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Involvement of $GABA_A$ receptors in the regulation of the prefrontal cortex on dopamine release in the rat dorsolateral striatum

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Abstract

The characteristics of the prefrontal cortex regulation of dopamine release in the rat dorsolateral striatum were investigated in focusing on the corticostriatal pathway, using dual-probe microdialysis in combination with simple behavioral procedures. Intracortical perfusion of the $GABA_A$ receptor antagonist bicuculline (0.03 and 0.1 mM) increased the striatal dopamine release, whereas the $GABA_A$ receptor agonist muscimol (0.1 and 1 mM) reduced the release dose dependently. Co-perfusion of muscimol (0.01 mM), which did not by itself affect the dopamine release, completely prevented the glutamate (1 mM)-stimulated dopamine release and behavioral activities, including locomotion and rearing. Muscimol (0.01 mM) also prevented the *N*-methyl-D-aspartate (NMDA; 1 mM)-induced increases in glutamate, as well as dopamine levels. These findings suggest that the prefrontal cortex functionally regulates dopamine release in the dorsolateral striatum, which is mediated via $GABA_A$ receptors. This cortical GABAergic system underlying the prefrontal cortex regulatory mechanism appears to be associated with the corticostriatal pathway.
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Keywords: Dopamine; Dorsolateral, striatum; Cortex prefrontal; $GABA_A$ receptor; Corticostriatal pathway

1. Introduction

The dorsal striatum, a structure of the basal ganglia, is innervated by dopamine nerve terminals from the substantial nigra pars compacta. This nigrostriatal dopaminergic pathway is known to play a crucial role in sensorimotor coordination and initiation of movements. The mesolimbic dopaminergic pathway originating from the ventral tegmental area, on the other hand, innervates in the ventral striatum, including the nucleus accumbens. These subcortical regions, i.e., dorsal and ventral striatum receive glutamatergic input from the cortex, the so-called corticostriatal pathway (Alexander and Crutcher, 1990; Groenewegen et al., 1997). Numerous studies have shown that the dopaminergic system in the ventral striatum is under the prefrontal cortex regulation directly or indirectly through the corticostriatal pathway (Taber and Fibiger, 1995; Taber et al., 1995; Karreman and Moghaddam, 1996). For instance, Karreman and Moghaddam (1996) demonstrated that the prefrontal cortex regulates facilitatory dopamine release in the nucleus accumbens indirectly via glutamatergic projection to the ventral tegmental area. Jackson et al. (2001) have recently shown that direct activation of the prefrontal cortex causes not increases but decreases in dopamine release in the nucleus accumbens, under the physiological conditions. An understanding of the prefrontal cortex regulatory mechanism through the corticostriatal pathway would provide insights into the pathophysiology of disorders associated with subcortical dopaminergic dysfunctions such as schizophrenia and Parkinson's disease. Few studies, however, have addressed concerning the prefrontal cortex regulatory mechanism of dopaminergic system in the dorsal striatum.

The dorsal striatum consists of heterogeneous structures, i.e., medial and lateral subregions (Graybiel, 1990). Recent studies have shown the differential characteristics of the corticostriatal pathway that project to each subregion: Partridge et al. (2000) reported that regional and postnatal heterogeneity of corticostriatal synaptic plasticity, a cellular mechanism underlying motor learning and memory (Charp-

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ier and Deniau, 1997), exists between the dorsolateral and dorsomedial striatum. The difference in synaptic plasticity may be attributable to the information processing and/or development of the circuitry of the basal ganglia. Indeed, each subcortical region appears to have different roles in learning and memory paradigms, i.e., lesions in the dorsolateral striatum disrupt sensorimotor function and stimulus–response learning (Reading et al., 1991; Tang and Lovinger, 2000), whereas lesions of the dorsomedial striatum produce impairment of cognitive–spatial tasks (Devan et al., 1996; Furtado and Mazurek, 1996). Devan and White (1999) suggested that the differential functions in each region are due to the corticostriatal connections, i.e., the dorsolateral striatum directly receives glutamatergic corticostriatal projections from the prefrontal cortex, whereas the corticostriatal pathway in the dorsomedial region cooperatively contacts the hippocampal system. To elucidate the prefrontal cortex regulation of dopamine release through the corticostriatal pathway, therefore, would be important for understanding the function of the dorsolateral striatum.

