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Dopaminergic and cholinergic stimulation of the ventrolateral striatum elicit rat jaw movements that are funnelled via distinct efferents

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Abstract

It has been reported that two distinct types of jaw movements can be elicited by bilateral injections of drugs into the ventrolateral striatum: (1) dopamine receptor-mediated jaw movements that are elicited by a mixture of (±)-6-chloro-7,8-dihydroxy-3-allyl-1-phenyl-2,3,4,5-tetrahydro-1*H*-3-benzazepine-7,8-diol (SKF 82958; 5 μg) and quinpirole (10 μg), and (2) acetylcholine receptor-mediated jaw movements that are elicited by carbachol (2.5 μg). In the present study, electromyographic analysis was used to characterise these movements: the dopamine receptor-mediated jaw movements were marked by a dominant digastric activity during jaw opening and a dominant masseter activity during jaw closing (digastric/masseter type), whereas the acetylcholine receptor-mediated jaw movements were marked by a dominant digastric activity during jaw opening without any significant change in masseter activity during jaw closing (digastric type). The main goal was to (in)validate the hypothesis that these two types of jaw movements are funnelled via distinct γ-aminobutyric acid (GABA)ergic output channels. Bilateral injections of both muscimol (25 and 50 ng/0.2 μl per side) and bicuculline (50 and 150 ng/0.2 μl per side) into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata essentially inhibited dopamine receptor-mediated jaw movements to various degrees. In contrast, acetylcholine receptor-mediated jaw movements were inhibited by muscimol given into the entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata, whereas these movements were enhanced by bicuculline. The acetylcholine receptor-mediated jaw movements were not affected by muscimol injections into the ventral pallidum, but were inhibited by bicuculline injections. Studies on such injections into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata of naive rats revealed that jaw movements of the digastric/masseter type were elicited either by muscimol injections into the dorsolateral part of the substantia nigra pars reticulata or by combined injections of muscimol and bicuculline into the entopeduncular nucleus, and that jaw movements of the digastric type were elicited only by combined injections of muscimol and bicuculline into the entopeduncular nucleus. Together, the data allow the conclusion that dopamine receptor-mediated and acetylcholine receptor-mediated jaw movements are two distinct types of jaw movements that are funnelled via separate GABAergic output channels. It is suggested that the three different profiles of responses to GABAergic drugs in animals showing either dopamine receptor-mediated or acetylcholine receptor-mediated jaw movements reflect the involvement of three distinct types of output neurons of the striatum, namely: type I neurons with collateralised axons to the ventral pallidum, entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata, mediating the dopamine receptor-mediated jaw movements; type II neurons with collateralised axons to the globus pallidus that, in turn, project to the entopeduncular nucleus and the dorsolateral part of the substantia nigra pars reticulata, mediating directly the acetylcholine receptor-mediated jaw movements; and type III neurons with a single axon to the ventral pallidum, mediating indirectly the acetylcholine receptor-mediated movements. It is evident that future studies are required to provide direct evidence in favour of the latter hypothesis.

Keywords: Jaw movement; Ventrolateral striatum; Striatal collateralised output pathway; Dopamine receptor; Acetylcholine receptor; $GABA_A$ receptor; (Rat)

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1. Introduction

One of the central tenets of the basal ganglia model is segregation of the striatal output pathways. Early retrograde cell labelling studies have supported the idea of distinct striatofugal projections arising from separate neuronal populations in the striatum (Fibiger and Crossman, 1984; Parent et al., 1984; Loopuijt and van der Kooy, 1985; Beckstead and Cruz, 1986). In the widely accepted scheme of basal ganglia organisation, the striatum is thought to exert its effect via a direct and an indirect pathway (Albin et al., 1989; DeLong, 1990; Gerfen and Wilson, 1996; Wichmann and DeLong, 1996; Smith et al., 1998). The direct pathway originates from γ-aminobutyric acid (GABA)ergic neurons that project monosynaptically upon neurons of the entopeduncular nucleus and substantia nigra pars reticulata, and the indirect pathway arises from GABAergic neurons that project polysynaptically to entopeduncular nucleus/substantia nigra pars reticulata via relays in the globus pallidus and the subthalamic nucleus; each of the pathways is thought to serve a particular function. However, recent single-cell labelling study in rats revealed the existence of striatal projection neurons with highly collateralised axons that provide branches to two or three of the striatal recipient structures. For instance, Wu et al. (2000) provided evidence that there exist three types of neurons in the striatum, i.e. (1) neurons that project to the globus pallidus, entopeduncular nucleus and substantia nigra pars reticulata, (2) neurons that project to the globus pallidus and substantia nigra pars reticulata and (3) neurons that project only to the globus pallidus. This finding suggests that each of these may serve its own function.

Previously, activation of dopamine or acetylcholine receptors in the ventrolateral part of the striatum has been found to elicit repetitive jaw movements in rats (Kelley et al., 1989; Delfs and Kelley, 1990), each of these marked by its own movement profile (Kikuchi de Beltrán et al., 1992). These behavioural findings have led to the hypothesis that the dopamine receptor-mediated jaw movements are mediated via mechanisms or substrates that differ from those involved in the acetylcholine receptor-mediated jaw movements.

GABAergic neurons arising from the ventrolateral striatum primarily converge onto neurons in the dorsolateral part of the substantia nigra pars reticulata (Von Krosigk et al., 1992; Iwata et al., 1996) and in the entopeduncular nucleus (Parent, 1990; Takada et al., 1994) which in turn project directly, or indirectly via the superior colliculus, to the parvicellular reticular formation (Yasui et al., 1992, 1994, 1995; Takada et al., 1994), a region that is directly connected with the orofacial motor nuclei. The ventral pallidum, a ventral portion of the globus pallidus, is also known to receive GABAergic input from the striatum, especially the ventral striatum encompassing the nucleus accumbens (Groenewegen and Russschen, 1984; Zahm and Brog, 1992).

In the first part of this study, we used electromyography and a phototransduction system to characterise (dis)similarities between the two types of jaw movements that are elicited by injections of dopamine and acetylcholine receptor agonists into the ventrolateral striatum. The second part of this study dealt with the hypothesis that the dopamine receptor-mediated and acetylcholine receptor-mediated jaw movements are funnelled via distinct GABAergic output channels. For that purpose, the agonist and the antagonist of $GABA_A$ receptors, muscimol and bicuculline, respectively, were injected bilaterally into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata of rats treated with bilateral injections into the ventrolateral striatum of a mixture of the dopamine D1 receptor agonist, (±)-6-chloro-7,8-dihydroxy-3-allyl-1-phenyl-2,3,4,5-tetrahydro-1*H*-3-benzazepine-7,8-diol (SKF 82958; 5 μg), and the dopamine D2 receptor agonist, quinpirole (10 μg), or the acetylcholine receptor agonist, carbachol (2.5 μg); the latter dopaminergic and cholinergic treatments have been found to be highly effective to elicit the above-mentioned dopamine receptor-mediated and acetylcholine receptor-mediated jaw movements (Kikuchi de Beltrán et al., 1992).

