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Effects of selective activation of dopamine D_2 and D_3 receptors on prolactin secretion and the activity of tuberoinfundibular dopamine neurons

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Abstract

Dopamine agonists with activity at both dopamine D_2 and D_3 receptor subtypes stimulate tuberoinfundibular dopamine neurons and inhibit prolactin secretion from the anterior pituitary. The purpose of the present study was to identify the dopamine receptor subtypes mediating these effects using recently developed selective agonists for dopamine D_2 (PNU-95,666) and D_3 (PD128907) receptors. The activity of tuberoinfundibular dopamine neurons was estimated by measuring either the synthesis (accumulation of 3,4-dihydroxyphenylalanine [DOPA] following inhibition of decarboxylase activity) or metabolism (3,4-dihydroxyphenylacetic acid [DOPAC] concentrations) of dopamine in the median eminence, the region of the hypothalamus containing axon terminals of these neurons. In one experiment, the activity of mesolimbic dopamine neurons was also determined by measuring DOPA accumulation in terminals of these neurons in the nucleus accumbens. Activation of dopamine D_2 receptors with PNU-95,666 caused dose- and time-related increases in DOPAC concentrations in median eminence which were temporally correlated with decreases in plasma prolactin concentrations. Activation of dopamine D_3 receptors with PD128907 decreased DOPA concentrations in the nucleus accumbens, but had no effect on concentrations of DOPAC or DOPA in the median eminence or prolactin in plasma. These results reveal that tuberoinfundibular dopamine neurons are regulated by dopamine D_2 rather than D_3 receptors, and suggest that the ability of mixed dopamine D_2/D_3 receptor agonists to increase the activity of these neurons is mediated by an action at dopamine D_2 receptors. Furthermore, these results confirm that tuberoinfundibular dopamine neurons are not regulated by inhibitory dopamine D_2 or D_3 autoreceptors. © 1997 Elsevier Science B.V.

Keywords: Dopamine receptor agonist; PNU-95,666; PD128907; DOPAC (3,4-dihydroxyphenylal acetic acid); Median eminence

1. Introduction

The major ascending nigrostriatal and mesolimbic dopamine neuronal systems in the brain are regulated by dopamine receptor-mediated mechanisms: i.e., dopamine agonists reduce, whereas dopamine receptor antagonists increase impulse flow and the neurochemical activities of these neurons (Bunney et al., 1987). These effects are chiefly mediated by inhibitory dopamine D_2 and/or D_3 autoreceptors located on neuronal perikarya, dendrites and/or axon terminals which function to maintain a homeostatic balance between impulse flow-induced neurotransmitter release and its replenishment by de novo synthesis (Elsworth and Roth, 1997). In contrast, hypothalamic tuberoinfundibular dopamine neurons lack dopamine autoreceptors, and are unresponsive to acute administration of non-selective dopamine receptor agonists (e.g. apomorphine and bromocriptine) and antagonists (e.g. haloperidol) (Moore, 1987). Rather, tuberoinfundibular dopamine neurons are regulated by prolactin secreted by anterior pituitary lactotrophs, and increases or decreases in circulating prolactin produce corresponding changes in the activity of these neurons. By virtue of their ability to alter prolactin secretion via an action on dopamine D_2 receptors in the anterior pituitary, non-selective dopamine receptor agonists and antagonists act indirectly to cause delayed alterations

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in the activity of tuberoinfundibular dopamine neurons (Moore and Lookingland, 1995).

