Pharmacology Biochemistry and Behavior, Vol. 66, No. 3, pp. 615–622, 2000

0091-3057/00/$–see front matter

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PII S0091-3057(00)00277-X

Involvement of the Medial Precentral Prefrontal Cortex in Memory Consolidation for Inhibitory Avoidance Learning in Rats

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Received 28 August 1999; Revised 6 January 2000; Accepted 18 January 2000

MELLO E SOUZA, T., M. R. M. VIANNA, C. RODRIGUES, J. QUEVEDO, B. A. MOLETA AND I. IZQUIERDO. *Involvement of the medial precentral prefrontal cortex in memory consolidation for inhibitory avoidance learning in rats.* PHARMACOL BIOCHEM BEHAV **66**(3) 615–622, 2000.—Adult male Wistar rats were trained in a step-down inhibitory avoidance learning task (3.0-s, 0.4-mA foot shock), received a 0.5-μl infusion of muscimol (0.02, 0.1, or 0.5 μg), AP5 (0.16, 0.34, 0.5, 1.6, or 5.0 μg), SCH 23390 (0.05, 0.34, 0.5, or 1.75 μg), saline, or vehicle (DMSO 20%) into the anterior medial precentral area (Fr2) (CI) immediately after training, and were tested 24 h later. Muscimol (0.02, 0.1, or 0.5 μg), AP5 (0.34 or 0.5 μg), or SCH (0.5 or 1.75 μg) were amnesic. Then, animals were infused with muscimol (0.1 or 0.5 μg), AP5 (0.34, 0.5, or 5.0 μg), or SCH (0.5 μg) at other posttraining times and/or into the junction of Fr1–Fr2 (CII). Muscimol (0.1 and 0.5 μg) or SCH into CI were amnesic when given 90 or 180 min after training, but not when given 270 min after training. Muscimol (0.5 μg, but not 0.1 μg) or SCH into CII were amnesic when given 90 min after training, but not when given 0 or 180 min after training. AP5 (0.5, but not 5.0 μg) was amnesic when given into CI, but not into CII, at 0 or 180 min posttraining, and a trend toward an amnesic effect was seen at 90 min posttraining. The results suggest that 1) the glutamatergic, GABAergic, and dopaminergic systems in Fr2 are involved in the consolidation of memory for inhibitory avoidance learning, either directly or as parts of modulatory systems; and 2) timing of involvement of anterior Fr2 (CI) is different from that of posterior Fr2 (CII). © 2000 Elsevier Science Inc.

Prefrontal cortex Dopamine NMDA receptors AMPA receptors GABA Memory consolidation

THE hippocampus, the amygdala, and the entorhinal, parietal, and posterior cingulate cortices are involved in the consolidation of memory for a step-down inhibitory avoidance learning task in rats through mechanisms sensitive to the *N*-methyl-D-aspartate (NMDA) glutamatergic receptor antagonist aminophosphonopentanoic acid (AP5) and to the γ-aminobutyric acid type A ($GABA_A$) receptor agonist muscimol. Infusion of muscimol or AP5 is amnesic when given immediately after training into the hippocampus and amygdala, 30–180 min after training into the entorhinal cortex, or 60–180 min after training into the parietal cortex (16–18). Infusion of muscimol is amnesic when given 90 min after training into the posterior cingulate (27). Taking into account that AP5 and muscimol interfere with the signaling of the most important excitatory and inhibitory neurotransmitters of the central nervous system, respectively, these results indicate that 1) the hippocampus and amygdala participate early on, and 2) the entorhinal, parietal, and cingulate cortices play a role in memory formation at least 0.5 h later (16–18,27). Otherwise, timing of involvement of other neurotransmitter systems in the hippocampus, such as the cholinergic, dopaminergic, and adrenergic systems, is different from that of the glutamatergic and gabaergic systems, probably because these three systems are involved in modulatory rather than in core mechanisms [see review in (18)].