The aim of present study is to elucidate the characteristics of the prefrontal cortex regulation of dopamine release in the rat dorsolateral striatum in focusing on the corticostriatal pathway. For this purpose, pharmacological manipulations were applied to the prefrontal cortex and extracellular levels of dopamine and/or glutamate were determined in the ipsilateral striatum using dual-probe microdialysis in combination with a behavioral procedure. The prefrontal cortex is composed of two major neuronal populations, glutamatergic neurons and GABAergic interneurons. Numerous neurochemical studies have shown that the cortical GABAergic neurons contribute to glutamatergic (Obrietan and van den Pol, 1999; Mitchell and Silver, 2000) and dopaminergic neuronal activity (Santiago et al., 1993; Feenstra et al., 1998; Doherty and Gratton, 1999). Berretta et al. (1997) reported that $GABA_A$ receptors are involved in the cortically evoked activation of immediate-early gene protein products in the dorsolateral striatum. These findings lead us to speculate that the GABAergic system in the cortical circuitry may be associated with the corticostriatal projection. Based on this assumption, the present experiment was performed in focusing on the cortical $GABA_A$ receptors.

2. Materials and methods

2.1. Animals

Male Wistar-strain rats (10–14 weeks old) were used. Rats were housed in a room with a 12-h light/dark cycle under the constant temperature (21 ± 2 °C). All animals handling was performed in accordance with guidelines for the Care and Use of Laboratory Animals of the Animal Research Committee of the Hokkaido University School of Medicine.

2.2. Dual probe microdialysis

Rats were anesthetized with ketamine (100 mg/kg, i.p.) and 3-mm concentric guide cannulae implanted into the prefrontal cortex (stereotaxic coordinates: anterior 3.2, lateral 0.7, ventral 1.0 mm) and the ipsilateral region of the dorsolateral striatum (posterior 1.4, lateral 4.2, ventral 4.2 mm) according to the atlas of Paxinos and Watson (1986). Two days after surgery, a concentric dialysis probe with a 3-mm tip (0.22 μm O.D. regenerated cellulose 50,000 MW cut-off, Eicom, Kyoto, Japan) was inserted through the guide cannulae. One probe was implanted in the prefrontal cortex, and the other probe was placed in the dorsolateral striatum. Drugs were infused by retrograde microdialysis into the prefrontal cortex. Dialysis probes were perfused at 1 μl/min with artificial cerebrospinal fluid (aCSF) (KCl 2.7, NaCl 140, $CaCl_2$ 1.2, $MgCl_2$ 1.0, NaH_2PO_4 0.3, Na_2HPO_4 1.7 mM) for 120–180 min to obtain the stable baseline before sampling. Successive samples were collected at 20 min intervals and were injected directly onto the HPLC column to determine the extracellular levels of dopamine in the dorsolateral striatum. Some samples were divided for simultaneous determination of glutamate levels. At the end of experiment, the localization of the probe was histologically examined.

2.3. Determination of extracellular levels of dopamine and glutamate

Extracellular levels of dopamine were analyzed using HPLC-ECD as previous methods (Matsumoto et al., 1998). Briefly, a graphite working electrode was maintained at 450 mV and the mobile phase consisted of 0.1 M sodium dihydrogenphosphate/0.1 M disodium hydrogen phosphate buffer (pH 6.0) with 1.16 mM of octanesulfonate, 0.15 mm EDTA-2Na and 18% (V/V) methanol. Samples for glutamate analysis were stored at −80 °C until assay within at least 2 weeks. The solution used to form fluorescent derivatives of glutamate was prepared by *o*-phthaldialdehyde containing mercaptoethanol. Ten μl of this solution was added to 30 μl of dialysate sample diluted with 50% methanol, and this mixture was allowed to react for 2.5 min before application to the column. The HPLC apparatus consisted of a reverse phase column together with a fluorometric detector (emission, 445 nm; extraction, 340 nm) (FLD-370, Eicom). The elute was a mixture of 0.1 M sodium dihydrogenphosphate/0.1 M disodium hydrogenphosphate buffer (pH 6.0) and methanol in the v/v proportion of 7:3.

2.4. Behavioral analysis

Motor activity was evaluated by analyzing simple behavioral procedure, i.e., locomotion and rearing. The locomotor activity was assessed by number of crossing line, which was subdivided each wall of clear polycarbonate cage (width × length × height: 30 × 30 × 30 cm). The numbers of single line crossing (passing a line with forepaws) and

rearing were counted before, during and after infusion of drugs.

2.5. Drugs

Drugs including L-glutamic acid (glutamate), *N*-methyl-D-aspartate (NMDA), MK-801 hydrogen maleate (MK 801), muscimol, (+)-bicuculline, GBR 12909 dihydrochloride (GBR 12909) and tetrodotoxin were purchased from RBI-Sigma. (*S*)-α-Amino-3-hydroxy-5-methyl-4-isoxaxolepropionic acid ((*S*)-AMPA) was purchased from Tocris Cookson. All drugs were diluted in the aCSF before application.