Utilization of second generation agonists which differentiate between the D_1-like and D_2-like subtype families of dopamine receptors (Schwartz et al., 1993; Jackson and Westlind-Danielsson, 1994) has revealed that tuberoinfundibular dopamine neurons are also regulated by a dopamine receptor-mediated mechanism which acts independently of prolactin (Eaton et al., 1993). Indeed, acute administration of dopamine agonists with preferential affinity for the D_2-like family of dopamine receptors (i.e., quinpirole (Bymaster et al., 1986) and quinelorane (Foreman et al., 1989)) stimulates the activity of tuberoinfundibular dopamine neurons (Berry and Gudelsky, 1991; Eaton et al., 1993), even in the absence of prolactin (Eaton et al., 1993). This dopamine receptor-mediated activation of tuberoinfundibular dopamine neurons likely occurs via an afferent neuronal mechanism involving, in part, inhibition of tonically active inhibitory dynorphinergic interneurons (Durham et al., 1996). Since ergoline-derived dopamine agonists such as quinpirole and quinelorane bind with equal affinity to D_2 and D_3 subtypes of the dopamine D_2-like receptor family (Smalstig and Clemens, 1984; Gackenheimer et al., 1995), it is not known which of these receptors mediates stimulation of tuberoinfundibular dopamine neurons.

The purpose of the present study was to examine the effects of two novel dopamine receptor agonists with discriminative actions at dopamine D_2 and D_3 receptors on prolactin secretion and the activity of tuberoinfundibular dopamine neurons. To this end, the effects of the selective dopamine D_2 receptor agonist PNU-95,666 (Smith et al., 1995) and the selective dopamine D_3 receptor agonist PD128907 (Pugsley et al., 1995) were examined on either the synthesis (accumulation of 3,4-dihydroxyphenylalanine [DOPA] following inhibition of decarboxylase activity) or metabolism (3,4-dihydroxyphenylacetic acid [DOPAC] concentrations) of dopamine in the median eminence. These results reveal that tuberoinfundibular dopamine neurons are regulated by dopamine D_2 rather than D_3 receptors, and suggest that the ability of mixed dopamine $D_{2/3}$ receptor agonists to increase the activity of these neurons is mediated by an action at dopamine D_2 receptors.

2. Materials and methods

2.1. Animals

Male Long-Evans rats weighing 200–225 g were obtained from Harlan Laboratories (Indianapolis, IN, USA), housed in a temperature- (72 ± 2°F) and light- (lights on between 05.00–19.00 h) controlled room, and provided food (Wayne Lablox) and tap water ad libitum.

2.2. Drug preparation and administration

(*R*)-5,6-dihydro-5-(methylamino)-4H-imidazo[4,5,1-ij]quinolin-2 (1H)-one (*Z*)-2-butenedioate maleate (PNU-95,666; Dr. P. VonVoigtlander, Pharmacia and Upjohn, Kalamazoo, MI, USA), and 3-hydroxybenzylhydrazine dihydrochloride (NSD 1015; Sigma, St. Louis, MO, USA) were dissolved in 0.9% saline. *R*-(+)-*trans*-3,4,4a,10b-tetrahydro-4-propyl-2H,5H-[1] benzopyrano[4,3-b]-1,4-oxazin-9-ol hydrochloride (PD-128907; Dr. M.D. Davis, Parke Davis, Ann Arbor, MI, USA) was dissolved in distilled water. Drugs were administered as indicated in the legends of the appropriate figures; doses of drugs were calculated as the respective salts.

2.3. Tissue dissection and biochemical determination of monoamines

Following drug treatments, rats were decapitated and brains were rapidly removed from the skull and frozen on aluminum foil placed over dry ice. Frontal brain sections (600 μm) beginning approximately at 9220 μm (König and Klippel, 1963) were prepared in a cryostat (−9°C), and the median eminence and nucleus accumbens were dissected from appropriate sections according to a modification (Lookingland and Moore, 1984) of the method of Palkovits (1973). Tissue samples were placed in 65 μl (median eminence) or 100 μl (nucleus accumbens) of 0.1 M phosphate–citrate buffer (pH 2.5) containing 15% methanol and stored at −20°C until assayed.

On the day of the assay, tissue samples were thawed, sonicated for 3 s (Sonicator Cell Disruptor, Heat Systems-Ultrasonic, Plainview, NY, USA) and centrifuged for 30 s in a Beckman 152 Microfuge. Contents of dopamine, DOPAC and DOPA in supernatants were determined by high-performance liquid chromatography coupled with electrochemical detection as described previously (Lindley et al., 1990). The amounts of these compounds in tissue samples were determined by comparing peak heights (as determined by a Hewlett Packard Integrator, Model 3395) with those obtained from external standards run on the same day. The lower limit of sensitivity of this assay for these compounds was approximately 1 pg per sample. Tissue pellets were dissolved in 1.0 M NaOH and assayed for protein (Lowry et al., 1951).