The search for the involvement of other areas, such as the prefrontal cortex (in the case of this work, its precentral area—Fr2), in the consolidation of memory for inhibitory avoidance learning is one of our main interest. The identification of the prefrontal cortex is based on reciprocal connections between this region and the medial dorsal nucleus of the thalamus (43),

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despite the inclusion of the rat medial precentral (Fr2) and the infralimbic (IL) areas as parts of the rat medial prefrontal cortex (12,38,39). Fr2 comprises an area equivalent to the sum of human Brodmann's areas (BA) 8 and 10 (5,6), which corresponds to part of the prefrontal association cortex.

The prefrontal cortex and other brain structures mediate working memory (10,32), which is defined as a system that processes and holds information for very short periods (seconds), and by which the animal perform cognitive tasks, such as comprehension, thinking, and planning (1,10). The prefrontal cortex is also fundamental for delayed responses (11,31,35) and, in the case of BA 8, for visual conditional learning (30). In humans, BA 10 is involved in autobiographical episodic memory (7).

Fr2 also seems to comprise an area corresponding to the primate premotor and supplementary motor areas (BA 6) (5,6,13,39), which receive converging axons from the prefrontal and parietal cortex and convert signals encoding desired actions into how the actions will be carried out (2). The primate premotor area is also involved in working memory and delayed responses (8,22,37,41), as well as in motor memory consolidation (36) and in visuomotor sequence learning (14). Neurons in a specific region of the primate supplementary area are activated particularly when subjects encounter a new context that requires motor plans to be updated (15).

Increased levels of dopamine in the prefrontal and premotor cortex were seen in primates performing a task related to working memory (41). Evidence has shown that D_1 dopaminergic receptors are involved in working memory (10,34,41,42). In a previous study, we showed that immediately posttraining infusion of the type 1 dopaminergic (D_1) receptor antagonist SCH23390 (0.5 μg) or of muscimol into the medial precentral area (Fr2, CI) was amnesic for long-term memory (test at 24 h), but not for short-term memory (test at 1.5 h), in a one-trial step-down inhibitory avoidance learning task in rats. In addition, infusion of these drugs 6 min prior to training impaired immediate memory for this task (19).

The objective of the present work is to analyze the involvement of the anterior medial precentral area (Fr2) in the consolidation of memory for a step-down inhibitory avoidance learning task in rats. To analyze the involvement of the two main neurotransmitters of the central nervous system, glutamate and GABA, infusion of AP5 or muscimol was given immediately after training in different concentrations, range of which included those concentrations used in previous works (16,17,27). Some of these concentrations, mainly those that were effective, were also infused 1) at other posttraining times, to see how long these drugs may affect consolidation of memory for this task, and/or 2) into a more posterior region of Fr2, because the rat Fr2 corresponds to different regions of the human brain (5,6,13,39) and might have functional differences along the anterior-posterior axis.

Because the dopaminergic system in the prefrontal cortex is involved in working memory (10,34,41,42) and might be involved in other functions, such as memory consolidation for inhibitory avoidance learning, we also intend to analyze the involvement of this system in Fr2. Therefore, different concentrations of SCH were infused into anterior Fr2 (CI) immediately after training, and the most effective concentration was also infused at other posttraining times and/or into CII.

METHOD

Subjects

Five hundred thirteen male Wistar rats (age, 60–90 days) were obtained from our breeding colony. The animals were housed five to a cage with food and water ad lib under a 12 L:12 D cycle (lights on at 0700 h) at a temperature of 23°C.

