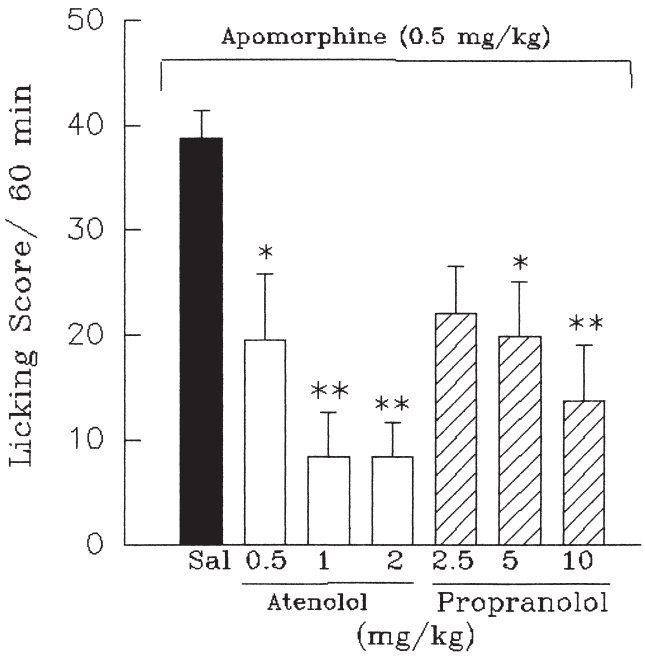

FIG. 5. Animals were treated intraperitoneally (IP) with either saline (5 ml/kg), dobutamine (0.5–2 mg/kg), or salbutamol (0.5–2 mg/kg) 15 min before apomorphine (0.5 mg/kg, IP) administration. Licking score was recorded for a period of 60 min after apomorphine injection. Each point is mean ± SEM of nine animals. $*p < 0.05$, $**p < 0.01$ different from respective control groups.

has been split into D_2, D_3, and D_4. The dopamine D_1 and D_2 receptors are needed to induce motor behavior (2) and licking behavior in rats (28,30).

The present data indicate that administration of different doses of apomorphine induced licking episodes in rats. The response was dose dependent. This is in agreement with previous work that D_1/D_2 dopamine agonist apomorphine (17) induced licking through activation of dopamine receptors (28,30).

In this study, we explored the influences of adrenergic agents on apomorphine-induced licking. The present data indicate that administration of the α_1-adrenoceptor phenylephrine, but not the α_2-adrenoceptor clonidine, decreased apomorphine-induced licking. Therefore, it can be suggested that the α_1-adrenergic mechanism may have an interaction with the present response. This may also be in agreement with other reports that adrenoceptor mechanisms may have influence on some other behaviors in the rat (4). It has been reported that there is an interaction between α_1-adrenoceptors and dopamine D_2 receptors in normal and catecholaminergic-depleted mice (6,14).

Although, the effect induced by phenylephrine was antagonized by the α_1-adrenoceptor antagonist prazosin, the highest dose of prazosin also reduced licking behavior produced by apomorphine. There are some reports indicating that prazosin decreased the behavioral effects produced by a systemic injection of apomorphine (13,20,21). This finding with the highest dose of prazosin may agree with a previous report that the α_1-antagonist, prazosin attenuates sniffing and locomotion (4), two behaviors that are induced by dopamine agonists (23,25). The reason why the highest dose of prazosin cannot reduce the response of all doses of the phenylephrine response also may be due to this inhibitory effect of the antagonist.

Stereotyped behaviors (e.g., head movement, licking, and localized sniffing) are more closely associated with the caudate striatum (12). Considering evidence for the role of D_1 and D_2 dopamine receptors in mediating apomorphine-induced licking behavior (28), and evidence that indicates an interaction between α_1-adrenoceptors and dopamine D_2 receptors in normal and catecholaminergic-depleted mice (6,14), a possible mechanism could involve a direct action of the α_1-adrenoceptor mechanism with postsynaptic D_2 receptor sites located in the striatum.