2.6. Statistics

Experimental values were given as means ± S.E.M. Values obtained by use of in vivo microdialysis were expressed as a percentage of the baseline level before drug perfusion. The area under the curves (AUC) of the time course changes is given to reflect the ensemble average. All statistical analysis was performed with the AUC value ($\times 10^3$ %/min) during 140 min after perfusion of drugs. For comparison with the experimental groups, Student's unpaired *t* test or Dunnett's test was conducted after ANOVA. $P < 0.05$ was considered significant.

3. Results

3.1. Pharmacological assessment of intracortical perfusion using dual-probe microdialysis

In this experiment, basal dopamine levels not corrected by the probe's recovery were 1.27 ± 0.18 fmol/μl (mean ± S.E.M., $n = 73$). These average values were not significantly different among the drug-treated groups.

The sodium channel blocker tetrodotoxin (0.01 mM), perfused directly into the dorsolateral striatum, reduced markedly dopamine release. Striatal dopamine levels were also decreased during intracotical perfusion of tetrodotoxin and gradually returned to the basal levels (Fig. 1). Significant decreases in dopamine release compared with controls, in which aCSF solution was perfused into the prefrontal cortex, were observed at 20 min (tetrodotoxin, 66.6 ± 11.2%, $n = 4$; controls, 103.8 ± 3.6%, $n = 7$, $P < 0.05$) and at 60 min (tetrodotoxin, 67.3 ± 8.9%, $n = 4$; controls, 97.0 ± 3.4%, $n = 7$, $P < 0.05$).

To examine whether tetrodotoxin perfused into the prefrontal cortex diffused to the ipsilateral striatum and exerted effects locally, fundamental experiments were performed. Direct perfusion of a selective dopamine reuptake inhibitor GBR 12909 (0.1 mM) markedly increased the striatal dopamine release, whereas intracortical perfusion of this drug did not affect the dopamine level (Fig. 1B). A high concentration of potassium (K^+, 60 mM), perfused into the dorsolateral striatum, elicited dopamine release by approximately three-fold. Intracortical perfusion of K^+, however, did not cause any change in the striatal dopamine release (Fig. 1C).

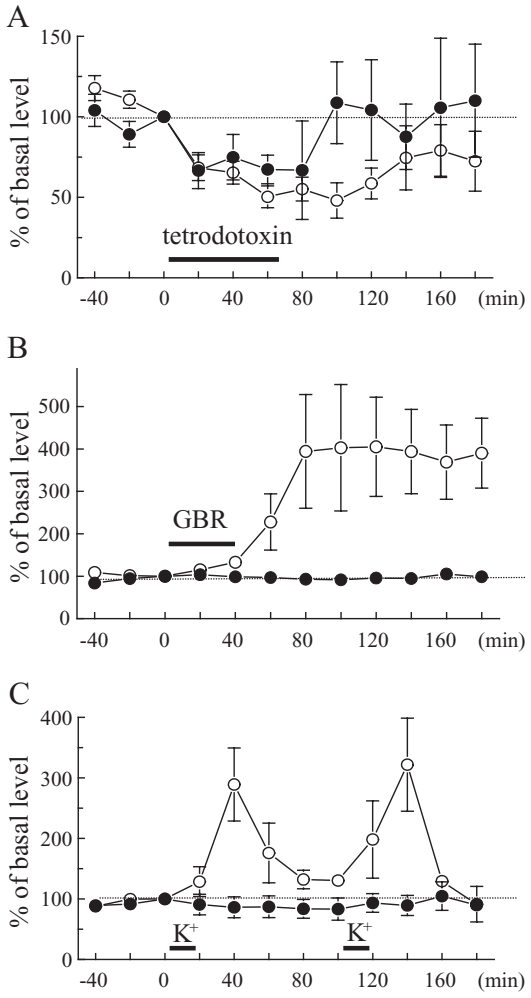

Fig. 1. Pharmacological assessment of intracortical perfusion using dual probe microdialysis. Effects of the sodium channel blocker tetrodotoxin (A), the selective dopamine reuptake inhibitor GBR 12909 (GBR) (B) and potassium (K^+) (C) on dopamine release in the dorsolateral striatum (STR). Tetrodotoxin (0.01 mM) was perfused via reversed dialysis into the prefrontal cortex (●, $n = 4$) or directly perfused into the STR (○, $n = 4$). GBR 12909 (0.1 mM) was perfused into the prefrontal cortex (●, $n = 3$) or perfusion into the STR (○, $n = 4$). A high concentration of K^+ (60 mM) were perfused into the prefrontal cortex (●, $n = 3$) or perfusion into the STR (○, $n = 4$). Values are expressed as a percentage of the baseline level before drug perfusion. Means ± S.E.M.

