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In vivo effects of aripiprazole on cortical and striatal dopaminergic and serotonergic function

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Abstract

In vivo microdialysis was used to monitor the effects of oral aripiprazole and olanzapine on basal extracellular concentrations of dopamine, 3,4-dihydroxyphenylacetic acid (DOPAC), homovanillic acid (HVA) and 5-hydroxyindole acetic acid (5-HIAA) in the medial prefrontal cortex and striatum of conscious, freely moving rats. Acute aripiprazole administration did not affect dopamine output, but produced moderate increases in DOPAC and HVA concentrations, in medial prefrontal cortex or striatum of drug-naïve rats. Similarly, aripiprazole did not affect dopamine output but produced moderate elevations in DOPAC and HVA concentrations in the striatum of chronic aripiprazole-pretreated rats. Olanzapine produced comparatively larger elevations in dopamine, DOPAC, and HVA in both regions, which, in the striatum, were diminished after chronic olanzapine exposure. Aripiprazole reduced extracellular 5-HIAA concentrations in the medial prefrontal cortex and striatum of drug-naïve rats, but not in chronic aripiprazole-pretreated rats. Together, these data provide in vivo evidence of aripiprazole-induced changes in forebrain dopaminergic and serotonergic function that may reflect its partial agonist activity at presynaptic dopamine D_2 and 5-HT_{1A} receptors and antagonist activity at 5-HT_{2A} receptors.

Keywords: Prefrontal cortex; Striatum; Dopamine; 5-HT (5-hydroxytryptamine, serotonin); Antipsychotic; Schizophrenia

1. Introduction

Aripiprazole, 7-{4-[4-(2, 3-dichlorophenyl)-1-piperazinyl]butoxy}-3,4-dihydro-2(1*H*)-quinolinone, is a next generation atypical antipsychotic that is active against the positive and negative symptoms of schizophrenia, has a low propensity for extrapyramidal symptoms, causes minimal weight gain and sedation, and produces minimal or no change in serum prolactin levels or QT_c interval prolongation (Kane et al., 2002). Aripiprazole has a mechanism of action that differs from all currently marketed typical and atypical antipsychotics. Unlike current antipsychotics, which act as dopamine receptor antagonists, biochemically, aripiprazole has been shown to be a potent, partial agonist at members of the D_2 family of dopamine receptors (Inoue et al., 1996; Lawler et al., 1999). In vivo, aripiprazole behaves as a dopamine D_2 receptor antagonist in rodent models of dopaminergic hyperactivity (e.g. inhibition of apomorphine-induced stereotypy) and as a dopamine D_2 receptor agonist in a model of dopaminergic hypoactivity (blockade of increased dopamine synthesis in reserpine-treated rats) (Kikuchi et al., 1995). Aripiprazole also provokes and blocks yawning behavior in drug-naïve and apomorphine-treated rats, respectively (Fujikawa et al., 1996). Taken together, these in vivo observations are consistent with in vitro evidence that aripiprazole is a partial agonist at dopamine D_2 receptors (Lawler et al., 1999; Burris et al., 2002). In addition, recent in vitro functional assays have revealed that aripiprazole also interacts with 5-HT receptor subtypes. In this respect, aripiprazole displays potent, partial agonist activity at cloned human (Jordan et al., 2002a) and native rat hippocampal (Jordan et al., 2002b) 5-HT_{1A} receptors, and antagonist activity at rat 5-HT_{2A} receptors (McQuade et al., 2002).

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The technique of in vivo microdialysis is commonly used to investigate drug effects on rat brain extracellular concentrations of neurotransmitters and their metabolites. Typical and atypical antipsychotic drugs most commonly produce relatively large increases in dopamine and its major metabolites, 3,4-dihydroxyphenylacetic acid (DOPAC) and homovanillic acid (HVA), in either the prefrontal cortex or striatum, or in both of these brain regions (Volonte et al., 1997; Westerink et al., 1998; Cartmell et al., 2000; Rayevsky et al., 1995; Watanabe and Hagino, 1999). These antipsychotic-induced increases in dopamine output and metabolism are in agreement with the antagonist activities of these drugs at dopamine D_2 receptors, and contrast with the inhibitory effects of dopamine D_2 receptor agonists on dopamine output and metabolism (Millan et al., 2000). In comparison, despite in vitro evidence that aripiprazole binds with high affinity to dopamine D_2 receptors in rat brain (Kikuchi et al., 1995; Lawler et al., 1999), in the only in vivo microdialysis study published to date on the effects of aripiprazole on forebrain dopaminergic function, aripiprazole produced changes in dopamine, DOPAC and HVA that are neither consistent with dopamine D_2 receptor agonist or antagonist activity. Rather, acute administration of aripiprazole slightly inhibited dopamine output and moderately stimulated DOPAC and HVA accumulation in rat prefrontal cortex and striatum (Semba et al., 1995), an overall profile that is distinct amongst all other antipsychotic drugs and which may represent a partial agonist activity of aripiprazole at dopamine D_2 receptors. The present study used in vivo microdialysis to confirm and extend upon the work of Semba et al. by examining the effects of aripiprazole on basal extracellular concentrations of dopamine, DOPAC, HVA and 5-hydroxyindole acetic acid (5-HIAA), a major metabolite of 5-HT that may be considered as an index of serotonergic neuronal activity (Stenfors et al., 1999). Concentrations were assessed in the medial prefrontal cortex and striatum of conscious, unrestrained rats. Measurements of 5-HIAA were included in this study to provide in vivo evidence that aripiprazole is a partial agonist at 5-HT_{1A} receptors. Aripiprazole was also studied for its effects on striatal dopamine, DOPAC, HVA and 5-HIAA in rats receiving chronic aripiprazole treatment, to determine whether potential changes in dopaminergic or serotonergic sensitivity develop after repeated aripiprazole exposure.