2.4. Neurochemical estimation of dopamine neuronal activity

The activities of tuberoinfundibular and mesolimbic dopamine neurons were estimated by determining dopamine synthesis or metabolism in axon terminals of these neurons in the median eminence and nucleus accumbens, respectively. Dopamine synthesis was determined by

measuring the accumulation of DOPA 30 min after administration of the decarboxylase inhibitor NSD 1015 (100 mg/kg, i.p.; Demarest and Moore, 1980), whereas dopamine metabolism was determined by measuring DOPAC concentrations in the absence of any drug pretreatment (Lookingland et al., 1987). Due to the tight coupling between the release and metabolism of dopamine and its replenishment by de novo synthesis, tissue concentrations of DOPAC and DOPA have been shown to reflect the activity of tuberoinfundibular (Demarest and Moore, 1980; Lookingland et al., 1987) and mesolimbic dopamine neurons (Roth et al., 1976; Demarest and Moore, 1979).

2.5. Radioimmunoassay for plasma prolactin

Trunk blood collected following decapitation was centrifuged for 20 min at 4°C. The plasma was drawn off and stored at −20°C for later determination of prolactin concentrations by double-antibody radioimmunoassay. Prolactin was measured using the reagents and procedures of the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) assay with rat prolactin (RP-3) as the standard (generously provided by Dr. A.F. Parlow and Dr. S. Raiti, NIDDK National Hormone and Pituitary Program). Using a 100 ml aliquot of plasma, the lower limit of sensitivity for prolactin was 120 pg/tube. The intra-assay coefficient of variation for prolactin was approximately 6%.

2.6. Statistical analyses

Statistical analyses were conducted using a one-way analysis of variance followed by Bonferroni *t*-test for multiple comparisons. Differences were considered significant if the probability of error was less than 5%.

3. Results

A hallmark of D_2 dopamine agonists is their ability to inhibit prolactin secretion via an action at D_2 dopamine receptors on anterior pituitary lactotrophs (Albert et al., 1997). As shown in Fig. 1, PNU-95,666 (10 mg/kg; s.c.) significantly reduced plasma prolactin concentrations by 30 min and this effect lasted for at least an additional 90 min. In contrast, PD128907 (300 μg/kg; i.p.) failed to alter plasma levels of prolactin at any of the times examined. These results are consistent with a direct agonist action of PNU-95,666 (but not PD128907) at D_2 dopamine receptors on lactotrophs in the anterior pituitary.

As shown in Fig. 2, PNU-95,666 caused a dose-dependent increase in DOPAC concentrations in the median eminence, whereas incremental pharmacologically-active doses of PD128907 (Pugsley et al., 1995) had no effect. A similar pattern emerged when time courses of the actions of these drugs were examined (Fig. 3). PNU-95,666 (10

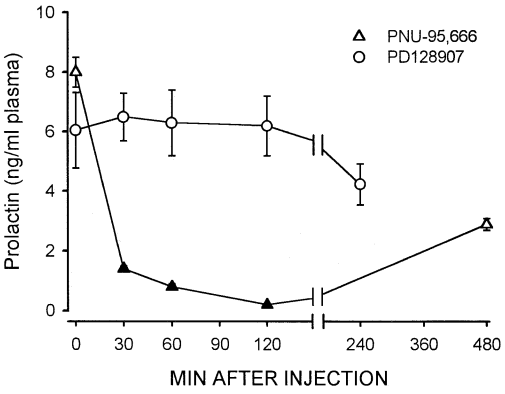

Fig. 1. Time course of the effects of PNU-95,666 and PD128907 on plasma prolactin concentrations. Rats were injected with either PNU-95,666 (10 mg/kg; s.c.) or PD128907 (300 μg/kg; i.p.) and decapitated either 30, 60, 120, 240 or 480 min later. Zero time controls were injected with vehicles for either PNU-95,666 (0.9% saline; 1 ml/kg; s.c.) or PD128907 (distilled water; 1 ml/kg; i.p.) 30 min prior to decapitation. Symbols represent means and vertical lines 1 S.E.M. of plasma prolactin concentrations in 5–7 rats; no vertical line is depicted when 1 S.E.M is less than the radius of the symbol. Solid symbols represent those values that are significantly different ($P < 0.05$) from zero time controls.