Surgery and Behavioral Procedures

The animals were bilaterally implanted under thionembutal anesthesia (30 mg/kg, IP) with 27-gauge guide cannulae. At least 48 h later, all animals were trained in a step-down inhibitory avoidance learning task (16,17,19,27). The rats were placed on a 2.5-cm high by 7.0-cm wide formica platform at the left of a 50 × 25 × 25-cm apparatus, the floor of which was a series of parallel 0.1-cm caliber stainless steel bars spaced 1.0 cm apart. Latency to step down placing the four paws on the grid was measured. In the training session, immediately upon stepping down, the animals received a 3.0-s, 0.4-mA foot shock. A retention test was carried out 24 h after training. The test session was procedurally identical except that no foot shock was given, and the step-down latency was cut off at 180 s; i.e.., test session values higher than 180 s were counted as 180 s. Retention test performance was taken as a measure of retention.

Infusion Procedure and Control of Cannula Placements

At the time of infusion, 30-gauge cannulae were fit into the guide cannulae (16,17,19,27). Animals received a bilateral infusion, which was given into anterior Fr2 (CI) immediately after training, of 0.5 μl of the $GABA_A$ agonist receptor muscimol HBr (0.02, 0.1, or 0.5 μg), of the glutamate NMDA antagonist 2-amino-S-phosphonopentanoic acid (AP5) (0.16, 0.34, 0.5, 1.6, or 5.0 μg), of the D_1 receptor antagonist R(+)SCH 23390 HCl (0.05, 0.34, 0.5, or 1.75 μg), of saline (phosphate buffer, pH 7.4), or of vehicle (solution of DMSO 20% in saline). To verify the involvement of Fr2 at other posttraining times, muscimol (0.1 and 0.5 μg), AP5 (0.5 and 5.0 μg), or SCH (0.5 μg) were also infused 90, 180, or 270 min after training (muscimol and AP5 were infused only in the higher concentration at 270 min posttraining). These posttraining time points were chosen because drugs presumably diffuse away within 90 min (26). To verify whether these drugs may also disrupt memory consolidation when infused into a more posterior region of Fr2, muscimol (0.5 μg), AP5 (5.0 μg), or SCH (0.5 μg) were also infused into the junction of Fr1–Fr2 (CII) immediately, 90, or 180 min after training. In addition, infusion of muscimol (0.1 μg) or AP5 (0.34 or 0.5 μg) was also administered 90 min after training into CII. All drugs were purchased from Research Biochemicals International (RBI), Natick, MA. The sites of infusion were chosen using coordinates (from bregma and dura) obtained from (29), as follows (units in cm): most anterior part of Fr2 (CI), A +0.47, L ±0.28, V −0.10 (Fig. 1A); and Fr1–Fr2 junction (CII), A +0.20, L ±0.20, V −0.01 (Fig. 1B).

Two to 24 h after the end of the behavioral procedure all animals received an infusion of 0.5 μl of 4% methylene blue through the infusion cannulae, and were killed by decapitation. Their brains were removed and stored in formalin for histological localization of infusion sites as explained elsewhere (16,17,19). Infusion placements were correct in 337 and 170 animals implanted in coordinates I and II, respectively. Only animals with correct cannula locations (Fig. 1) were included in the final statistical analysis.

Statistics

Data are reported as median (interquartile range) of the retention test performance. Training session performances of groups of the same site and time of infusion were compared