2. Materials and methods

2.1. Surgical procedures

Male Sprague–Dawley rats weighing 260–330 g were housed in cages (27 × 45 × 20 cm) that were kept at constant room temperature (23 ± 2 °C) and relative humidity (55 ± 5%) under a 12-h light/dark cycle (lights on at 0700 h), with free access to food and water.

Rats were anaesthetised with halothane (0.5–4.0% when appropriate) and supplemented with ketamine HCl (10.0 mg/kg, i.p.). The surgical and recording procedures were as described previously (Koshikawa et al., 1989, 1990a,b, 1991; Cools et al., 1995). After cannulation of the right external jugular vein, a small light-emitting diode was fixed to the mandible. The animal was placed in a stereotactic frame so that the head was kept in constant relation to a light-sensitive transducer, which detected the vertical movements of the diode. Bipolar electrodes were placed into the masseter and digastric muscles to record electromyographic (EMG) activity. After surgery, the animals continuously received ketamine in a dose (10.0 mg/kg, i.v.) unable to influence either the jaw movements under study (Koshikawa et al., 1989) or dopamine metabolism in the striatum (Koshikawa et al., 1988). Lignocaine HCl (2% gel) was applied to all incisions to ensure complete analgesia. Rectal temperature was maintained at 37.0 °C with a thermostatically controlled heating pad. Monitored concentrations of expired O_2 and CO_2 during the experiment were 19–21% and 2.0–2.5%, respectively. The jaw movements and EMG activity were recorded on an eight-channel tape recorder

(RD-180T; TEAC) for off-line analyses. The recordings were analysed automatically, using a spike trigger that counted vertical jaw movements per 5 min, and the EMG data were full-wave rectified. Averaging of the vertical output of the movement transducer and jaw muscle EMG activity was performed with a computer system (signal processor 1000; NEC). The recording period lasted 60 or 120 min. The recorded EMG activity and movements of the mandible were averaged in order to establish the relationship between EMG activity and the movements more clearly.

Guide cannulas (0.5 mm o.d., 0.3 mm i.d., 6.0 mm length) were implanted bilaterally into the brain according to previously described procedures (Koshikawa et al., 1989). The coordinates based on the atlas of Paxinos and Watson (1986) were: anterior = 8.6 mm, vertical = 3.0 mm, lateral = 4.0 mm (ventrolateral striatum); anterior = 8.7 mm, vertical = 2.2 mm, lateral = 2.0 mm (ventral pallidum); anterior = 6.7 mm, vertical = 2.0 mm, lateral = 2.8 mm (entopeduncular nucleus); anterior = 3.8 mm, vertical = 2.0 mm, lateral = 3.0 mm (dorsolateral part of the substantia nigra pars reticulata). The injection was made slowly in a volume of 0.2 μl per side over 20 s, and the needle was left in situ for an additional 20-s period after completion of the injection. Damage to the target site was minimised by implanting the tips of the guide cannulas 1.6 mm (ventrolateral striatum), 2.2 mm (dorsolateral part of the substantia nigra pars reticulata; entopeduncular nucleus) or 2.0 mm (ventral pallidum) above the desired injection site. Wire stylets were placed in the guide cannulas to prevent occlusion.

These experiments were approved by the Animal Experimentation Committee of Nihon University School of Dentistry, and were performed in accordance with Institutional guidelines for the care and use of experimental animals that were in compliance with the UK Animals Scientific Act 1986.

2.2. Drugs

The animals ($n = 6–8$ per experiment) received bilateral injections of the full dopamine D1 receptor agonist, (±)-6-chloro-7,8-dihydroxy-3-allyl-1-phenyl-2,3,4,5-tetrahydro-1*H*-3-benzazepine-7,8-diol hydrobromide (SKF 82958; 5 μg; Research Biochemicals International), and the dopamine D2 receptor agonist, quinpirole (10 μg; Research Biochemicals International), combination (cocktail) or the nonselective acetylcholine receptor agonist, carbachol (2.5 μg; carbamylcholine, Sigma), into the ventrolateral striatum. The $GABA_A$ receptor agonist, muscimol (25 or 50 ng; 5-aminomethyl-3-hydroxyisoxazole, Sigma), or the antago-