3.2. Effects of bicuculline and muscimol, perfused into the prefrontal cortex, on extracellular levels of dopamine in the dorsolateral striatum

The $GABA_A$ receptor antagonist bicuculline was perfused into the prefrontal cortex via retrograde dialysis for 20 min. Extracellular levels of dopamine in the dorsolateral striatum were increased by bicuculline (0.03 and 0.1 mM) in a concentration-dependent manner (Fig. 2A). Significant

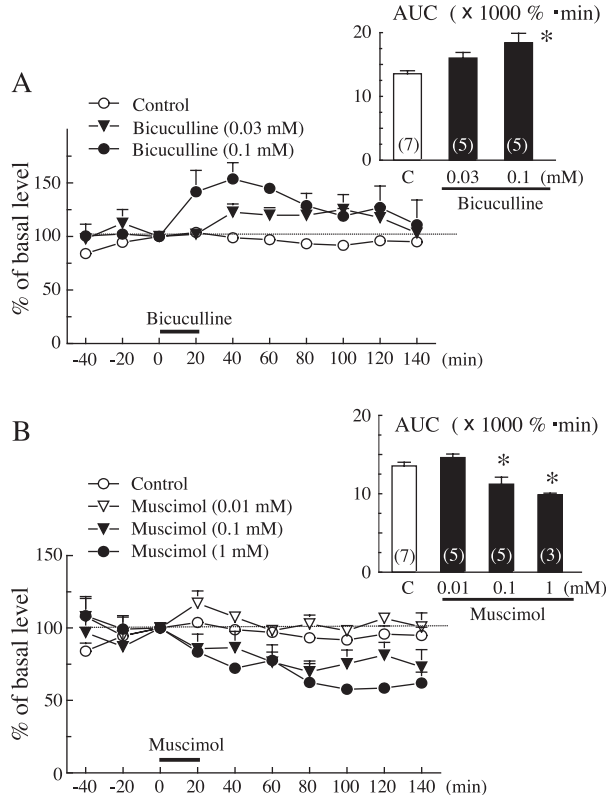

Fig. 2. Effects of bicuculline and muscimol, perfused into the prefrontal cortex, on extracellular levels of dopamine in the dorsolateral striatum. The $GABA_A$ receptor antagonist bicuculline (0.03 and 0.1 mM) (A) or the $GABA_A$ receptor agonist muscimol (0.01, 0.1 and 1 mM) (B) was perfused into the prefrontal cortex during 20 min. The time-course response and area under the curves (AUC) during 0–140 min are shown. Values are expressed as a percentage of the baseline level before drug perfusion. Means ± S.E.M. The number in each column indicates the number of rats tested. *$P<0.05$ vs. controls (C), which were perfused with artificial cerebrospinal fluid (aCSF) solution into the prefrontal cortex.

increases in dopamine release were observed after perfusion of 0.1 mM bicuculline. A high concentration of bicuculline (1 mM) was not studied because of the induction of strong behavioral activation such as hyperlocomotion, rearing and turning.

Intracortical perfusion of the $GABA_A$ receptor agonist muscimol (0.1 and 1 mM) caused significant and dose-dependent decreases in dopamine levels (Fig. 2B). A low concentration of muscimol (0.01 mM) did not affect the dopamine release.

3.3. Effects of muscimol, co-perfused into the prefrontal cortex, on the striatal dopamine release induced by glutamate

Intracortical perfusion of glutamate (1 mM) significantly increased the striatal dopamine level with a maximum response of 205.9 ± 30.7% ($n=6$), whereas 0.1 mM glutamate did not affect the dopamine levels. Glutamate (1 mM)-induced increase in dopamine release was completely prevented by co-perfusion of muscimol (0.01 mM) (Fig. 3). Muscimol (0.01 mM) by itself did not affect the extracellular level of dopamine during 80 min of perfusion.