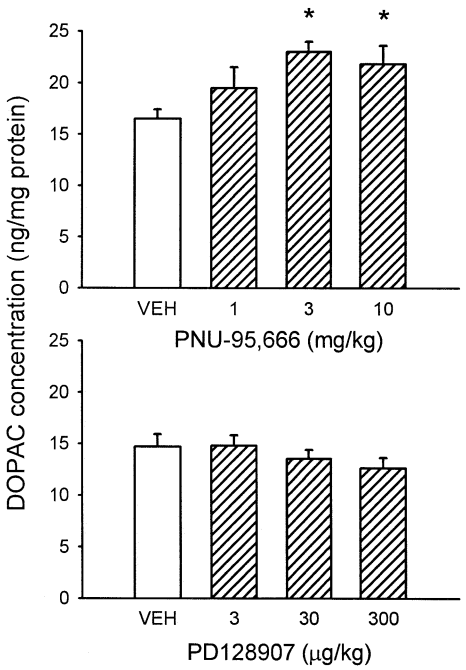

Fig. 2. Dose response effects of PNU-95,666 and PD128907 on DOPAC concentrations in the median eminence. (Top panel) Rats were injected with either PNU-95,666 (1, 3 or 10 mg/kg; s.c.) or its 0.9% saline vehicle (1 ml/kg; s.c.) and decapitated 30 min later. (Bottom panel) Rats were injected with either PD128907 (3, 30 or 300 μg/kg; i.p.) or its distilled water vehicle (1 ml/kg; i.p.) and decapitated 60 min later. Columns represent means and vertical lines 1 S.E.M. of 7–9 determinations of DOPAC concentrations in median eminence of vehicle- (VEH; open columns) or drug-treated (hatched columns) rats. * Values in drug-treated rats that are significantly different ($P < 0.05$) from vehicle-treated controls.

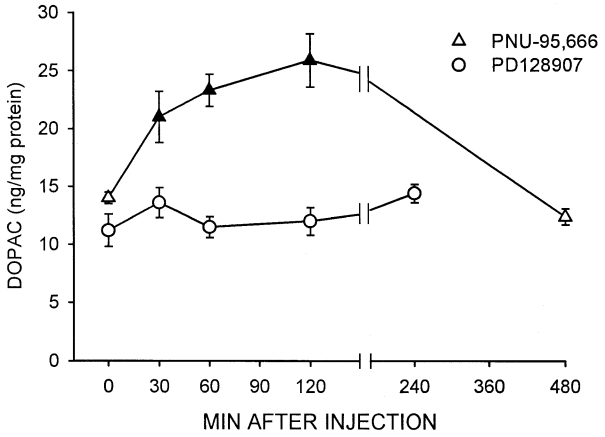

Fig. 3. Time course of the effects of PNU-95,666 and PD128907 on DOPAC concentrations in the median eminence. Rats were injected with either PNU-95,666 (10 mg/kg; s.c.) or PD128907 (300 μg/kg; i.p.) and decapitated either 30, 60, 120, 240 or 480 min later. Zero time controls were injected with vehicles for either PNU-95,666 (0.9% saline; 1 ml/kg; s.c.) or PD128907 (distilled water; 1 ml/kg; i.p.) 30 min prior to decapitation. Symbols represent means and vertical lines 1 S.E.M of DOPAC concentrations in the median eminence of 5–8 rats. Solid symbols represent those values that are significantly different ($P < 0.05$) from zero time controls.

mg/kg; s.c.) caused a prompt (within 30 min) and a sustained (for at least 120 min) increase in DOPAC concentrations in the median eminence, while PD128907 (300 μg/kg; i.p.) failed to alter median eminence DOPAC at any of the times examined.