2. Methods

2.1. Animals and surgery

Male Sprague–Dawley rats (Charles River Laboratories, Raleigh, NC) were housed under conditions of controlled temperature (23 ± 1 °C) and lighting (12-h light–dark cycle, lights on at 0700 h), with unlimited access to food and water. All animals were adapted to the laboratory for 14 days prior to surgery, at which time they weighed 270–330 g. The experimental protocol was approved and conducted in accordance with Otsuka America Pharmaceutical's Institutional Animal Care and Use Committee (IACUC).

2.2. In vivo microdialysis

Rats were anesthetized with pentobarbital (65 mg/kg, i.p.) and mounted in a stereotaxic frame (Stoelting, Wood Dale, IL). A CMA-11 guide cannula (CMA-Microdialysis, Acton, MA) was stereotaxically implanted into the dorsal medial prefrontal cortex or central striatum. Two days after cannulation, a microdialysis probe (CMA-12, 4 mm tip length) was inserted into the guide cannula so that its tip terminated in either the medial prefrontal cortex (AP = + 3.2 mm, LAT = − 0.6 mm, DV = − 5.3 mm from dura) or striatum (AP = + 1.2 mm, LAT = − 3.0 mm, DV = − 7.2 mm from dura) using coordinates according to the atlas of Paxinos and Watson (1998). Each probe was continuously perfused at 1 μl/min with sterile artificial cerebrospinal fluid (aCSF) (1.2 mM Na_2HPO_4, 0.27 mM NaH_2PO_4, 140 mM NaCl, 3 mM KCl, 2.5 mM $CaCl_2$, 7.2 mM glucose, pH 7.29) using a CMA-102 microinfusion pump. Animals were individually housed for the duration of the experiment in a Raturn® animal system (BAS, West Lafayette, IN) and microdialysate samples were collected at 30 min intervals (CMA-142 microfraction collector) into silanized microvials containing 5 μl of 0.1N HCl to reduce oxidation of monoamines. Vehicle and drug treatments were administered 5 h after microdialysis probe implantation and microdialysate sampling continued for 3 h thereafter. A dissecting microscope was used to verify correct microdialysis probe placement at the end of each experiment.

2.3. Drug treatments

All vehicle and drug treatments were administered by oral gavage using a 5 ml/kg dose volume. Rats received either an acute administration of vehicle, aripiprazole (2, 10 and 40 mg/kg) or olanzapine (1, 10 and 20 mg/kg) or were administered chronically with vehicle, aripiprazole (10 and 40 mg/kg) or olanzapine (20 mg/kg) daily for 21 consecutive days.

2.4. Biochemical conditions

An isocratic, high-performance liquid chromatography–electrochemical detection (HPLC–ECD) assay was used to simultaneously quantitate dopamine, DOPAC, 5-HIAA and HVA in single 30-μl samples of microdialysate. A commercial mobile phase (MD-TM, ESA, Chelmsford, MA), containing 75 mM NaH_2PO_4, 1.7 mM 1-octanesulfonic acid sodium salt, 25 μM EDTA and 10% acetonitrile (adjusted to pH 3.0 with phosphoric acid), was pumped at 0.4 ml/min

through an ESA MD-150 RP C-18 (150 × 4.6 mm, 3 μm ODS) column. Analyte detection was performed using an ESA Coulochem II Multi-Electrode Detector equipped with a 5014B Microdialysis Cell (first electrode set at −275 mV, second electrode at 250 mV; guard cell = 500 mV). Online data capture was performed using Waters Millennium Software version 3.20 for HPLC.