3.4. Effects of muscimol, co-perfused into the prefrontal cortex, on behavioral changes induced by glutamate

Behavioral activity was estimated by locomotion and rearing, during the microdialysis experiment. Locomotor activity and rearing were markedly enhanced during perfusion of glutamate (1 mM) into the prefrontal cortex. As shown in Fig. 4, glutamate-stimulated behavioral activities were reduced in the presence of muscimol (0.01 mM). Muscimol alone did not influence either behavior. Locomotion and rearing were dramatically increased during perfusion of bicuculline (0.1 mM) and continued 40 min after perfu-

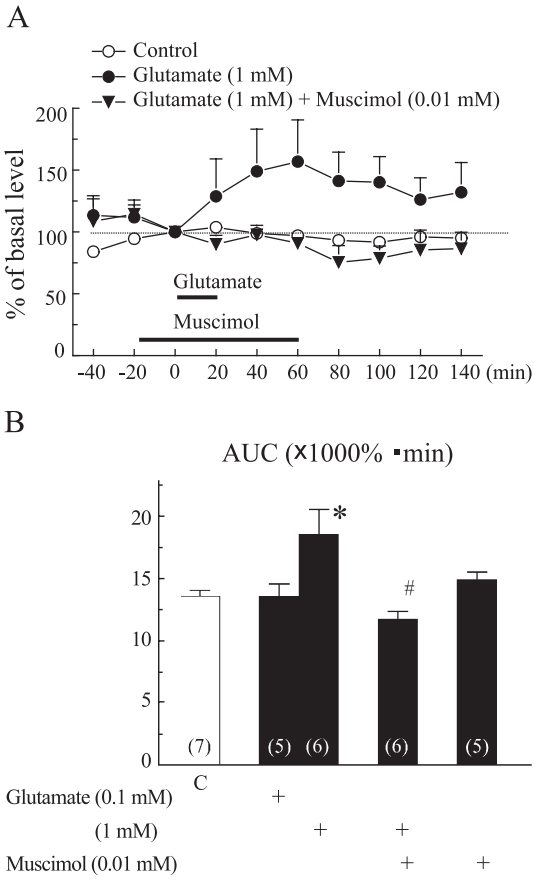

Fig. 3. Effects of muscimol, co-perfused into the prefrontal cortex, on the striatal dopamine release induced by glutamate. Glutamate (0.1 and 1 mM) was perfused into the prefrontal cortex for 20 min. The $GABA_A$ receptor agonist muscimol (0.01 mM) was perfused 20 min before glutamate (1 mM) administration and continued for 80 min. The time-course response (A) and area under the curves (AUC) during 0–140 min (B) are shown. Values are expressed as a percentage of the baseline level before glutamate perfusion. Means ± S.E.M. The number in each column indicates the number of rats tested. *$P<0.05$ and $^{\#}P<0.05$ vs. controls (C), which were perfused with artificial cerebrospinal fluid (aCSF) solution and rats given glutamate alone, respectively.

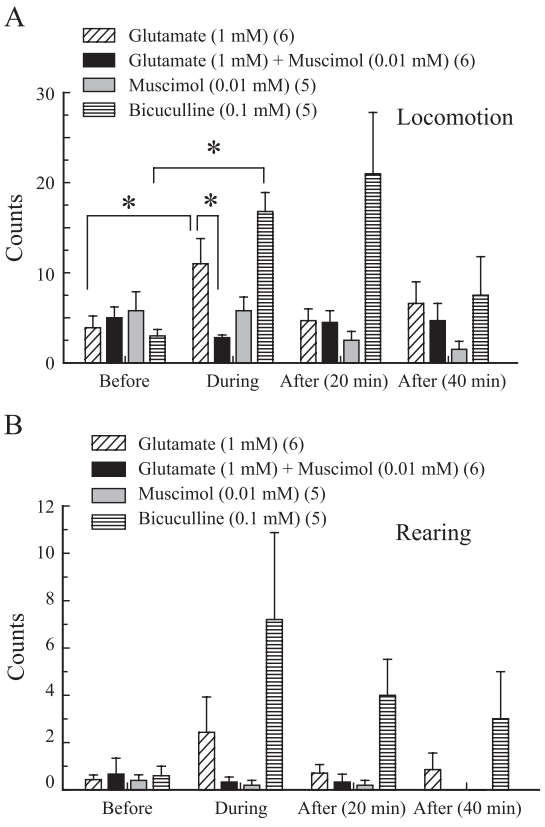

Fig. 4. Effects of muscimol, co-perfused into the prefrontal cortex, on behavioral changes induced by glutamate. Motor activity was estimated as locomotion (A) and rearing (B). These behaviors were counted before, during, 20 min and 40 min after perfusion of drugs, accompanied by in vivo microdialysis. Glutamate (1 mM) and the $GABA_A$ receptor antagonist bicuculline (0.1 mM) were perfused into the prefrontal cortex for 20 min. The $GABA_A$ receptor agonist muscimol was perfused 20 min before glutamate (1 mM) application and continued for 80 min. Means ± S.E.M. The numbers of rats tested are given in parentheses. $^*P < 0.05$.

sion. Bicuculline (0.1 mM)-stimulated behavioral activities were greater than those induced by glutamate (1 mM).