Since PD128907 failed to alter concentrations of DOPAC in the median eminence it was imperative to demonstrate that this drug has dopamine D_3 receptor agonist properties at the doses employed in these studies. A previous report (Pugsley et al., 1995) demonstrated that PD128907 reduced dopamine synthesis in the terminals of mesolimbic dopamine neurons in the nucleus accumbens. Confirming this report, the results presented in Fig. 4 reveal that PD128907 (300 μg/kg; i.p.) reduced DOPA accumulation in the nucleus accumbens. Consistent with the results of studies in which DOPAC concentrations were used as an index of the activity of tuberoinfundibular dopamine neurons (Figs. 2 and 3), PD128907 failed to alter DOPA accumulation in the median eminence (Fig. 4).

4. Discussion

Central dopamine receptors were originally divided into two pharmacologically distinct subtypes on the basis of their biochemical effects on adenylyl cyclase activity (Kebabian and Calne, 1979). Dopamine D_1 receptors were linked to the stimulation of adenylyl cyclase; whereas dopamine D_2 receptors inhibited adenylyl cyclase. More recent binding and molecular cloning studies have established multiple variants of these receptors which form two separate families, the dopamine D_1-like and D_2-like receptors (Schwartz et al., 1993; Civelli et al., 1993). The D_1-like family is composed of two subtypes (D_1 and D_5) which are predominantly distributed as post-synaptic receptors (Richtand et al., 1995), whereas the D_2-like family consists of three subtypes (D_2, D_3 and D_4) which function both as pre- and post-synaptic receptors (Richtand et al., 1995). The discovery of multiple subtypes of the dopamine receptor led to the development of second generation agonists selective for dopamine D_1- and D_2-like receptors, including the substituted benzazepines (e.g. SKF 38393) with dopamine D_1-like receptor affinity and ergoline-derived compounds (e.g. quinpirole, quinelorane) with affinity for dopamine D_2-like receptors. More recently, a newer generation of compounds has been developed with utility for distinguishing among receptors comprising the dopamine D_2-like receptor family including the selective dopamine D_2 receptor agonist PNU-95,666 (Smith et al.,

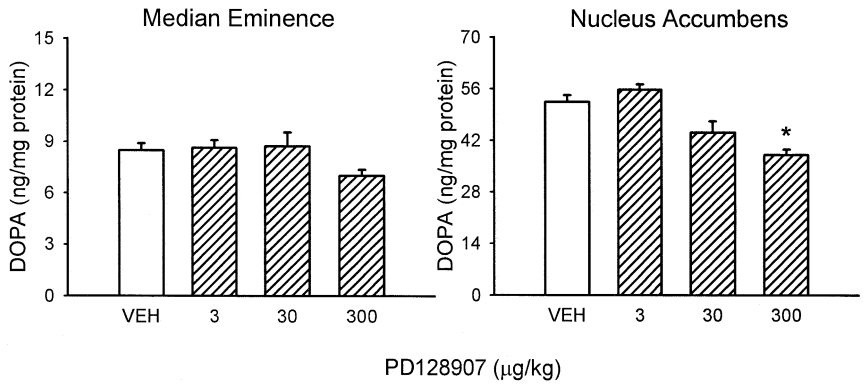

Fig. 4. Dose response effects of PD128907 on DOPA accumulation in the median eminence and nucleus accumbens. Rats were injected with either PD128907 (3, 30 or 300 μg/kg; i.p.) or its distilled water vehicle (1 ml/kg; i.p.) and decapitated 60 min later. All rats were injected with NSD 1015 (100 mg/kg; i.p.) 30 min prior to decapitation. Columns represent means and vertical lines 1 S.E.M. of 7–9 determinations of DOPA concentrations in median eminence of vehicle- (VEH; open columns) or drug-treated (hatched columns) rats. * Values in drug-treated rats that are significantly different ($P < 0.05$) from vehicle-treated controls.