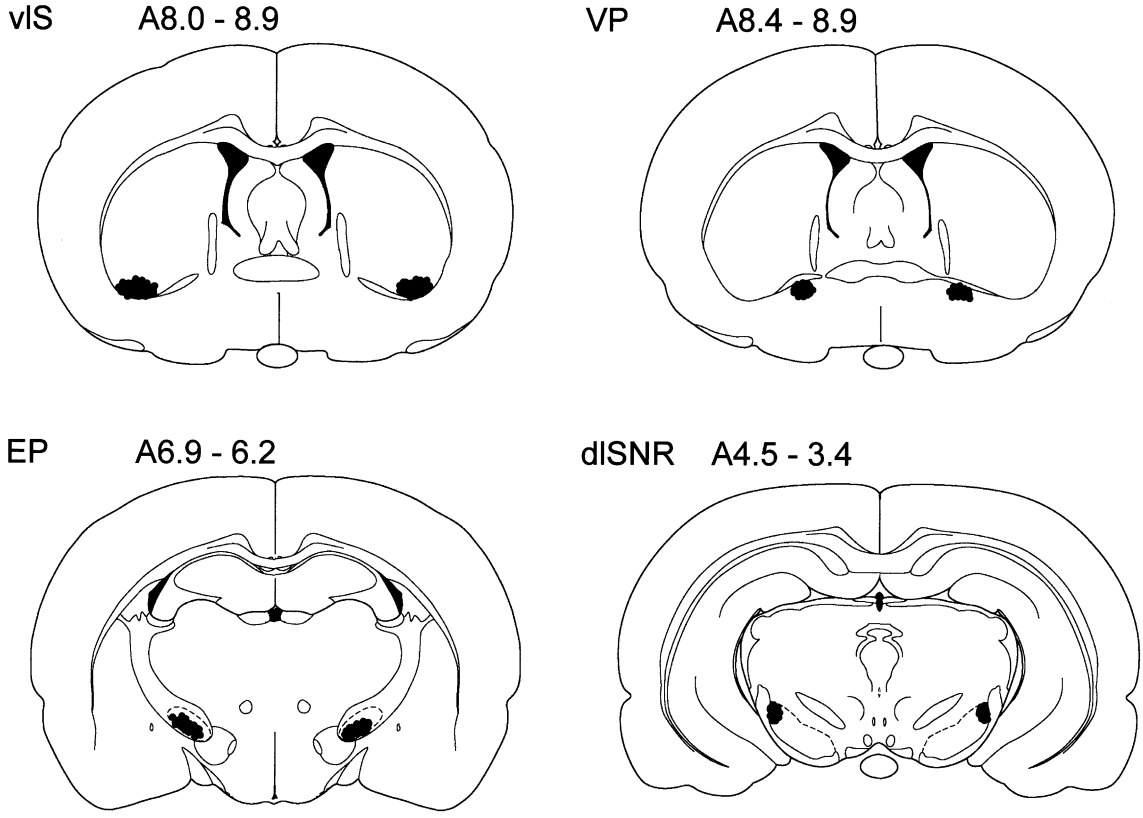

Fig. 1. Location of injection sites in the ventrolateral striatum (vlS, upper left), ventral pallidum (VP, upper right), entopeduncular nucleus (EP, lower left) and dorsolateral part of the substantia nigra pars reticulata (dlSNR, lower right). Planes are modified to a series of two or three sections for each brain area from the atlas of Paxinos and Watson (1986); approximate coordinates indicated are in mm anterior to the interaural line.

nist, (−)-bicuculline methylbromide (50 or 150 ng; Research Biochemicals International), was injected into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata immediately before the injection of the mixture of SKF 82958 and quinpirole or carbachol into the ventrolateral striatum. All drugs were dissolved in saline immediately before use. Doses employed were based on previously published studies (Koshikawa et al., 1990a; Kikuchi de Beltrán et al., 1992; Cools et al., 1995). The animals were used only once.

2.3. Histology

At the end of each experiment, the rats were deeply anaesthetised with sodium pentobarbitone (80 mg/kg, i.p.) and perfused transcardially with 10% formalin. The brains were removed, sectioned at 50 μm and stained with Cresyl violet to visualise the injection site; only data from animals in which the injections were correctly placed were included in subsequent analyses. Fig. 1 gives a survey of the injection sites located in the ventrolateral striatum, ventral pallidum, entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata.

2.4. Data analysis

All values are expressed as means ± S.E.M. and analysed using a two-way (group × time) analysis of variance (ANOVA), where appropriate. A probability value of $P < 0.05$ was considered statistically significant.

3. Results

3.1. Difference in pattern of jaw movements elicited by dopamine and acetylcholine receptor agonists

Fig. 2 shows the results of the comparison of the averaged EMG activity recorded from digastric and masseter muscle and the vertical component of jaw movements. After bilateral injections of the mixture of SKF 82958 (5 μg) and quinpirole (10 μg) into the ventrolateral striatum (Fig. 2, left panel), increased EMG activity in the digastric and masseter muscles was associated with movements of both jaw opening and closing: digastric activity was dominant during jaw opening, and masseter activity was dominant during jaw closing (digastric/masseter type). After bilateral injections of carbachol (2.5 μg) into the ventrolateral striatum (Fig. 2, right panel), increased EMG activity in the digastric muscle was associated with the movement of jaw opening, whereas the masseter muscle maintained a stable moderate tone of EMG activity, which did not change during jaw movements (digastric type).

3.2. Effects of bilateral injections of muscimol and bicuculline into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata on production of jaw movements

Injections of muscimol (50 ng; $n = 7$) and bicuculline (150 ng; $n = 6$), alone or in combination ($n = 7$), into the ventral pallidum did not essentially elicit repetitive jaw

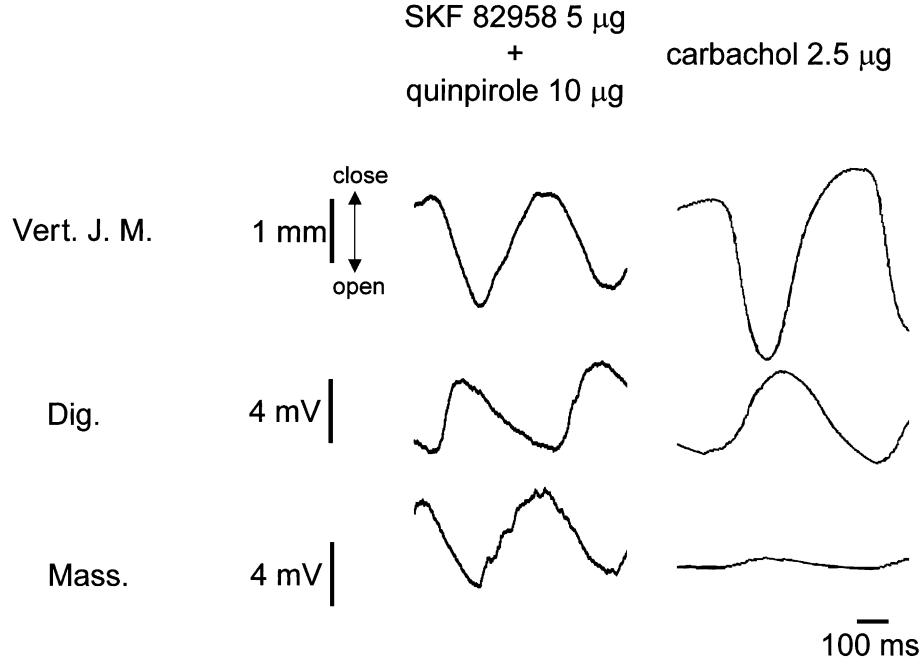

Fig. 2. Difference in pattern of jaw movements elicited by dopamine D1/D2 receptor and acetylcholine receptor stimulation in the ventrolateral striatum. For dopamine D1/D2 receptor stimulation, injection of a mixture of SKF 82958 (5 μg) and quinpirole (10 μg) was used and acetylcholine receptor stimulation was with the injection of carbachol (2.5 μg). Averaged values of 10 consecutive vertical component of jaw movements (Vert. J.M.) and associated electromyographic (EMG) activity recorded from digastric (Dig.) and masseter (Mass.) muscles are shown.

movements (Fig. 3, top). Injections of bicuculline ($n=15$) into the entopeduncular nucleus elicited a small number of jaw movements, whereas muscimol ($n=7$) remained without any effect. Combination of the two drugs ($n=12$) induced prominent short-lasting (30 min) jaw movements (Fig. 3, middle); analysis of the movement pattern revealed that 7 out of 12 rats displayed jaw movements of the digastric type and that the remaining 5 rats displayed jaw movements of digastric/masseter type. When given into the dorsolateral part of the substantia nigra pars reticulata, muscimol (50 ng; $n=8$) readily induced repetitive jaw movements of the digastric/masseter type. The effects of muscimol were completely abolished by bicuculline (150 ng; $n=7$) that itself ($n=6$) did not elicit jaw movements (Fig. 3, bottom).