3.5. Effects of NMDA, perfused into the prefrontal cortex, on the extracellular levels of dopamine and glutamate in the dorsolateral striatum

Glutamate (1 mM)-induced increases in dopamine levels were not found in the presence of the non-selective NMDA receptor antagonist MK-801 (10 μM) (AUC ($\times 10^3$ %/min): glutamate; 19.1 ± 1.8, $n = 6$, glutamate + MK 801; 13.0 ± 0.8, $n = 5$, $P < 0.05$), suggesting that the NMDA receptors are involved in the prefrontal cortex regulation of striatal dopamine release. A high concentration of NMDA (1 mM) significantly increased dopamine release compared with the controls (AUC ($\times 10^3$ %/min): NMDA; 17.1 ± 1.0, $n = 7$; controls; 13.6 ± 0.5, $n = 7$, $P < 0.05$). Intracortical perfusion of AMPA (1 mM) did not cause any changes in the dopamine levels (AUC ($\times 10^3$ %/min): 14.1 ± 2.0, $n = 4$).

To further elucidate the involvement of NMDA receptors in the glutamatergic regulatory mechanism, extracellular levels of glutamate, as well as dopamine, were determined in the dorsolateral striatum. The basal level of glutamate not corrected by probe recovery was 3.23 ± 0.52 pmol/μl (mean ± S.E.M., $n = 24$). There were no significant differences of the basal values among the groups. As shown in Fig. 5, intracortical perfusion of NMDA (1 mM) significantly increased the glutamate level, with a maximum response of $157.9 \pm 1.6\%$ ($n = 5$). This facilitatory effect appeared to be identical to that of dopamine release induced by NMDA (maximum response; $153.0 \pm 12.9\%$, $n = 7$).

Additional experiments were performed to examine the dynamic changes in the dopamine and glutamate levels after direct perfusion of NMDA into the dorsolateral striatum. Significant increases in dopamine levels were observed at

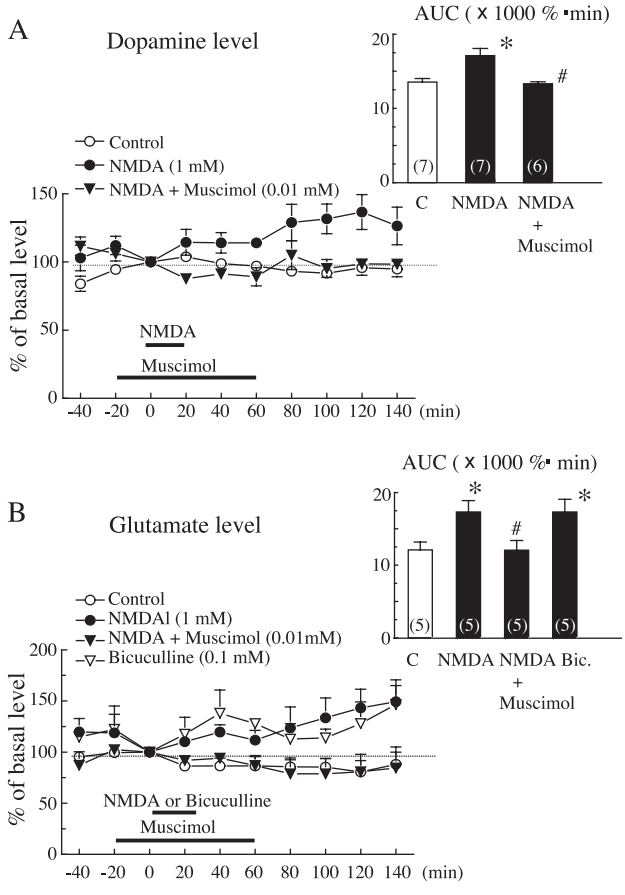

Fig. 5. Effects of NMDA, perfused into the prefrontal cortex, on the extracellular levels of dopamine and glutamate in the presence of muscimol. NMDA (1 mM) or the $GABA_A$ receptor antagonist bicuculline (Bic.) (0.1 mM) were perfused into the prefrontal cortex for 20 min. The $GABA_A$ receptor agonist muscimol (0.01 mM) was perfused 20 min before NMDA administration and continued for 80 min. The time-course response and area under the curves (AUC) during 0–140 min of dopamine (A) and glutamate levels (B) are shown. Values are expressed as a percentage of the baseline level before NMDA perfusion. Means ± S.E.M. The number in each column indicates the number of rats tested. $^*P < 0.05$ and $^{\#}P < 0.05$ vs. controls (C) and rats given NMDA alone, respectively.