2.5. Statistics

Statistically significant differences ($P < 0.05$) (treatment × time) between vehicle and drug treatment effects on post-treatment extracellular concentrations of dopamine, DOPAC, HVA and 5-HIAA in the medial prefrontal cortex and striatum were established by repeated measures analysis of variance. Statistically significant differences between vehicle and individual drug dose effects on cortical and striatal dopamine, DOPAC, HVA and 5-HIAA were revealed by a post hoc Dunnett's multiple comparison test (dose × time). All statistical analyses were performed using complete post-dose microdialysate data sets collected 30–180 min post-dose.

2.6. Drugs

Aripiprazole and olanzapine were synthesized by Otsuka Pharmaceutical (Tokushima, Japan). Tragacanth gum and HVA were purchased from Sigma (St. Louis, MO). Dopamine, 5-HIAA and DOPAC were purchased from RBI (Natick, MA). Artificial CSF (aCSF) was filtered (0.2 μm) prior to use. All drugs were suspended in a 0.5% (w/v) tragacanth gum-distilled water vehicle.

3. Results

Mean basal concentrations of dopamine, DOPAC, HVA and 5-HIAA were each significantly higher in microdialysates collected from the striatum than from the medial prefrontal cortex in naïve animals (Table 1; $P < 0.001$, Mann–Whitney rank sum test). Chronic administration of either aripiprazole or olanzapine did not produce a significant ($P > 0.05$, unpaired t-test) change in mean basal concentrations of dopamine, DOPAC, HVA and 5-HIAA compared with mean basal concentrations of these neurochemicals in rats chronically administered vehicle (Table 2). These mean basal concentrations were calculated from microdialysates collected 21 h after rats were administered their penultimate daily exposure to drug or vehicle. The present HPLC–ECD system was capable of measuring an external standard of 5-HT dissolved in aCSF, with a lower limit of quantitation of 2.5 pg on column, although 5-HT was unable to be reliably detected in microdialysate samples and was therefore excluded from our analysis.

3.1. Acute effects of aripiprazole in the medial prefrontal cortex

Acute administration of aripiprazole did not alter extracellular dopamine concentrations in the medial prefrontal cortex (Fig. 1A). However, aripiprazole induced significant increases in medial prefrontal cortex DOPAC ($P < 0.001$; $F = 9.65$) and HVA ($P < 0.01$; $F = 6.0$) concentrations that were accompanied by a significant inhibition of medial prefrontal cortex 5-HIAA ($P < 0.0001$; $F = 21.3$). Post hoc analysis revealed that these effects on DOPAC and HVA were significant at the 10 and 40 mg/kg doses of aripiprazole (Fig. 1B,C), whereas a significant reduction in 5-HIAA was only produced at a dose of 40 mg/kg (Fig. 1D).

3.2. Acute effects of aripiprazole in the striatum

No statistically significant differences could be detected between the effects of individual doses of aripiprazole and vehicle upon striatal extracellular dopamine concentrations (Fig. 2A). Aripiprazole treatment significantly elevated extracellular levels of DOPAC ($P = 0.003$; $F = 7.4$) in the striatum, this effect being significantly different from vehicle at the 10 mg/kg dose only (Fig. 2B). Aripiprazole treatment also produced a significant rise in extracellular levels of HVA ($P = 0.002$; $F = 8.1$) that differed significantly from vehicle at the 10 and 40 mg/kg doses (Fig. 2C). Acute administration of aripiprazole significantly inhibited striatal extracellular 5-HIAA ($P < 0.0001$; $F = 17.8$) concentrations and this effect was significant at all doses tested (Fig. 2D).

3.3. Acute effects of olanzapine in the medial prefrontal cortex

Acute administration of olanzapine produced a significant ($P < 0.0001$; $F = 19.8$) increase in basal dopamine output; that at 10 and 20 mg/kg doses was considerably higher than that observed after vehicle (Fig. 3A). A single

Table 1
Mean basal dopamine (DA), DOPAC, HVA and 5-HIAA concentrations (pg/μl ± S.E.M. (N)) in microdialysate samples from medial prefrontal cortex and striatum of drug-naïve conscious, freely moving rats used for acute drug treatment studies

Brain region	DA	DOPAC	HVA	5-HIAA
Medial prefrontal cortex	0.11 ± 0.01 (41)	16.4 ± 2.0 (48)	26.0 ± 3.8 (51)	61.9 ± 5.8 (51)
Striatum	0.45 ± 0.03[a] (45)	283.6 ± 19.7[a] (65)	119.7 ± 7.4[a] (61)	106.3 ± 7.5[a] (60)

[a] $P < 0.001$ versus mean basal concentrations in medial prefrontal cortex (Mann–Whitney rank sum test).