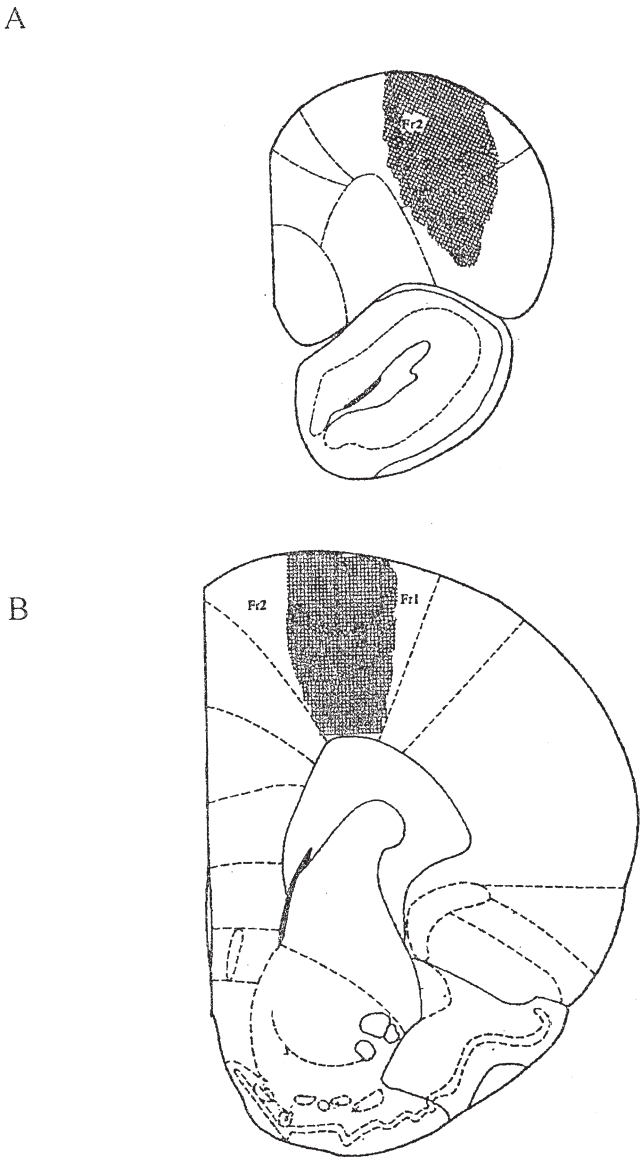

FIG. 1. (A, B) Schematic drawings of rat brain sections at planes +0.47 and +0.22, respectively, from (29) showing (stippled) the extension of the areas reached by infusions into coordinates I and II. In each animal, maximum extension of the site(s) reached by the infusions was less than 1.5 mm^3, as ascertained by the spread of a 0.5 μl infusion of 4% methylene blue into each of the structures, 24 h after the last behavioral manipulation.

by the Kruskal–Wallis analyzes of variance. Differences between training and test session performances in each group were evaluated by a Wilcoxon test. Differences from the control group of the same time of infusion in test session performances were evaluated by Mann–Whitney *U*-test, two-tailed. $p < 0.05$ was considered to indicate statistical significance.

RESULTS

Training vs. Test Session Performances

There is no difference among groups regarding the training session performances (overall median, 4.9 s; overall interquartile range, 3.3/7.6 s; Kruskal–Wallis ANOVA, $p > 0.10$; data not shown).

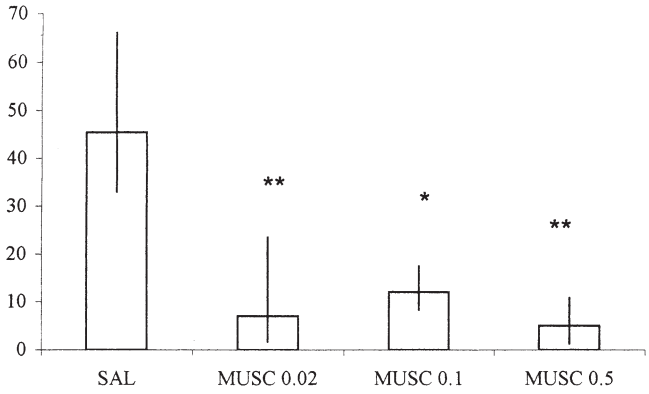

FIG. 2. Median (interquartile range) test session latency in groups infused bilaterally into coordinate I of Fr2 with saline (SAL) or muscimol (0.02, 0.1, or 0.5 μg) immediately after training. Double and single asterisks indicates statistical significance in Mann–Whitney *U*-tests, two tailed, at $p < 0.01$ and at $p < 0.05$, respectively, from the respective control group. *N* per group was 10.