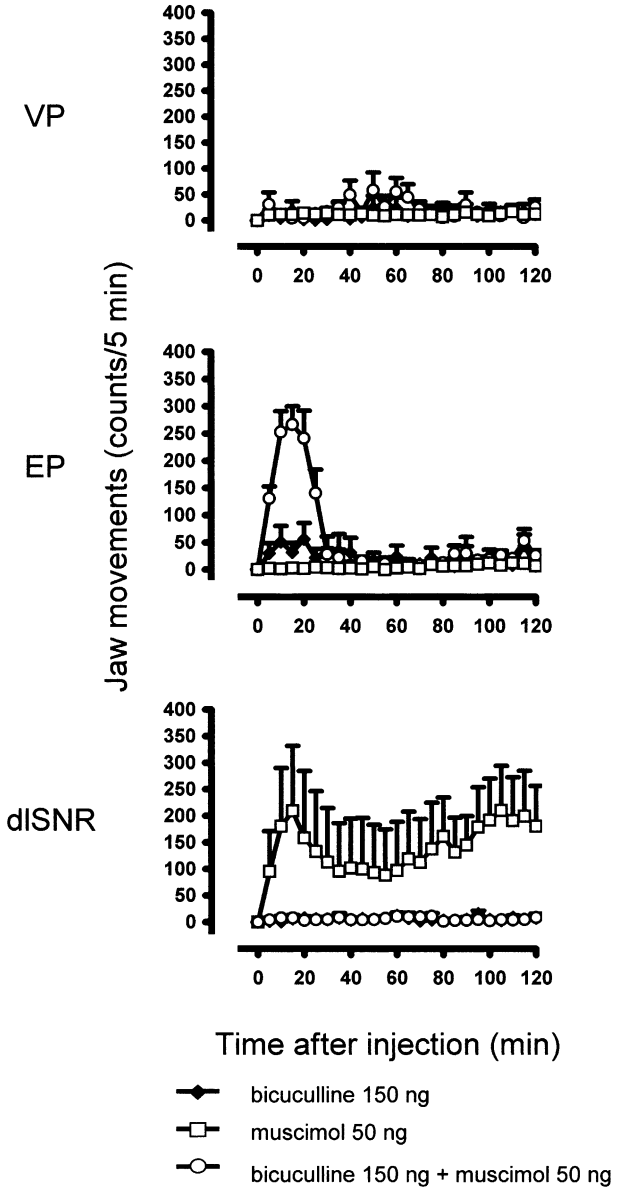

Fig. 3. The time-dependent effects of bilateral injections of saline (0.2 μl/side), muscimol (50 ng/side), bicuculline (150 ng/side) or the mixture of muscimol (50 ng/side) and bicuculline (150 ng/side) into the ventral pallidum (VP, top), entopeduncular nucleus (EP, middle) or dorsolateral part of the substantia nigra pars reticulata (dlSNR, bottom) on production of jaw movements. The data are expressed as the mean number of jaw movements occurring in 5-min observation periods ($n=6-15$). Vertical bars indicate S.E.M.

3.3. Effects of bilateral injections of muscimol into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata on jaw movements elicited by administration of SKF 82958 and quinpirole mixture into the ventrolateral striatum

The time-dependent effects of the mixture of SKF 82958 and quinpirole are shown in Fig. 4. This figure illustrates that the jaw movements occurred initially at low levels but subsequently increased markedly and then declined over the 120-min observation period. When given into the ventral pallidum, muscimol produced a dose-dependent inhibition of the jaw movements elicited by the mixture of SKF 82958 and quinpirole injected into the ventrolateral striatum (25 ng vs. control: $F(1,406)=53.68$, $P<0.0001$, $n=8$; 50 ng vs. control: $F(1,358)=184.89$, $P<0.0001$, $n=6$; saline: $n=9$; Fig. 4, left part, top); in the entopeduncular nucleus, muscimol produced complete inhibition (25 ng vs. control: $F(1,334)=129.43$, $P<0.0001$, $n=7$; 50 ng vs. control: $F(1,286)=119.4$, $P<0.0001$, $n=6$; saline: $n=6$; Fig. 4, left part, middle). Injections of muscimol into the dorsolateral part of the substantia nigra pars reticulata dose dependently suppressed the jaw movements elicited by the mixture at 30–90 min after injection (25 ng vs. control: $F(1,180)=5.72$, $P<0.02$, $n=7$; 50 ng vs. control: $F(1,193)=23.06$, $P<0.0001$, $n=8$; saline: $n=7$; Fig. 4, left part, bottom).

3.4. Effects of bilateral injections of bicuculline into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata on jaw movements elicited by administration of SKF 82958 and quinpirole mixture into the ventrolateral striatum

Injections of bicuculline into the ventral pallidum produced a dose-dependent but short-lasting (until 60 min) inhibition of the jaw movements elicited by the SKF 82958 and quinpirole mixture (0–60 min: 50 ng vs. control, $F(1,178)=11.29$, $P<0.001$, $n=6$; 150 ng vs. control, $F(1,178)=32.30$, $P<0.0001$, $n=6$; Fig. 4, right part, top). When given into the entopeduncular nucleus or the dorsolateral part of the substantia nigra pars reticulata, bicuculline dose dependently inhibited the jaw movements elicited by the mixture injected into the ventrolateral striatum, respectively (entopeduncular nucleus: 50 ng vs. control, $F(1,334)=9.47$, $P<0.003$, $n=8$; 150 ng vs. control: $F(1,310)=203.57$, $P<0.0001$, $n=7$; dorsolateral part of the substantia nigra pars reticulata: 50 ng vs. control, $F(1,310)=53.80$, $P<0.0001$, $n=6$; 150 ng vs. control, $F(1,334)=$