There is some evidence for an interaction between α_2-adrenoceptors and dopamine systems (4). However, the α_2-adrenoceptor agonist clonidine did not alter the apomorphine response in the present study. Therefore, direct interaction between α_2-adrenoceptor mechanisms in apomorphine-induced licking seems unlikely. The α_2-adrenoceptor antagonist yohimbine also reduced licking induced by apomorphine. The α_2-antagonist has been shown to inhibit stereotyped behavior (8,20,31), which may be mediated through a direct inhibition of dopamine receptors (15,16). Nevertheless, yohimbine may block α_2 presynaptic adrenoceptors and, in turn, may release noradrenaline, which may cause inhibition of apomorphine's response through activation of the α_1-adrenoceptor mechanism. The reason why the α_2-adrenoceptor agonist clonidine did not induce opposite effects may indicate that the central adrenergic system through adrenergic receptors does not have a negative influence on apomorphine-induced licking.

The involvement of β-adrenoceptors in licking behavior is not clear. The present study showed that the apomorphine-induced licking was reduced by atenolol and propranolol. This may indicate that even β-adrenoceptors are involved in the behavior. However, the β-adrenoceptor agonists dobutamine and salbutamol did not alter apomorphine response. Various interactions between dopamine and 5-HT systems have been demonstrated (10). Because both yohimbine and propranolol bind to 5-HT_{1A} receptors, the possible involvement of 5-HT mechanism(s) needs to be examined.

It is concluded that adrenoceptor mechanism(s) may be involved in licking behavior. However, more experiments are needed to clarify the exact mechanism(s).

REFERENCES

1. Anden, N. E.; Rubensson, A.; Fuxe, K.; Hokfelt, T.: Evidence of dopamine receptor stimulation by apomorphine. J. Pharm. Pharmacol. 19:627–629; 1967.
2. Braun, A.; Chase, T. N.: Obligatory D-1/D-2 receptor interaction in the generation of dopamine agonist related behaviours. Eur. J. Pharmacol. 123:109–114; 1986.
3. Cools, A. R.; Van Rossum, J. M.: Multiple receptors for brain dopamine in behaviour regulation: Concept of dopamine-E Dopamine-I receptors. Life Sci. 27:1237–1253; 1980.
4. Dickinson, S. I.; Brian, G.; Tulloch, I. F.: α_1- and α_2-adrenoceptor antagonists differentially influence locomotor and stereotyped behaviour induced by *d*-amphetamine and apomorphine in the rat. Psychopharmacology (Berlin) 96:522–527; 1988.
5. Ernst, A. M.: Mode of action of dopamine and dexamethasone in