1995) and the selective dopamine D_3 receptor agonist PD128907 (Pugsley et al., 1995).

The results of the present study verify the dopamine D_2 receptor agonist properties of PNU-95,666 by demonstrating time-dependent inhibitory effects of this compound on prolactin secretion. This effect is likely mediated by either short (D_{2S}) or long (D_{2L}) forms of D_2 dopamine receptors which utilize similar signaling mechanisms to decrease cAMP generation and Ca^{2+} influx into pituitary lactotrophs (Albert et al., 1997). On the other hand, PD128907 (which exhibits 18-fold selectivity for dopamine D_3 versus D_{2L} receptors; Pugsley et al., 1995) has no effect on prolactin secretion, demonstrating that when administered at a dose as high as 300 μg/kg this compound does not activate dopamine D_2 receptors. Taken together, these results indicate that PNU-95,666 and PD128907 may be used to discriminate between dopamine D_2 and D_3 receptors and identify the role of these D_2-like receptor subtypes in mediating central dopamine neuronal function.

Acute administration of the dopamine receptor agonist quinelorane increases the activity of tuberoinfundibular dopamine neurons (Eaton et al., 1993) by a mechanism involving either dopamine D_2 or D_3 receptors. The results from the present study suggest that these actions are mediated by dopamine D_2 rather than D_3 receptors. Indeed, acute administration of PNU-95,666 caused dose- and time-dependent increases in dopamine metabolism in the median eminence, whereas PD128907 was without effect, even at a dose (300 μg/kg) which decreased dopamine synthesis in the nucleus accumbens. These latter results reveal that tuberoinfundibular dopamine neurons are not regulated by synthesis-regulating dopamine D_3 autoreceptors like mesolimbic dopamine neurons (Aretha et al., 1995). In addition, the stimulatory (rather than inhibitory) effect of dopamine D_2 receptor activation on tuberoinfundibular dopamine neurons is consistent with the conclusion that these neurons also lack dopamine D_2 autoreceptors, and are not regulated by dopamine D_2 receptor-mediated afferent neuronal inhibition (Demarest and Moore, 1979; Durham et al., 1996). Rather, these results are consistent with the conclusion that tuberoinfundibular dopamine neurons are stimulated by a post-synaptic dopamine D_2 receptor-mediated mechanism, possibly involving afferent neuronal inhibition of tonically active inhibitory dynorphinergic neurons (Durham et al., 1996).

The finding that selective activation of dopamine D_2 receptors with PNU-95,666 stimulates the activity of tuberoinfundibular dopamine neurons raises the question as to why acute administration of non-selective dopamine agonists with dopamine D_2 receptor properties such as apomorphine or bromocriptine have no effect on the activity of these neurons (Demarest and Moore, 1979). This may be due to the ability of these agonists to also activate dopamine D_1 receptors since tuberoinfundibular dopamine neurons are inhibited following administration of selective dopamine D_1 receptor agonists (Berry and Gudelsky, 1990; Johnson et al., 1996). Evidently, non-selective dopamine agonists activate both stimulatory dopamine D_2 and inhibitory D_1 receptors which masks their individual effects on tuberoinfundibular dopamine neurons. The present results demonstrate the utility of selective dopamine receptor agonists in studies designed to discern the opposing roles of these dopamine subtype receptors in the regulation of tuberoinfundibular dopamine and other dopamine neuronal systems.

In conclusion, the results of the present study reveal that tuberoinfundibular dopamine neurons in the male rat are stimulated following activation of dopamine D_2 receptors, and confirm that these neurons are not regulated by inhibitory dopamine D_2 or D_3 autoreceptors. Furthermore, these results suggest that the ability of dopamine receptor agonists to inhibit mesolimbic dopamine neurons without affecting tuberoinfundibular dopamine neuronal activity or prolactin secretion may represent a characteristic in vivo response useful for discriminating dopamine D_2 versus D_3 receptor agonist activity.

