Effects of YM-14673, a thyrotropin-releasing hormone analogue, injected into the shell and the core of the nucleus accumbens on production of repetitive jaw movements in rats: Comparison with the effects of a dopamine D₁ and D₂ receptor agonist combination

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(Received 13 June and accepted 1 August 1997)

Abstract: The present study examined whether the shell and the core of the nucleus accumbens play a differential role in the display of YM-14673-induced jaw movements in rats. For that purpose the effects of YM-14673 were compared to those of a SKF 82958 and quinpirole combination, a dopamine D₁ and a D₂ receptor agonist respectively, that is known to functionally differentiate these two subregions of the nucleus. Consistent with the previous report, bilateral injections of a mixture of SKF 82958 (5 µg) and quinpirole (10 µg) into the shell of the nucleus accumbens produced repetitive jaw movements, whereas similar injections of the mixture into the core did not induce such an effect. In contrast, there was no regional difference in the effects of YM-14673 on the production of repetitive jaw movements. Thus, both bilateral injections of YM-14673 (0.1 or 1.0 µg) into the shell or the core produced similar repetitive jaw movements in a dose-related manner. Moreover, the pattern of oral movements induced by YM-14673 differed from that induced by the mixture of SKF 82958 and quinpirole; frequent tongue protrusions were evident in rats treated with the mixture but were not seen in YM-14673-treated rats. It therefore appears that, unlike the effects of the mixture of dopamine D₁ and D₂ receptor agonists, the effects of YM-14673 in the shell on the production of rat jaw movements do not differ from the effects of the compound in the core.

Key words: accumbal shell; accumbal core; repetitive jaw movements; YM-14673; dopamine D₁/D₂ receptor; rat.

Introduction

The thyrotropin-releasing hormone (TRH) analogue, N°-[(S)-4-oxo-2-azetidinyl]-carbonyl]-L-histidyl-L-prolinamide dihydrate (YM-14673), injected bilaterally into the nucleus accumbens elicits repetitive jaw movements in rats (1). The mechanisms giving rise to the display of jaw movements after intra-accumbal injections of YM-14673 remain unknown. However, stimulation of the dopamine D₁/D₂ receptors may at least partly contribute to these effects, since the non-selective dopamine receptor antagonist, cis-(Z)-flupentixol, significantly reduced the response produced by the administration of YM-14673 into the nucleus accumbens (1).

Recently, it has become evident that the nucleus accumbens is a heterogeneous structure. At least two different parts, i.e. the shell and the core, can be discerned (2-8). Moreover, it has been shown that the shell plays a critical role in dopamine-specific behaviours such as oral movements (9-11) and turning behaviour (12). These studies have shown that the administration of a highly specific dose-combination of the dopamine D₁ receptor agonist (±)-1-phenyl-2, 3, 4, 5- tetrahydro-1H-3-benzazepine-7, 8-diol (SKF 38393, 5 µg) or (±)-6-chloro-7, 8-dihydroxy-3-allyl-1-phenyl-2, 3, 4, 5-tetrahydro-1H-3-benzazepine (SKF 82958, 5 µg) and the dopamine D₂ receptor agonist quinpirole (10 µg) into the shell, but not the core, of the nucleus elicits jaw movements and turning behaviour in rats.

Given the behavioural evidence that YM-14673 injection into the nucleus accumbens induces jaw movements that can be significantly attenuated by the dopamine D₁/D₂ receptor antagonist cis-(Z)-flupentixol (1) and that jaw movements induced by dopamine receptor stimulation in the nucleus are shell-specific (9-11), we have decided to examine whether the shell of the nucleus accumbens plays a role in the display of YM-14673-induced jaw movements that differs from that played by the core of the nucleus. For that purpose, the effects of YM-14673 injection into the shell or the core were compared to those of a mixture of SKF 82958 and quinpirole; a drug combination that has previously been shown to be useful as a tool to functionally differentiate these two subregions of the nucleus (10).
Materials and Methods