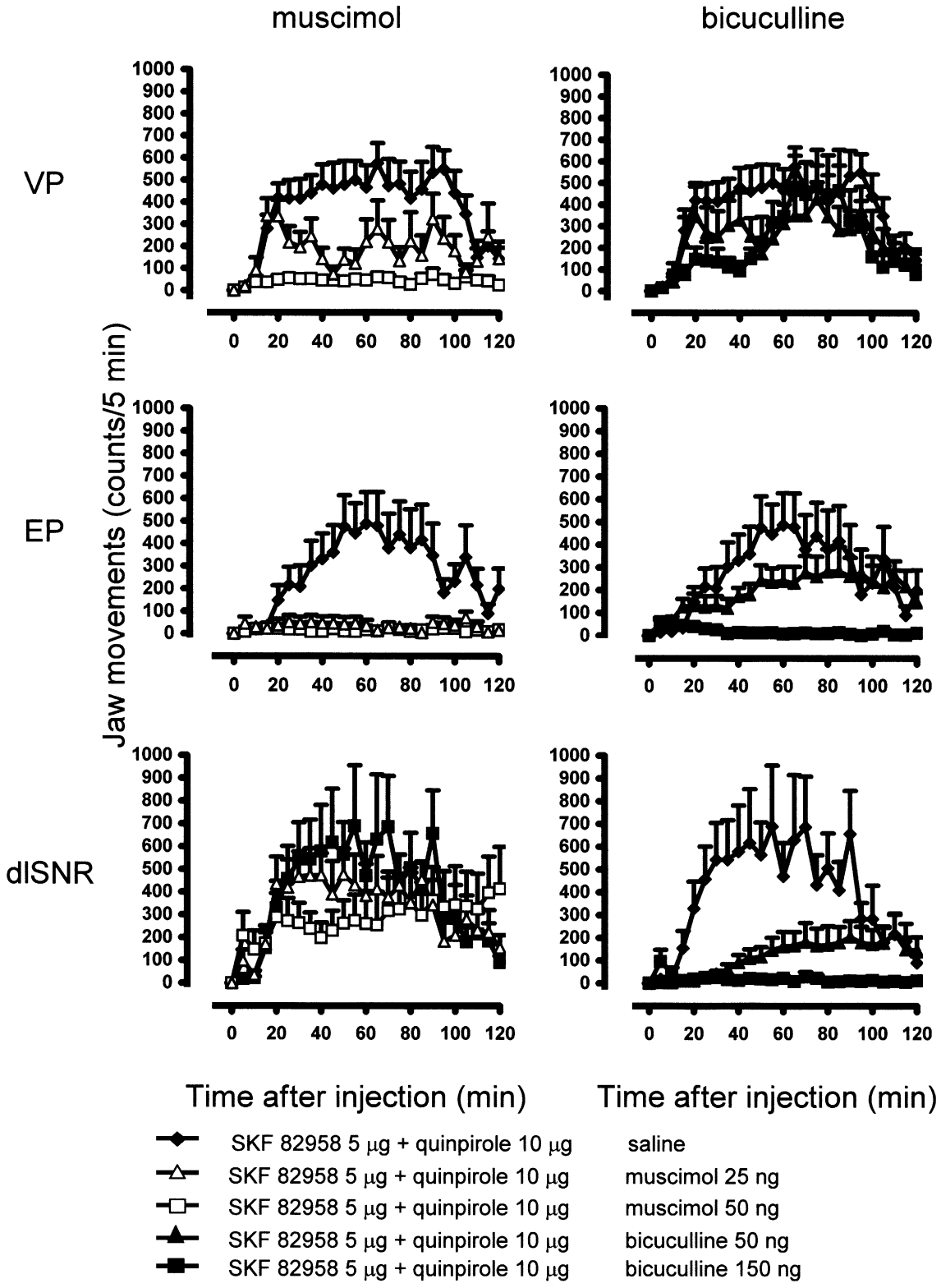

Fig. 4. Effects of muscimol (25 and 50 ng/side, left part) or bicuculline (50 and 150 ng/side, right part) injection into the ventral pallidum (VP, top), entopeduncular nucleus (EP, middle) or dorsolateral part of the substantia nigra pars reticulata (dlSNR, bottom) on production of jaw movements induced by the mixture of SKF 82958 (5 μg) and quinpirole (10 μg) given into the ventrolateral striatum (vlS). The data are expressed as the mean number of jaw movements occurring in 5-min observation periods ($n = 6–9$). Vertical bars indicate S.E.M.

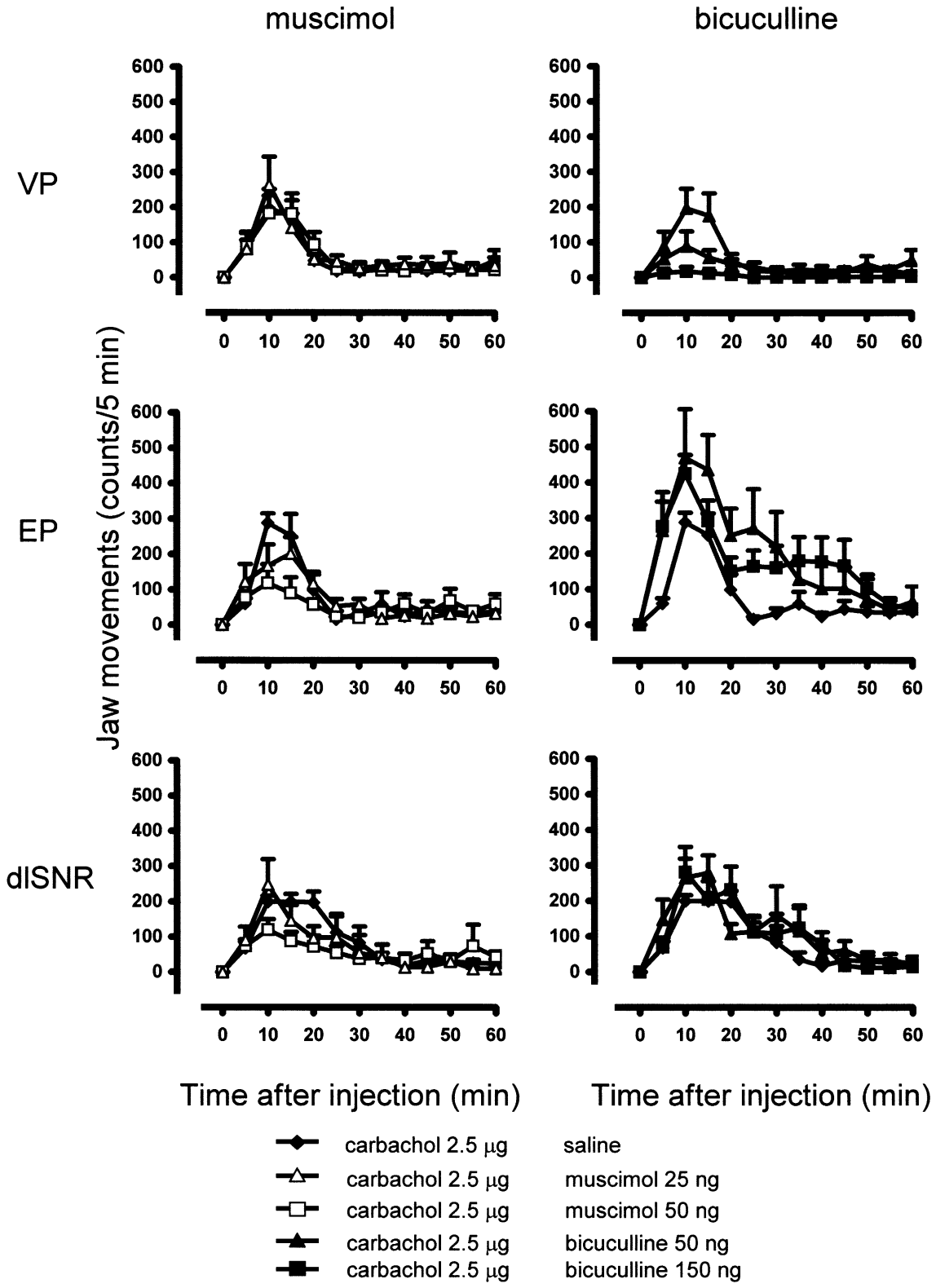

Fig. 5. Effects of muscimol (25 and 50 ng/side, left part) or bicuculline (50 and 150 ng/side, right part) injection into the ventral pallidum (VP, top), entopeduncular nucleus (EP, middle) or dorsolateral part of the substantia nigra pars reticulata (dlSNR, bottom) on production of jaw movements induced by carbachol (2.5 μg) given into the ventrolateral striatum (vlS). The data are expressed as the mean number of jaw movements occurring in 5-min observation periods ($n = 6–7$). Vertical bars indicate S.E.M.