gnawing compulsion in rats. Psychopharmacology (Berlin) 10:316–323; 1967.
6. Eshel, G.; Ross, S. B.; Kelder, D.; Edis, L. E. M.; Jackson, D. M.: α_1(but not α_2)-adrenoceptor agonist in combination with the dopamine D_2 agonist quinpirole produce locomotor stimulation in dopamine-depleted mice. Pharmacol. Toxicol. 67:123–131; 1990.
7. Givelli, O.; Bunzow, J. R.; Grandy, D. K.: Molecular diversity of the dopamine receptors. Annu. Rev. Pharmacol. Toxicol. 32:281–307; 1993.
8. Grabowska-Anden, M.: Modification of the amphetamine-induced stereotypy in rats following inhibition of the noradrenaline release by FLA136. J. Pharm. Pharmacol. 29:566–567; 1977.
9. Jackson, D. M.; Anden, N. E.; Dahlstrom, A.: A functional effect of dopamine in the nucleus accumbens and in some other dopamine-rich parts of the rat brain. Psycopharmacology (Berlin) 5:139–149; 1975.
10. Jackson, D. M.; Westlind-Danielsson, A.: Dopamine receptors: Molecular biology, biochemistry and behavioural aspects. Pharmacol. Ther. 64:291–369; 1994.
11. Kebabian, J. W.; Calne, D. B.: Multiple receptors for dopamine. Nature 277:93–96; 1979.
12. Kelly, P. H.; Seviour, P. W.; Iverson, S. D.: Amphetamine and apomorphine responses in the rat following 6-OHDA lesions of the nucleus accumbens, septi and corpus striatum. Brain Res. 94:507–522; 1975.
13. Mogilnicka, E.; Braestrup, C.: Noradrenergic influence on the stereotyped behaviour induced by amphetamine, phenethylamine and apomorphine. J. Pharm. Pharmacol. 28:253–255; 1976.
14. Rubinstein, M.; Schinder, A. F.; Gershanik, O.; Stefano, F. J. D.: Positive interaction between alpha-1 adrenergic and dopamine-2 receptors in locomotor activity of normo and supersensitive mice. Life Sci. 44:337–346; 1989.
15. Scatton, B.; Zivkovic, B.; Dedek, J.: Antidopaminergic properties of yohimbine. J. Pharmacol. Exp. Ther. 215:494–499; 1980.
16. Scatton, B.; Dedek, J.; Zivkovic, B.: Lack of involvement of α_2-adrenoceptors in the regulation of striatal dopaminergic transmission. Eur. J. Pharmacol. 86:427–433; 1983.
17. Seeman, P.: Brain dopamine receptors. Pharmacol. Rev. 32:229–313; 1980.
18. Sibley, D. R.; Monsma, F. J.: Molecular biology of dopamine receptors. Trends Pharmacol. Sci. 13:61–69; 1992.
19. Stoof, J. C.; Kebabian, J. W.: Two dopamine receptors: Biochemistry, physiology and pharmacology. Life Sci. 35:2281–2296; 1984.
20. Thomas, K. V.; Handley, S. L.: Modulation of dexamphetamine-induced compulsive gnawing-including the possible involvement of presynaptic α-adrenoceptors. Psychopharmacology (Berlin) 56:61–67; 1978.
21. Wiszniowska-Szafraniec, G.; Danek, L.; Reichenberg, K.; Vetulani, J.: Facilitation by α-adrenolytics of apomorphine gnawing behaviour: Depression of threshold apomorphine concentration in the striatum of the rat. Pharmacol. Biochem. Behav. 19:19–21; 1983.
22. Zarrindast, M. R.; Amin, R.: Role of D_1 and D_2 receptors in apomorphine-induced pecking in chicks. Psychopharmacology (Berlin) 106:67–70; 1992.
23. Zarrindast, M. R.; Eliassi, A.: Differential effects of dopamine agonists on locomotion in intact and reserpine-treated mice. Gen. Pharmacol. 22:1027–1031; 1991.
24. Zarrindast, M. R.; Hajian-Heydari, A.; Hoseini-Nia, T.: Characterization of dopamine receptors involved in apomorphine-induced pecking in pigeons. Gen. Pharmacol. 23:427–430; 1992.
25. Zarrindast, M. R.; Naghashi, H.: Bromocriptine requires D-1 receptor stimulation for the expression of sniffing behaviour in rats. J. Psychopharmacol. 5:160–165; 1991.
26. Zarrindast, M. R.; Moini-Zanjani, T.; Manaheji, H.; Fathi, F.: Influences of dopamine receptors on chewing behaviour in rats. Gen. Pharmacol. 23:915–919; 1992.
27. Zarrindast, M. R.; Poursoltan, M.: Interactions of drugs acting on central dopamine receptors and cholinoceptors on yawning responses in the rat induced by apomorphine, bromocriptine or physostigmine. Br. J. Pharmacol. 96:843–848; 1989.
28. Zarrindast, M. R.; Roushan-Zamir, F.; Amir-Rahmat, F.: Potentiation of licking in rats by stimulation of both D-1 and D-2 dopamine receptors. J. Psychopharmacol. 6:395–398; 1992.
29. Zarrindast, M. R.; Farahvash, H.: Effects of GABA-ergic drugs on penile erection induced by apomorphine in rats. Psychopharmacology (Berlin) 115:249–253; 1994.
30. Zarrindast, M. R.; Sharifzadeh, M.: Effects of adenosine drugs on apomorphine-induced licking in rats. Gen. Pharmacol. 26:1119–1123; 1995.
31. Zetler, G.: Clonidine sensitizes mice for apomorphine-induced stereotypic gnawing. Antagonism by neuroleptics and cholecystokinin-like peptides. Eur. J. Pharmacol. 111:309–315; 1985.