Surgical procedures

Male Sprague-Dawley rats weighing 260-330 g were anaesthetized with ketamine HCl (10 mg/kg i.p.), supplemented during surgery with halothane (0.5-4 % when appropriate). The surgical and recording procedures were as previously described (10, 13-16). A small light-emitting diode was fixed to the mandible and 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 and horizontal movements of the diode. Bipolar electrodes were placed into the masseter and digastric muscles to record electromyographic (EMG) activity. The spinal cord was transected at the C1 level to confine drug-induced movements to the head region. The jaw movements were recorded on a polygraph for later quantification and were automatically counted with a spike trigger. The sessions lasted 240 min. Guide cannulae (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 (10,11,13). The coordinates based on the atlas of Paxinos and Watson (17) were: for the shell, anterior = 10.6 mm, vertical = 2.0 mm, lateral = 0.5 mm; for the core, anterior = 10.6 mm, vertical = 3.0 mm, lateral = 1.2 mm (Fig. 1). The cannulae directed at the shell were angled 21 degrees from the mid-sagittal plane and those directed at the core were angled 18 degrees from this plane to avoid the ventricular system. The injection of 0.2 µl per side was delivered over a 20 s period, and the needle was left in position for an additional 20 s period after completion of the injection. Damage to the target site was minimized by implanting the tips of the guide cannulae 1.2 mm (for the core) or 2.0 mm (for the shell) above the desired injection site. Wire stylets were placed in the guide cannulae to prevent occlusion. After surgery, the animals were maintained under anaesthesia by continuous infusion of ketamine (10 mg/h i.v.). Lignocaine HCl (2 %) gel was applied to all incisions and the rectal temperature was maintained at 37°C with a thermostatically controlled heating pad. Monitored concentrations of O2 and CO2 expired during the experiment were 19-21 % and 2.0-2.5 %, respectively. The number of jaw openings greater than 1 mm (measured from the diode displacement) was counted in consecutive 5 min periods for 240 min according to our previously described paradigms (16,18). Jaw movements and EMG activity were also recorded on a polygraph for further analysis of the pattern of jaw movements (16,18). All experiments were performed according to institutional and national guidelines for animal experimentation.

Drugs

The animals (n = 6-7 per experiment) received bilateral injections of the TRH analogue YM-14673 (0.1 µg or 1.0 µg; N'-[(S)-4-oxo-2-azetidinyl]- carbonyl]-L-histidyl-L-prolinamide dihydrate, Yamanouchi Pharmaceutical Lo.), the full dopamine D1 receptor agonist SKF 82958 (5 µg; (±)-6-chloro-7,8-dihydroxy-3-allyl-1-phenyl-2, 3, 4, 5-tetrahydro-1H-3-benzazepine-7, 8-diol hydrobromide, Research Biochemicals International) and the dopamine D2 receptor agonist quinpirole (10 µg; Research Biochemicals International). Control animals received the drug vehicle (0.9 % saline solution). Doses and time-schedule were based on previously published studies (1,10,14,19). Animals were used only once.

Histology

At the end of each experiment, the rats were deeply anaesthetized with sodium pentobarbitone and perfused transcardially with 10 % formalin. The brains were removed, sectioned (50 µm) and stained with cresyl violet to visualize the injection site. Only data from animals in which the injections were correctly placed were analyzed.

Data analysis

All values are expressed as means ±S.E.M. and analyzed using two-way (group×time) analysis of variance (ANOVA). Differences were considered significant when P < 0.05.

Results

Effects of injecting combined SKF 82958 and quinpirole into the shell and the core of the nucleus accumbens

The overall effects of saline and the SKF 82958/quinpirole combination are shown in Fig. 2. The mixture of SKF 82958 (5 µg) and quinpirole (10 µg) was ineffective when injected into the core (Fig. 2A); the number of jaw movements in the treated rats did not
significantly differ from that in vehicle treated animals. In contrast, this mixture was highly effective when injected into the shell (p < 0.001, two-way ANOVA). These time-dependent effects are shown in Fig. 2B. This figure clearly illustrates that the effect started almost immediately after the injection, reached its peak around 40 min and vanished at about 120 min after injection.

Effects of YM-14673 injection into the shell and the core of the nucleus accumbens on jaw movements

Bilateral injections of YM-14673 (0.1 or 1.0 µg/0.2 µl per side) into the shell or the core elicited similar dose-dependent repetitive jaw movements. The effects of YM-14673 reached maximum level at around 30 min after injection and remained stable until at least 4 h after injection (Fig. 3A, B).

The pattern of jaw movements including EMG changes induced by YM-14673 and the combination of SKF 82958 and quinpirole

The pattern of jaw movements occurring after administration of YM-14673 (0.1 or 1.0 µg) into the shell compared to the core of the nucleus accumbens was similar. Injections of YM-14673 into both sites produced large amplitude opening and closing movements in the vertical direction that were followed by small amplitude opening and closing movements; the latter were very rapid in comparison with the former. Jaw movements in the horizontal direction (lateral jaw movements) were also present. The jaw movements were associated with a pronounced activity of the digastric muscle and the masseter muscle. The movements were not accompanied by tongue protrusions. This pattern was identical to that previously described (1), but differed from that produced by injections of the dopamine D1 and D2 receptor agonist combination (SKF 82958 5 µg + quinpirole 10 µg/0.2 µl) into the shell of the nucleus accumbens. This combination produced a continuous series of opening and closing movements of the jaw with varying amplitude.