125.85, $P<0.0001$, $n=7$; Fig. 4, right part, middle and bottom).

3.5. Effects of bilateral injections of muscimol into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata on jaw movements elicited by administration of carbachol into the ventrolateral striatum

The time-dependent effects of carbachol are shown in Fig. 5. This figure clearly illustrates that the effect started nearly immediately after injection, reached its peak around 10–25 min and vanished about 40 min after injection. When given into the ventral pallidum, muscimol did not affect the jaw movements elicited by carbachol into the ventrolateral striatum (0–30 min: 25 ng vs. control, $F(1,66)=0.28$, $P=0.596$, $n=6$; 50 ng vs. control, $F(1,66)=0.15$, $P=0.702$, $n=6$; saline: $n=7$, Fig. 5, left part, top). Injections of muscimol into the entopeduncular nucleus and the dorsolateral part of the substantia nigra pars reticulata produced a small but significant inhibition of the jaw movements elicited by carbachol (0–30 min: entopeduncular nucleus, 25 ng vs. control, $F(1,66)=0.05$, $P=0.817$, $n=6$; 50 ng vs. control, $F(1,66)=8.70$, $P<0.005$, $n=7$; saline: $n=6$; dorsolateral part of the substantia nigra pars reticulata: 25 ng vs. control, $F(1,66)=0.54$, $P=0.466$, $n=6$; 50 ng vs. control, $F(1,72)=19.88$, $P<0.0001$, $n=7$; saline: $n=7$; Fig. 5, left part, middle and bottom).

3.6. Effects of bilateral injections of bicuculline into the ventral pallidum, entopeduncular nucleus or dorsolateral part of the substantia nigra pars reticulata on jaw movements elicited by administration of carbachol into the ventrolateral striatum

Injections of bicuculline bilaterally into the ventral pallidum produced a dose-dependent inhibition of the jaw movements induced by carbachol (0–30 min: 50 ng vs. control, $F(1,66)=4.80$, $P<0.05$; $n=6$; 150 ng vs. control, $F(1,66)=20.82$, $P<0.0001$; saline: $n=6$; Fig. 5, right part, top). In contrast, bicuculline significantly enhanced the jaw movements elicited by injections of carbachol into the ventrolateral striatum (50 ng vs. control: $F(1,142)=16.64$, $P<0.0001$; $n=6$; 150 ng vs. control: $F(1,142)=19.57$, $P<0.0001$; $n=6$; Fig. 5, right part, middle) when given into the entopeduncular nucleus, and less so in the dorsolateral part of the substantia nigra pars reticulata (50 ng vs. control: $F(1,166)=2.75$, $P=0.099$; $n=7$; 150 ng vs. control: $F(1,154)=12.12$, $P<0.001$; $n=6$; Fig. 5, right part, bottom).

4. Discussion

The present study clearly demonstrated that two distinct types of jaw movements can be elicited by bilateral injections of drugs into the ventrolateral striatum: (1) dopamine receptor-mediated jaw movements that are elicited by a mixture of SKF 82958 (5 μg) and quinpirole (10 μg), and (2) acetylcholine receptor-mediated jaw movements that are elicited by carbachol (2.5 μg). The dopamine receptor-mediated jaw movements are characterised by a dominant digastric activity during jaw opening and a dominant masseter activity during jaw closing (digastric/masseter type), whereas the acetylcholine receptor-mediated jaw movements are characterised by a dominant digastric activity during jaw opening without any significant change in masseter activity during jaw closing (digastric type). These results confirm previously reported findings (Kikuchi de Beltrán et al., 1992). The present study provides evidence that these two types of jaw movements are funnelled via distinct GABAergic output channels of the ventrolateral striatum. This evidence is discussed below.

4.1. Effects of GABAergic drugs on jaw movements of digastric/masseter type

The first observation of note concerns the finding that bilateral injections of both the $GABA_A$ receptor agonist, muscimol, and the $GABA_A$ receptor antagonist, bicuculline, into the ventral pallidum, entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata inhibited to various degrees the dopamine receptor-mediated jaw movements that were elicited from the ventrolateral striatum. These effects were not as specific, because they were dose-dependent and injections of the solvent of these drugs into the regions under study did not produce any inhibition. The most likely explanation is that the regions under study are heterogeneous, encompassing an area sensitive to the $GABA_A$ receptor agonist because the tonus of its GABAergic input is low, and an area sensitive to the $GABA_A$ receptor antagonist because the tonus of its GABAergic input is high. Although such heterogeneity has not been described for regions such as the dorsolateral part of the substantia nigra pars reticulata, it is well known for pallidal regions such as the ventral pallidum and the entopeduncular nucleus (Filion et al., 1994; Matsumura et al., 1995). According to the latter authors, pallidal segments are marked by a centre-surround organisation, with hypoactivity in the peripheral outer part and hyperactivity in the central inner part. In fact, they demonstrated that there is a topological antagonistic centre-surround organization, providing a topological arrangement that allows an informational convergence on a limited number of pallidal neurons in the centre. Such an organisation implies that the GABAergic input is high in the peripheral outer part, but low in the central inner part, or vice versa. Considering our above-mentioned data in the context of these findings, it is suggested that the dopaminergic treatment reduced the tonus of the GABAergic input of the peripheral outer parts of all three regions examined with the consequence that it indirectly enhanced the GABAergic tonus at the level of the

output neurons in the central inner part, resulting in a hypoactive output of the region under study. In this manner, the target structures, namely the orofacial motor nuclei, are released from tonic inhibition and, accordingly, give rise to the initiation of jaw movements (cf. Hikosaka and Wurtz, 1985). According to this model, bicuculline inhibited the dopamine receptor-mediated jaw movements because it inhibited the high GABAergic input of the inner part, and muscimol inhibited these jaw movements because it enhanced the GABAergic input in the peripheral outer part. Comparing the effectiveness of the drugs in the three distinct regions gives the following rank order for muscimol's potency: ventral pallidum > entopeduncular nucleus> dorsolateral part of the substantia nigra pars reticulata, whereas the reverse rank order is seen for bicuculline. According to the above-mentioned model, these data imply that the ratio outer part/inner part increases from the ventral pallidum, entopeduncular nucleus to dorsolateral part of the substantia nigra pars reticulata.