Table 2
Mean basal concentrations (pg/μl ± S.E.M. (*N*)) of dopamine (DA), DOPAC, HVA and 5-HIAA in striatal microdialysate samples collected from conscious, freely moving rats chronically treated with vehicle, aripiprazole or olanzapine

Treatment (mg/kg, p.o.)	DA	DOPAC	HVA	5-HIAA
Vehicle	0.49 ± 0.11 (4)	348.9 ± 62.3 (4)	168.1 ± 30.9 (4)	81.3 ± 10.4 (4)
Aripiprazole, 10 × 20 days	0.55 ± 0.12 (6)	463.3 ± 92.9 (5)	170.3 ± 75.8 (6)	100.2 ± 15.0 (6)
Aripiprazole, 40 × 20 days	0.31 ± 0.08 (3)	428.4 ± 56.3 (5)	196.1 ± 55.7 (4)	92.4 ± 12.1 (6)
Olanzapine, 20 × 20 days	0.61 ± 0.15 (4)	443.0 ± 75.3 (5)	145.6 ± 38.5 (6)	86.0 ± 15.8 (5)

Mean basal concentrations of DA, DOPAC, HVA and 5-HIAA did not differ significantly ($P>0.05$, unpaired *t*-test) between vehicle and drug treatment groups.

exposure to olanzapine also stimulated large dose-dependent increases in extracellular concentrations of DOPAC ($P<0.0001$; $F=36.8$; significant at 10 and 20 mg/kg) and HVA ($P=0.0002$; $F=12.7$; significant at 1, 10 and 20 mg/kg) (Fig. 3B,C). In contrast, no significant changes in cortical 5-HIAA ($P<0.10$; $F=2.5$) concentrations were detected after acute administration of olanzapine (Fig. 3D).

3.4. Acute effects of olanzapine in the striatum

Acute administration of olanzapine considerably elevated striatal extracellular dopamine concentrations ($P<0.0001$; $F=29.8$) and this effect was significant at the 10 and 20 mg/kg doses (Fig. 4A). Olanzapine also produced large dose-dependent increases in concentrations of striatal DOPAC ($P<0.0001$; $F=23.7$; Fig. 4B) and HVA ($P=0.0006$; $F=10.5$; Fig. 4C) that were significant at all doses tested. These effects of olanzapine were accompanied by a small decrease in striatal 5-HIAA accumulation ($P<0.0001$; $F=18.6$), although this effect was only significant at the lowest dose (1 mg/kg) tested (Fig. 4D).

3.5. Chronic effects of aripiprazole in the striatum

Treatment with vehicle alone reduced extracellular dopamine in microdialysates collected from the striatum of rats administered chronically with vehicle (Fig. 5A). Aripiprazole produced a comparatively weaker inhibitory effect on striatal dopamine in the rats that had been administered chronically with aripiprazole ($P=0.01$; $F=14.9$; no significant difference between each dose of aripiprazole and vehicle). In addition to this, aripiprazole produced increases in extracellular DOPAC ($P=0.005$; $F=9.2$; Fig. 5B) and HVA ($P=0.04$; $F=4.5$; Fig. 5C) concentrations in striatal