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References

Albert, P.R., Ghahremani, M.H., Morris, S.J., 1997. Mechanisms of dopaminergic regulation of prolactin secretion. In: Neve, K.A., Neve, R.L. (Eds.), The Dopamine Receptors. Humana Press, Totowa, NJ, pp. 359–381.

Aretha, C.W., Sinha, A., Galloway, M.P., 1995. Dopamine D3-preferring ligands act at synthesis modulating autoreceptors. J. Pharmacol. Exp. Ther. 274, 609–613.

Berry, S.A., Gudelsky, G.A., 1990. D_1 receptors function to inhibit the activation of tuberoinfundibular dopamine neurons. J. Pharmacol. Exp. Ther. 254, 677–682.

Berry, S.A., Gudelsky, G.A., 1991. Effect of D_2 dopamine agonists on tuberoinfundibular dopamine neurons. Neuropharmacology 30, 961–965.

Bunney, B.S., Sesack, S.R., Silva, N.L., 1987. Midbrain dopaminergic systems: Neurophysiology and electrophysiological pharmacology. In: Meltzer, H.Y. (Ed.), Psychopharmacology: The Third Generation of Progress. Raven Press, New York, NY, pp. 113–126.

Bymaster, F., Reid, L., Nichols, C., Kornfeld, E., Wong, D., 1986. Elevations of acetylcholine levels in striatum of rats by LY 163502, *trans*-(−)-5,5a,6,7,8,9a,10-octahydro-6-propyl-pyrimido < 4,5-g > quinolin-2-aminedihydrochloride, a potent and stereospecific dopamine D_2 agonist. Life Sci. 38, 317–322.

Civelli, O., Bunzow, J.R., Grandy, D.K., 1993. Molecular diversity of the dopamine receptors. Annu. Rev. Pharmacol. Toxicol. 33, 281–307.

Demarest, K.T., Moore, K.E., 1979. Comparison of dopamine synthesis regulation in the terminals of nigrostriatal, mesolimbic, tuberoinfundibular and tuberohypophyseal neurons. J. Neural. Trans. 46, 263–277.

Demarest, K.T., Moore, K.E., 1980. Accumulation of L-Dopa in the median eminence: An index of tuberoinfundibular dopaminergic nerve activity. Endocrinology 106, 463–468.

Durham, R.A., Johnson, J.D., Moore, K.E., Lookingland, K.J., 1996. Evidence that D_2 receptor-mediated activation of hypothalamic tuberoinfundibular dopaminergic neurons in the male rat occurs via inhibition of tonically active afferent dynorphinergic neurons. Brain Res. 732, 113–120.

Eaton, M.J., Gopalan, C., Kim, E.J., Lookingland, K.J., Moore, K.E., 1993. Comparison of the effects of the dopamine D_2 agonist quinelorane on tuberoinfundibular dopaminergic neuronal activity in male and female rats. Brain Res. 629, 53–58.

Elsworth, J.D., Roth, R.H., 1997. Dopamine autoreceptor pharmacology and function. In: Neve, K.A., Neve, R.L. (Eds.), The Dopamine Receptors. Humana Press, Totowa, NJ, pp. 233–265.

Foreman, M.M., Fuller, R.W., Hynes, M.D., Gidda, J.S., Nichols, C.L., Schaus, J.M., Kornfeld, E.C., Clemens, J.A., 1989. Preclinical studies on quinelorane, a potent and highly selective D_2-dopaminergic agonist. J. Pharmacol. Exp. Ther. 250, 227–235.

Gackenheimer, S.L., Schaus, J.M., Gehlert, D.R., 1995. [^{3}H]-Quinelorane binds to D_2 and D_3 dopamine receptors in the rat brain. J. Pharmacol. Exp. Ther. 274, 1558–1565.

Jackson, D.M., Westlind-Danielsson, A., 1994. Dopamine receptors: Molecular biology, biochemistry and behavioural aspects. Pharmacol. Ther. 64, 291–369.