40–80 min after perfusion of NMDA with a maximum response of 231.5 ± 47.9% ($n = 4$). AUC ($\times 10^3$% min) after NMDA perfusion (22.3 ± 3.9) was significantly increased compared with controls (13.6 ± 0.5, $n = 7$, $P < 0.05$). Striatal glutamate levels were also significantly enhanced after perfusion of NMDA (1 mM) (AUC ($\times 10^3$% min): NMDA and control: 21.5 ± 1.2, $n = 4$ and 12.1 ± 1.1, $n = 5$, respectively).

3.6. Effect of muscimol, co-perfused into the prefrontal cortex, on striatal dopamine and glutamate release induced by NMDA

NMDA (1 mM)-induced increases in dopamine release were significantly prevented by co-perfusion of 0.01 mM muscimol (AUC ($\times 10^3$ %/min): NMDA, 17.1 ± 1.0, $n = 7$; NMDA + muscimol, 13.3 ± 0.3, $n = 6$, $P < 0.05$). NMDA (1 mM)-stimulated glutamate, as well as dopamine levels, were completely abolished by co-perfusion of muscimol (0.01 mM) (Fig. 5). The $GABA_A$ receptor antagonist bicuculline (0.1 mM) caused significant increases in not only dopamine (see Fig. 2A) but also the glutamate levels. A low concentration of bicuculline (0.03 mM) tended to enhance the NMDA (1 mM)-induced increases in glutamate (AUC ($\times 10^3$ %/min): NMDA; 12.1 ± 1.1, $n = 5$, NMDA + bicuculline; 19.3 ± 2.0, $n = 5$), but there were no significant difference between two groups.

4. Discussion

In this study, the $GABA_A$ receptor antagonist bicuculline, perfused into the prefrontal cortex, enhanced dopamine release, whereas the $GABA_A$ receptor agonist muscimol reduced dopamine levels in the dorsolateral striatum. Intracortical perfusion of glutamate increased dopamine release and behavioral activities, and these facilitatory responses were completely inhibited by a low concentration of muscimol. These findings suggest that $GABA_A$ receptors contribute to the prefrontal cortex regulation of dopamine release in the dorsolateral striatum. Furthermore, the cortical GABAergic system underlying the prefrontal cortex regulatory mechanism appears to be associated with the corticostriatal pathway.

In the present experiment using dual-probe microdialysis, a technical concern was the close proximity of the prefrontal cortex and ipsilateral striatum, i.e., the drugs applied into the prefrontal cortex might diffuse to the dorsolateral striatum and exert their effects locally. This possibility cannot be excluded completely, however, we observed that intracortical perfusion of neither a high concentration of K^+ nor a selective dopamine reuptake inhibitor GBR 12909 altered the striatal dopamine levels. The changes in dopamine release observed in this study were, therefore, not likely due to the diffusion of drugs, at least under the present experimental conditions.

Intracortical perfusion of tetrodotoxin significantly decreased striatal dopamine release, suggesting that the dopaminergic neuronal activity in the dorsolateral striatum was under the tonic excitatory control of the prefrontal cortex. Previous studies showed that electrical and chemical stimulation of the prefrontal cortex enhanced the striatal dopamine release (Moghaddam et al., 1990; Taber and Fibiger, 1993). Consistent with these findings, a high concentration of glutamate perfused into the prefrontal cortex, increased dopamine release in the dorsolateral striaum. Thus, exogenous glutamate can directly stimulate the corticostriatal pathway and might subsequently enhance the dopamine release from the nerve terminals. In turn, glutamatergic projection seems to tonically activate the dopamine release through the corticostriatal pathway. Intracortical perfusion of the $GABA_A$ receptor antagonist bicuculline also increased the striatal dopamine levels. Since $GABA_A$ receptor antagonists suppress the actions of cortical inhibitory GABAergic interneurons (Connors et al., 1988), it is conceivable that disinhibition of the prefrontal cortex by bicuculline produces increases in the striatal dopamine release. In other words, activation of the corticostriatal pathway is likely to be caused by a reduction in $GABA_A$ receptor-mediated intracortical inhibition. This possibility is strengthened by the finding that glutamate-induced increases in dopamine release were completely abolished by the $GABA_A$ receptor agonist muscimol.