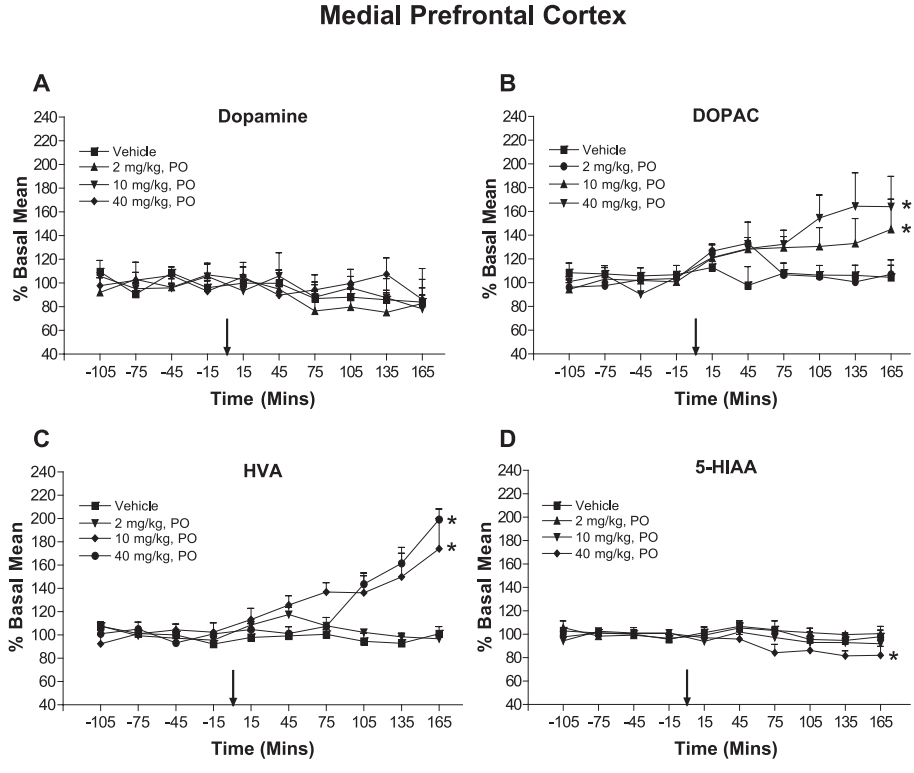

Fig. 1. Effect of acute administration of aripiprazole in medial prefrontal cortex: (A) dopamine (DA) output ($N=3-7$ per group); (B) DOPAC output ($N=3-10$ per group); (C) HVA output ($N=3-12$ per group); (D) 5-HIAA output ($N=3-10$ per group). Arrow indicates time of drug administration; * indicates statistical difference ($P<0.05$) between individual aripiprazole doses and vehicle (post hoc analysis using Dunnett's multiple comparison test).

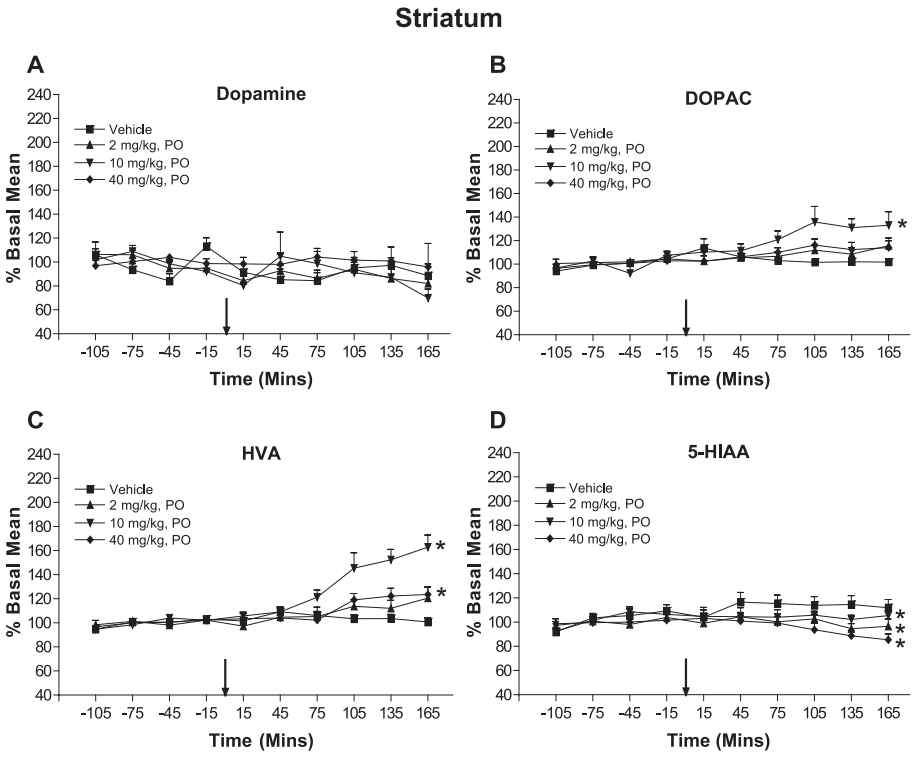

Fig. 2. Effect of acute administration of aripiprazole in striatum: (A) dopamine (DA) output ($N = 5-9$ per group); (B) DOPAC output ($N = 6-11$ per group); (C) HVA output ($N = 7-11$ per group); (D) 5-HIAA output ($N = 3-10$ per group). Arrow indicates time of drug administration; * indicates statistical difference ($P < 0.05$) between individual aripiprazole doses and vehicle (post hoc analysis using Dunnett's multiple comparison test).