It is evident that the above-mentioned explanation of the present findings is open to discussion. Despite this, the data available clearly show that (1) all three regions are heterogeneous as far as concerns the GABAergic input, and that (2) all three regions directly and/or indirectly modulate dopamine receptor-mediated jaw movements that are elicited from the ventrolateral striatum (see Fig. 6).

4.2. Output stations of dopamine receptor-mediated jaw movements

The experiments with the GABAergic drugs given to naive rats revealed that two treatments were able to elicit jaw movements of the digastric/masseter type: (1) bilateral injections of muscimol into the dorsolateral part of the substantia nigra pars reticulata and (2) bilateral injections of muscimol plus bicuculline into the entopeduncular nucleus. The effects of muscimol injections into the dorsolateral part of the substantia nigra pars reticulata were GABA-specific, because these were well inhibited by bicuculline. According to the above-mentioned model, muscimol enhanced the GABAergic input of the central inner part of the dorsolateral part of the substantia nigra pars reticulata with the result that it inhibited the firing of the efferents. The ability of intra-nigrally administered muscimol to elicit jaw movements of the digastric/masseter type and the above-mentioned data about the ability of intra-nigrally injected GABAergic drugs to inhibit the dopamine receptor-mediated jaw movements that were elicited from the ventrolateral striatum allow the conclusion that the dorsolateral part of the substantia nigra pars reticulata has to be considered as an important station that mediates dopamine receptor-mediated jaw movements elicited from the ventrolateral striatum (see Fig. 6). Because jaw movements of the digastric/masseter

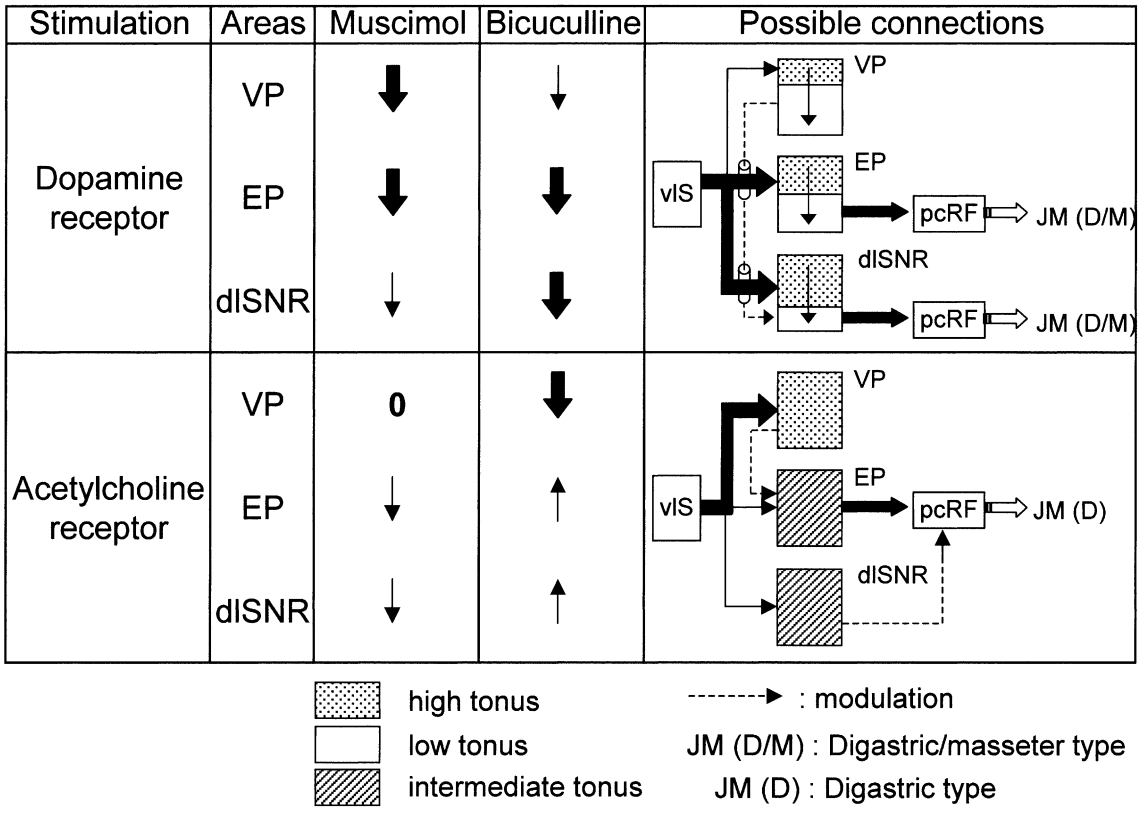

Fig. 6. Summary of overall effects of local injections of muscimol and bicuculline on dopamine and acetylcholine receptor-mediated jaw movements (JM), and possible connections from the ventrolateral striatum (vlS). VP: ventral pallidum; EP: entopeduncular nucleus; dlSNR: dorsolateral part of the substantia nigra pars reticulata; pcRF: parvicellular reticular formation.

type could be elicited from the entopeduncular nucleus as well, this region should also be considered as a station that can mediate dopamine receptor-mediated jaw movements (see Fig. 6). It was the combined intra-entopeduncular injection of both muscimol and bicuculline that elicited jaw movements of the digastric/masseter type in naive rats. According to the above-mentioned model, muscimol enhanced the GABAergic input of the central inner part, whereas bicuculline inhibited the GABAergic input of the peripheral outer part. It is consistent with this notion that intra-entopeduncular injections of bicuculline alone also elicited jaw movements of the digastric/masseter type. The finding that such injections of muscimol alone were unable to elicit jaw movements of the digastric/masseter type in naive rats might be ascribed to the fact that muscimol has a very restricted ability to diffuse (cf. Arnt et al., 1979) and, therefore, could not spread throughout the central inner part of the entopeduncular nucleus; this problem does not apply to the dorsolateral part of the substantia nigra pars reticulata because the latter structure is far smaller than the entopeduncular nucleus. Finally, none of the GABAergic injections into the ventral pallidum of naive rats elicited jaw movements of the digastric/masseter type, thus excluding the possibility that the ventral pallidum has to be considered as a station that mediates dopamine receptor-mediated jaw movements. As mentioned above, intra-pallidal injections of bicuculline, however, did inhibit dopamine receptor-mediated jaw movements. This finding becomes understandable in view of the fact that neurons of the ventral pallidum are known to exert an inhibitory effect on cells in the dorsolateral part of the substantia nigra pars reticulata (Maurice et al., 1997). According to the above-mentioned model, we speculate that the efferents of the ventral pallidum project onto cells in the central inner part of the dorsolateral part of the substantia nigra pars reticulata that, in turn, inhibited the output neurons of the dorsolateral part of the substantia nigra pars reticulata. The finding that intra-pallidal injections of muscimol did not elicit the digastric/masseter jaw movements might be ascribed to the fact that muscimol has a very restricted ability to diffuse and, therefore, could not spread throughout the rather large inner part of the ventral pallidum.