![Fig. 2](image1.png)  
Fig. 2 The time-dependent effects of bilateral injections of saline (○) or the mixture of SKF 82958 and quinpirole (●) into the shell (A) or the core (B) of the nucleus accumbens (0.2 µl/side) on production of jaw movements. 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.

![Fig. 3](image2.png)  
Fig. 3 The time-dependent effects of bilateral injections of YM-14673 (0.1 µg, A or 1.0 µg, B; 0.2 µl/side) into the shell (●) or the core (○) of the nucleus accumbens on production of jaw movements. 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.
Digastric and masseter muscles showed continuous activity throughout the period of maximum drug effect. Frequent tongue protrusions occurred during the large amplitude jaw openings. This pattern was similar to that previously described (16,18).

**Discussion**

As mentioned in the Introduction, there exists increasing evidence showing that the shell and the core of the nucleus accumbens differ in terms of anatomy (2-8) and pharmacology (9-12). The outcome of the present study confirms the earlier reported findings that the shell, but not the core, of the nucleus accumbens is involved in the control of oral movements, especially jaw movements induced by co-stimulation of dopamine D₁ and D₂ receptors (10-12): the combined administration of SKF 82958 and quinpirole was only effective when injected into the shell, but not the core, of the nucleus. The finding that the shell of the nucleus accumbens is especially involved in oral behaviour is understandable in view of the fact that the shell projects to a part of the ventral pallidum (20) that is known to be involved in oral behaviour (21).

It has also been shown that the nucleus accumbens play a role in jaw movements elicited by YM-14673 (1). Since the response to YM-14673 was attenuated by the dopamine D₁/D₂ receptor antagonist cis-(Z)-flupentixol (1), it is suggested that YM-14673 directly or indirectly enhanced the dopamine activity at the level of dopamine D₁/D₂ receptors. These findings give rise to the hypothesis that the shell of the nucleus accumbens plays a role in the display of the YM-14673-induced jaw movements that differs from that played in the core. The outcome of the present study shows a lack of significant difference between the effects of shell and the core; thus, YM-14673 injected into the shell and the core of the nucleus accumbens similarly elicited repetitive jaw movements in a dose-related manner. The absence of any significant difference between the shell and the core in this respect give rise to two alternative explanations. First, both treatments elicited identical effects, because YM-14673 diffused from the shell to the core, and/or vice versa. In that case, it is still possible that the role of the shell in YM-14673-induced jaw movements differs from that of the core. Second, the shell shares its role with that of the core. Further research is required to solve this problem.

This study reports at least two findings indicating that YM-14673 has pharmacologically distinct effects from the D₁/D₂ dopamine-receptor agonists. First, the pattern of jaw movements induced by YM-14673 is not identical to that elicited by stimulation of dopamine D₁ and D₂ receptors. Although the differences in number and amplitude of jaw movements may have resulted from differences in the degree in which the dopaminergic postsynaptic activity is altered by YM-14673 and by direct receptor stimulation with SKF 82958 + quinpirole, the absence (YM-14673) or presence (SKF 82958 + quinpirole) of tongue protrusions cannot be ascribed to such differences. Secondly, it has reported that the effects of YM-14673 could be blocked only partially by a dose of the dopamine D₁/D₂ receptor antagonist cis-(Z)-flupentixol (1) that is highly effective in blocking the effects of dopamine receptor agonists upon jaw movements (15). Together, these data strongly suggest that YM-14673 has distinct effects, apart from those that rely upon dopamine transmission.

Taking the above-mentioned findings and considerations together, it appears that the TRH-analogue YM-14673 can induce behavioural effects by the mechanisms which can directly and/or indirectly enhance the dopamine activity at the level of dopamine D₁/D₂ receptors in the shell and/or the core of the nucleus accumbens. Although such mechanisms may at least partly contribute to the ability of YM-14673 to elicit jaw movements after injections into the structures, it is evident that additional mechanisms must play a role in the display of jaw movements.

**Acknowledgements**

We thank Yamanouchi Pharmaceutical Co. for the gift of YM-14673.

**References**

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