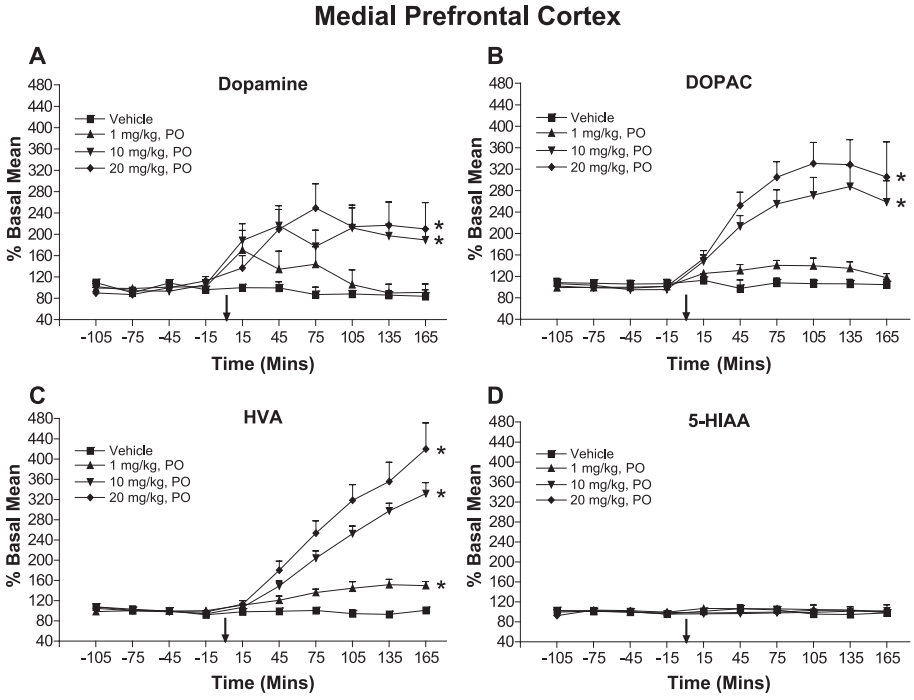

Fig. 3. Effect of acute administration of olanzapine in medial prefrontal cortex: (A) dopamine (DA) output ($N = 4-7$ per group); (B) DOPAC output ($N = 6-10$ per group); (C) HVA output ($N = 5-11$ per group); (D) 5-HIAA output ($N = 6-12$ per group). Arrow indicates time of drug administration; * indicates statistical difference ($P < 0.05$) between individual aripiprazole doses and vehicle (post hoc analysis using Dunnett's multiple comparison test).

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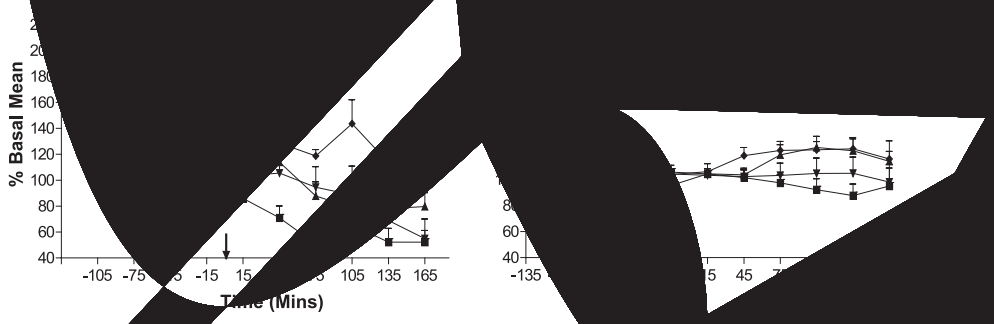

3.6. Chronic effects of olanzapine in the striatum

At the only dose tested (20 mg/kg), olanzapine produced significant increases in striatal dopamine, DOPAC, HVA and 5-HIAA concentrations in microdialysate samples collected from rats that had been treated chronically with olanzapine (Fig. 5A–D).