4.3. Effects of GABAergic drugs on jaw movements of the digastric type

The effects of GABAergic drugs on acetylcholine receptor-mediated jaw movements were similar for injections into the entopeduncular nucleus and the dorsolateral part of the substantia nigra pars reticulata, but different from those of such injections into the ventral pallidum. Injections of muscimol into the entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata inhibited the acetylcholine receptor-mediated jaw movements, whereas injections of bicuculline into these structures potentiated the jaw movements. Apparently, the tonus of the GABAergic input of the entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata regions of these animals was neither high nor low, implying that the carbachol treatment produced effects in these regions that differed fully from those elicited by the dopaminergic treatment (see above). On the other hand, injections of bicuculline into the ventral pallidum inhibited the acetylcholine receptor-mediated jaw movements elicited from the ventrolateral striatum, whereas injections of muscimol into the ventral pallidum remained without effect. This points to a high GABAergic tonus in the region under study. However, it cannot be excluded that the lack of ability of muscimol to produce any change was due its limited diffusion (see above).

4.4. Output stations of jaw movements of the digastric type

The experiments in which the GABAergic drugs were given to naive rats revealed that only one treatment was able to elicit jaw movements of the digastric type, namely bilateral injections of the combination of muscimol and bicuculline into the entopeduncular nucleus. This implied that only the entopeduncular nucleus, but not the ventral pallidum and the dorsolateral part of the substantia nigra pars reticulata, might be a station that can mediate acetylcholine receptor-mediated jaw movements. The finding that intra-entopeduncular injections of muscimol and bicuculline inhibited or potentiated, respectively, acetylcholine receptor-mediated movements elicited from the ventrolateral striatum fits with this suggestion. The mechanisms that underlie these effects might be comparable to that underlying the ability of the combined injections of muscimol and bicuculline into the entopeduncular nucleus to elicit jaw movements similar to, but not identical to (see below) the digastric/masseter type of jaw movements (see above). Although neither the ventral pallidum nor the dorsolateral part of the substantia nigra pars reticulata is a station that can mediate acetylcholine receptor-mediated jaw movements (see above), injections of muscimol and bicuculline inhibited and potentiated, respectively, acetylcholine receptor-mediated jaw movements. The modulatory role of the ventral pallidum, however, becomes understandable in view of the fact that efferents of the ventral pallidum impinge upon cells in the entopeduncular nucleus (Bevan et al., 1997; Bolam and Smith, 1992) and, accordingly, can modify the function of the entopeduncular nucleus. Furthermore, it is known that efferents of both the dorsolateral part of the substantia nigra pars reticulata (Von Krosigk et al., 1992; Iwata et al., 1996) and the entopeduncular nucleus (Parent, 1990; Takada et al., 1994) project directly, or indirectly via the superior colliculus, to the parvicellular reticular formation (Yasui et al., 1992, 1994, 1995; Takada et al., 1994), a region that sends neurons to orofacial motor nuclei of rats. It is this circuitry that might underlie the ability of the dorsolateral part of the substantia nigra pars reticulata to modulate acetylcholine receptor-mediated movements that are funnelled via the entopeduncular nucleus (see Fig. 6).

4.5. Overall conclusion

The present study provided hard evidence that the two distinct types of jaw movements that are elicited from the ventrolateral striatum, namely the dopamine receptor-mediated digastric/masseter type and the acetylcholine receptor-mediated digastric type, are at least partly funnelled via distinct mechanisms and output channels of the ventrolateral striatum. First, both the dorsolateral part of the substantia nigra pars reticulata and entopeduncular nucleus were found to mediate the jaw movements of the digastric/masseter type, whereas only the entopeduncular nucleus was found to mediate the jaw movements of the digastric type. Although the mechanism in the entopeduncular nucleus that mediates the digastric/masseter type of jaw movements might be comparable to that mediating the digastric type of jaw movements, the actual pathways allow, at best, parallel processing in neuronal channels because of the different nature of each type of jaw movement. Overall, these results are consistent with the previously reported finding that the ventrolateral striatum sends GABAergic projections to both the dorsolateral part of the substantia nigra pars reticulata (Von Krosigk et al., 1992; Iwata et al., 1996) and the entopeduncular nucleus (Parent, 1990; Takada et al., 1994) that, in turn, project directly, or indirectly via the superior colliculus, to the parvicellular reticular formation (Yasui et al., 1992, 1994, 1995; Takada et al., 1994), a region that is directly connected with the orofacial motor nuclei of rats. Second, the GABAergic mechanisms in each of the three brain structures investigated modulated differentially the dopamine receptor-mediated and acetylcholine receptor-mediated jaw movements, as discussed above.

As a final remark, it is noteworthy that our study revealed three distinct profiles of responses to injections of the GABAergic agents in animals, showing either dopamine receptor-mediated or acetylcholine receptor-mediated jaw movements. The first pattern was characterised by an inhibiting effect of both muscimol and bicuculline: this pattern was seen after injections into the ventral pallidum, entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata of rats showing dopamine receptor-mediated jaw movements. The second pattern was characterised by an inhibiting effect of muscimol and a potentiating effect of bicuculline: this pattern was seen after injections into the entopeduncular nucleus and the dorsolateral part of the substantia nigra pars reticulata of rats showing acetylcholine receptor-mediated jaw movements. The third pattern was characterised by an inhibiting effect of bicuculline without any effect of muscimol: this pattern was seen after injections into the ventral pallidum in rats showing acetylcholine receptor-mediated jaw movements. Given the recent studies of Parent's group (Parent et al., 2000; Wu et al., 2000), it is highly attractive to suggest that the three distinct patterns reflect the involvement of three distinct types of output neurons of the striatum, namely: type I neurons with collateralised axons to the ventral pallidum, entopeduncular nucleus and dorsolateral part of the substantia nigra pars reticulata, mediating the dopamine receptor-mediated jaw movements; type II neurons with collateralised axons to the dorsolateral part of the substantia nigra pars reticulata and the globes pallidus that, in turn, project to the entopeduncular nucleus (Bolam and Smith, 1992), mediating directly the acetylcholine receptor-mediated jaw movements; and type III neurons with a single axon to the ventral pallidum, mediating indirectly the acetylcholine receptor-mediated movements. It is evident that future studies are required to provide direct evidence in favour of the latter hypothesis.

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