Johnson, J.D., Durham, R.A., Moore, K.E., Lookingland, K.E., 1996. Opposing roles for D_1 and D_2 receptors in the regulation of tuberoinfundibular dopamine neurons in male rats. 26th Annual Meeting of the Society for Neuroscience, Washington, DC, Abstract No. 696.9.

Kebabian, J.W., Calne, D.B., 1979. Multiple receptors for dopamine. Nature 277, 93–96.

König, J.F.R., Klippel, R.A., 1963. The Rat Brain: A Stereotaxic Atlas of the Forebrain and Lower Parts of the Brainstem. Krieger, Huntington, NY.

Lindley, S.E., Gunnet, J.W., Lookingland, K.J., Moore, K.E., 1990. 3,4-dihydroxyphenylacetic acid concentrations in the intermediate lobe and neural lobe of the posterior pituitary gland as an index of tuberohypophysial dopaminergic neuronal activity. Brain Res. 506, 133–138.

Lookingland, K.J., Moore, K.E., 1984. Dopamine receptor-mediated regulation of incertohypothalamic dopaminergic neurons in the male rat. Brain Res. 304, 329–338.

Lookingland, K.J., Jarry, H.D., Moore, K.E., 1987. The metabolism of dopamine in the median eminence reflects the activity of tuberoinfundibular dopamine neurons. Brain Res. 419, 303–310.

Lowry, O.H., Rosebrough, N.J., Farr, A.L., Randall, R.J., 1951. Protein measurement with the Folin phenol reagent. J. Biol. Chem. 193, 265–275.

Moore, K.E., 1987. Hypothalamic dopaminergic neuronal systems. In: Meltzer, H.Y. (Ed.), Psychopharmacology: The Third Generation of Progress. Raven Press, New York, NY, pp. 127–140.

Moore, K.E., Lookingland, K.E., 1995. Dopaminergic neuronal systems in the hypothalamus. In: Bloom, F.E., Kupfer, D.J. (Eds.), Psychopharmacology: The Fourth Generation of Progress. Raven Press, New York, NY, pp. 245–256.

Palkovits, M., 1973. Isolated removal of hypothalamic or other brain nuclei in the rat. Brain Res. 59, 449–450.

Pugsley, T.A., Davis, M.D., Akunne, H.C., Mackenzie, R.G., Shih, Y.H., Damsma, G., Wikstrom, H., Whetzel, S.Z., Georgic, L.M., Cooke, L.W., Demattos, S.B., Corbin, A.E., Glase, S.A., Wise, L.D., Dijkstra, D., Heffner, T.G., 1995. Neurochemical and functional characterization of the preferentially selective dopamine D3 agonist PD 128907. J. Pharmacol. Exp. Ther. 275, 1355–1366.

Richtand, N.M., Kelsoe, J.R., Segal, D.S., Kuczenski, 1995. Regional quantification of D1, D2, and D3 dopamine receptor mRNA in rat brain using a ribonuclease protection assay. Mol. Brain Res. 33, 97–103.

Roth, R.H., Murrin, L.C., Walters, J.R., 1976. Central dopaminergic neurons: Effects of alterations in impulse flow in the accumulation of dihydroxyphenylacetic acid. Eur. J. Pharmacol. 36, 163–172.

Schwartz, J.-C., Levesque, D., Martres, M.-P., Sokoloff, P., 1993. Dopamine D3 receptor: Basis and clinical aspects. Clin. Neuropharmacol. 16, 295–314.

Smalstig, B., Clemens, J., 1984. Dopamine agonist activity of LY 163502, a potent 'partial' ergoline. Pharmacologist 26, 136.

Smith, M.W., Bienkowski, M.J., Darlington, W.H., Dinh, D.M., Harris, D.W., Huff, R.M., Lajiness, M.E., Lawson, C.F., McCall, R.B., McGuire, J.C., Moon, M.W., Schlachter, S.K., 1995. Monoaminergic receptor profiles of imidazoquinolinones: Agonists potentially useful for the treatment of parkinson's disease. 25th Annual Meeting of the Society for Neuroscience, San Diego, CA, Abstract No. 340.8.