4. Discussion

The technique of in vivo brain microdialysis was used to monitor the effects of orally administered aripiprazole and olanzapine on basal extracellular concentrations of dopamine, DOPAC, HVA and 5-HIAA in the medial prefrontal cortex and striatum of conscious, freely moving rats. Acute administration of aripiprazole did not affect dopamine output in either the medial prefrontal cortex or striatum of drug-naïve rats. In a previous microdialysis study when aripiprazole was administered by intraperitoneal injection, reductions in dopamine output in the medial prefrontal cortex and striatum were observed in drug-naïve rats (Semba et al., 1995). The present lack of effect of aripiprazole on forebrain dopamine output may be related to pharmacokinetic differences resulting from the administration of aripiprazole by the oral rather than the intraperitoneal route. Nevertheless, both of these studies are in agreement in that they did not detect increases in frontocortical dopamine in response to aripiprazole. Based upon the effectiveness of aripiprazole against the negative symptoms and cognitive deficits of schizophrenia (Kane et al., 2002; Carson et al., 2002), these data challenge the view that atypical antipsychotic drugs improve these symptoms of schizophrenia by selectively increasing cortical dopamine release. In addition, and in accordance with the findings of Semba et al., aripiprazole did produce moderate increases in DOPAC and HVA concentrations in the medial prefrontal cortex and striatum of drug-naïve rats. We also observed a similar effect of aripiprazole in the striatum of rats that were chronically treated with aripiprazole that might indicate that long-term exposure to aripiprazole might not be expected to induce tolerance in striatal dopaminergic metabolism. In contrast with aripiprazole, acute administration with olanzapine produced relatively large dose-dependent increases in extracellular concentrations of dopamine, DOPAC and HVA in both the medial prefrontal cortex and striatum, and these effects of olanzapine on striatal dopamine and DOPAC concentrations were diminished in rats chronically treated with olanzapine. Previous microdialysis studies have demonstrated similar effects of olanzapine on these biochemical indices of forebrain dopaminergic function, which have been attributed to the activity of olanzapine as an antagonist at presynaptic dopamine D_2 receptors (Volonte et al., 1997; Li et al., 1998; Westerink et al., 1998). Aripiprazole also dose-dependently reduced extracellular 5-HIAA concentrations in the medial prefrontal cortex and striatum of drug-naïve rats in the current study. In contrast, aripiprazole had no effect on extracellular concentrations of this metabolite of 5-HT in the striatum of rats that had been chronically treated with aripiprazole, suggesting that chronic exposure to aripiprazole may induce a rapid functional desensitization of somatodendritic 5-HT_{1A} receptors, which has previously been demonstrated for 5-HT_{1A} receptor agonists (Kennett et al., 1987; Fuller and Perry, 1989). Aripiprazole has been shown to behave as a potent, partial agonist at rat hippocampal 5-HT_{1A} receptors (Jordan et al., 2002b) and cloned human 5-HT_{1A} receptors (Jordan et al., 2002a). In vivo electrophysiological experiments have also shown that aripiprazole dose-dependently inhibits the activity of 5-HT-containing neurons in the rat dorsal raphe nucleus (Sharp et al., personal communication), an effect also produced by the 5-HT_{1A} receptor partial agonist buspirone (Millan et al., 1992). Collectively, these and the present data suggest that aripiprazole activates presynaptic 5-HT_{1A} receptors to reduce either the biosynthesis or release of 5-HT that in turn would account for the inhibitory effects of aripiprazole on forebrain extracellular 5-HIAA concentrations. Further microdialysis studies are required to confirm whether aripiprazole inhibits forebrain 5-HT output and whether the effects of aripiprazole on forebrain serotonergic function are specifically mediated by 5-HT_{1A} receptors, as would be determined by their sensitivity to the 5-HT_{1A} receptor selective antagonist *N*-[2-[4-(2-methoxyphenyl)-1-piperazinyl]ethyl]-*N*-2-pyridinylcyclohexanecarboxamide (WAY-100635). In addition, in vitro experiments should also be performed to test the alternative possibility that aripiprazole may interfere with the enzymatic processes involved in the biosynthesis or metabolism of 5-HT.

Initial pharmacological evidence identified aripiprazole as a mixed agonist/antagonist at pre- and postsynaptic dopamine D_2 receptors, respectively (Kikuchi et al., 1995), as previously also demonstrated for 3-(3-hydroxyphenyl)-*N*-*n*-propylpiperidine (3-PPP), a partial agonist at dopamine D_2 receptors (Clark et al., 1985). Additional data have since accumulated to demonstrate that aripiprazole is also a partial agonist at dopamine D_2 receptors (Fujikawa et al., 1996; Inoue et al., 1996; Lawler et al., 1999). As a dopamine D_2 receptor partial agonist, aripiprazole could be expected to act as a functional agonist at presynaptic dopamine D_2 receptors, which have receptor reserve and low endogenous dopamine tone, and as a functional antagonist at post-synaptic dopamine D_2 receptors, which have little or no receptor reserve and a relatively high endogenous dopamine tone (Meller et al., 1987; Yokoo et al., 1988). Based on this model, a partial agonist activity of aripiprazole at presynaptic dopamine D_2 receptors could account for its ability to produce no change in dopamine while moderately increasing DOPAC and HVA in both the medial prefrontal cortex and striatum. These effects of aripiprazole are intermediate between the relatively large stimulatory and inhibitory effects typically produced by dopamine D_2 receptor antagonist and agonist drugs, respectively, upon forebrain dopamine and its metabolites