A functional link between the cortical $GABA_A$ receptors and the glutamatergic pathway was supported by the behavioral findings: intracortical perfusion of glutamate caused increases in locomotion and rearing, which were prevented by co-perfusion of muscimol. Furthermore, we observed that bicuculline, which increased extracellular levels of both dopamine and glutamate, enhanced motor activity. These neurochemical and behavioral findings appear to be in line with the results reported by Berretta et al. (1997), i.e., the $GABA_A$ receptor antagonist picrotoxin stimulated the cortically evoked activation of c-Fos and Jun B in the dorsolateral striatum. It should be noted that bicuculline produced heightened locomotor activity compared with that induced by glutamate, despite each drug causing identical elevation of dopamine release. We could not exclude the possibility that other neurotransmitter systems and/or other brain regions are implicated in the motor outcome observed in this study. Nevertheless, however, the present data strongly indicate that the cortical $GABA_A$ receptors are associated with the dorsolateral striatal functions such as motor control.

Glutamate-induced increases in dopamine release were abolished by the non-selective NMDA receptor antagonist MK-801, suggesting that the prefrontal cortex regulates the striatal dopamine release, at least in part, via NMDA receptors. Indeed, intracortical perfusion of NMDA, not AMPA, caused increases not in only dopamine but also in glutamate levels in the dorsolateral striatum. NMDA-induced increases in dopamine and glutamate levels were

prevented by co-perfusion of the $GABA_A$ receptor agonist muscimol. Conversely, the $GABA_A$ receptor antagonist bicuculline increased glutamate as well as dopamine release. These results strongly support the present finding that striatal dopamine release was regulated via cortical $GABA_A$ receptors through the corticostriatal pathway.

Electrophysiological studies have shown that the glutamatergic corticostriatal afferent induces long-term potentiation and/or long-term depression in the dorsolateral striatum (Centonze et al., 1999, 2001; Reynolds and Wickens, 2002). Although the precise cellular mechanism of corticostriatal synaptic plasticity remains unknown, it is generally considered that the NMDA component plays a critical role in the cortically evoked long-term potentiation (Charpier and Deniau, 1997; Malenka and Nicoll, 1993; Partridge et al., 2000). The present finding that NMDA receptors are involved in the glutamatergic regulation of dopamine release suggests the functional relation between the cortically evoked long-term potentiation and dopaminergic system in the dorsolateral striatum.

Although morphological studies showed close proximity of dopaminergic and glutamatergic neurons in the dorsolateral striatum (Smith and Bolam, 1990), the precise location of NMDA receptors is not clear. In this study, we observed that direct perfusion of NMDA (1 mM) into the striatum produced increases in both dopamine and glutamate levels to approximately 2.5-fold. These findings indicate the possibility that glutamate acting at NMDA receptors can stimulate dopamine neurons, in turn, the stimulation of glutamatergic projection may cause increases in dopamine release via NMDA receptors, which are located on dopamine nerve terminals. Further studies, however, are required about the precise location of NMDA receptors underlying the prefrontal cortex regulation of dopamine release.

It should be noted, furthermore, the present data do not exclude the possibility that dopamine release in the dorsolateral striatum is indirectly regulated by afferents from the prefrontal cortex. Considering the extensive connectivity of the prefrontal cortex with cortical and subcortical regions, alterations in the activity of the prefrontal cortex afferents would affect the function of another cortical region and might consequently influence the striatal dopaminergic system. Interestingly, however, a recent imaging study (Strafella et al., 2001) showed that dopamine release in the dorsal striatum was increased when the repetitive transcranial magnetic stimulation, which is considered to exert therapeutic effects in Parkinson's diseases and some psychiatric disorders (Gerschlager et al., 2001; Grunhaus et al., 2000), was applied to the prefrontal cortex. It has been proposed that the repetitive transcranial magnetic stimulation treatments directly affect the corticostriatal projection and consequently stimulate dopamine neurons in the ipsilateral striatum (Strafella et al., 2001). In other words, dopaminergic neuron in the dorsolateral striatum would be regulated directly by afferents from the prefrontal cortex through the corticostriatal pathway.

In conclusion, the present study revealed that the prefrontal cortex functionally regulates dopamine release in the dorsolateral striatum, which is mediated via $GABA_A$ receptors. The cortical GABAergic neurons underlying the prefrontal cortex regulation appear to be associated with the corticostriatal pathway. In other words, GABAergic system in the cortical circuitry would play an important role in not only the regulation of dopamine release but also the physiological function of the dorsolateral striatum.

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