(Ferre and Artigas, 1995; Cartmell et al., 2000; Rayevsky et al., 1995; Millan et al., 2000). Indeed, a close inverse correlation exists between the relative intrinsic activity of drugs at dopamine D_2 receptors and their corresponding effects on striatal extracellular HVA concentrations (Lahti et al., 1992), and based upon this relationship aripiprazole would be predicted to behave as a partial agonist at dopamine D_2 receptors with a low relative intrinsic activity. Alternatively, the current effects of aripiprazole on forebrain dopaminergic function may be explained by its combined activity as a partial agonist at dopamine D_2 receptors, a partial agonist at 5-HT_{1A} receptors, and an antagonist at 5-HT_{2A} receptors. In a manner similar to that believed to account for the ability of other partial 5-HT_{1A} receptor agonist drugs to produce increases in striatal DOPAC and HVA (Protais et al., 1998; Nomikos et al., 1992; Ballarin et al., 1994; Meltzer, 1999; Millan et al., 2000), aripiprazole might activate 5-HT_{1A} autoreceptors in the dorsal raphe nucleus to lower the level of serotonergic tone on 5-HT_{2A} and 5-HT_{2C} heteroreceptors, which respectively exert excitatory and indirect inhibitory effects on forebrain dopaminergic function. Aripiprazole might also regulate dopaminergic metabolism by acting directly as a competitive antagonist at 5-HT_{2A} heteroreceptors. Other antipsychotic drugs have been shown to regulate cortical dopamine release via their combined antagonistic activities at dopamine D_2 and 5-HT_{2A} receptors (Ichikawa et al., 2001). In addition, aripiprazole has been recently reported to behave as a partial agonist at dopamine D_3 receptors (Shapiro et al., 2003), suggesting the present effects of aripiprazole might partly be mediated through dopamine D_3 autoreceptors. However, it is uncertain as to whether dopaminergic inhibitory autoreceptors are exclusively of the D_2 or D_3 subtype, or even whether dopamine D_3 receptors primarily regulate dopamine release rather than turnover (Millan et al., 2000). While the present study clearly demonstrates that the effects of aripiprazole on dopaminergic metabolites are clearly distinct from those of all other antipsychotic drugs, including olanzapine (Li et al., 1998), additional experiments, using dopamine D_2, D_3 and 5-HT_{1A}, 5-HT_{2A} receptor selective compounds, are however warranted to interpret whether this unique profile of aripiprazole is representative of its independent or collective activity at dopamine D_2, D_3 or 5-HT_{1A}, 5-HT_{2A} receptors, all of which appear to have the capacity to directly or indirectly regulate dopaminergic neuronal function.

In the present study, administration of vehicle reduced basal striatal dopamine output in microdialysates collected from rats that had been chronically administered with vehicle. The absence of a similar response in drug-naïve rats suggests this effect might represent an adaptive response of the nigrostriatal dopaminergic system to the effects of repeated oral gavage stress. This possible stress effect was less prominent when aripiprazole was administered to rats that had been chronically exposed to aripiprazole. These possible effects of oral gavage stress require further investigation, although they are not too surprising as other acute stressors have been shown to modify the sensitivity of the dopaminergic system to subsequent stress or pharmacological challenge (Fdez Espejo and Gil, 1997; Pani et al., 2000; Pacchioni et al., 2002).

In summary, aripiprazole produced moderate increases in basal DOPAC and HVA concentrations in the medial prefrontal cortex and striatum of rats that were drug-naïve and chronically exposed to aripiprazole. These modest effects on forebrain dopaminergic metabolism, and the lack of effect of individual aripiprazole doses on cortical and striatal dopamine output may reflect a partial agonist activity of aripiprazole at presynaptic dopamine D_2 receptors. Partial agonist activity of aripiprazole at 5-HT_{1A} receptors could indirectly contribute to these effects on forebrain dopaminergic function and to its suppression of striatal 5-HIAA in drug-naïve rats but not in rats chronically treated with aripiprazole. The results of the present study, together with other data demonstrating that aripiprazole is a partial agonist at dopamine D_2 receptors, a partial agonist at 5-HT_{1A} receptors, and an antagonist at 5-HT_{2A} receptors, suggest that aripiprazole may be best described as a dopamine–serotonin system stabilizer representing the first member of a new generation of antipsychotic drugs (Grady et al., 2003). This preclinical profile is supportive of the clinical efficacy of aripiprazole in the treatment of schizophrenia, including activity against both positive and negative symptoms with minimal risk of side effects such as extrapyramidal symptoms, sedation or elevated prolactin levels (Carson et al., 2002; Kane et al., 2002). Further microdialysis studies, using receptor-selective compounds, are required to investigate the role(s) played by individual subtypes of dopaminergic and serotonergic receptors in mediating the effects of aripiprazole on forebrain dopaminergic and serotonergic function.

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