Animals infused into CI with AP5 (0.34 μg) immediately after training, SCH (0.5 μg) 0, 90, or 180 min after training, or the higher dose of muscimol 0 or 180 min after training did not show a difference between training and test session performance (Wilcoxon test, $p > 0.10$). Animals infused into CII with the higher dose of muscimol 90 min after training did not show a difference between both sessions (Wilcoxon test, $p > 0.10$). The other groups showed a difference between both sessions (Wilcoxon test, $p < 0.05$).

Effects of Infusion Into Anterior Fr2 (CI) Given Immediately After Training

Three dose–response curves were obtained from infusions into CI immediately after training (Figs. 2, 3 and 4). Muscimol (0.02, 0.1, or 0.5 μg), SCH (0.5 or 1.75 μg, but not 0.05 or 0.34 μg), or AP5 (0.34 or 0.5 μg, but not 0.16, 1.6, or 5.0 μg) were amnesic when given into CI immediately after training

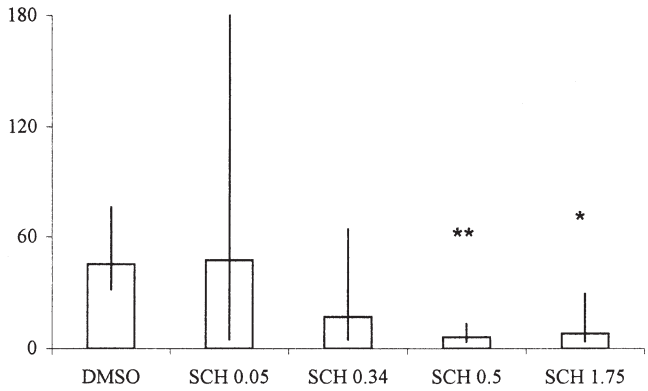

FIG. 3. Same of the previous figure but for infusions of SCH (0.05, 0.34, 0.5, or 1.75 μg). *N* per group was 9–13.

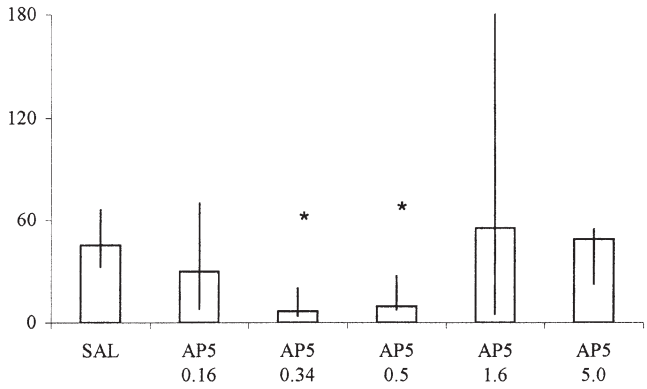

FIG. 4. Same of the previous figure but for infusions of AP5 (0.16, 0.34, 0.5, 1.6, or 5.0 μg). *N* per group was 10.

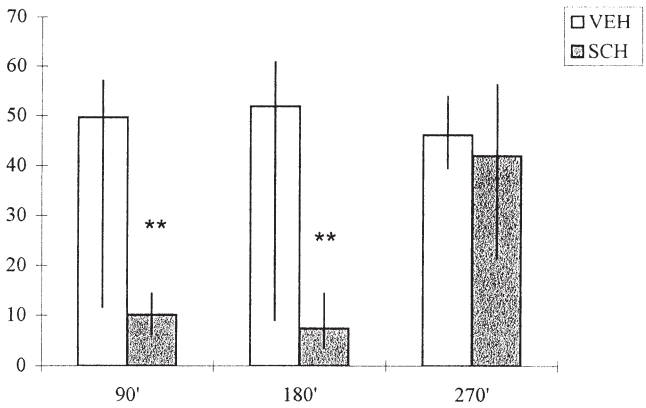

FIG. 6. Same of the previous figure but for infusions of SCH (0.5 μg). *N* per group was 9–11.

(Mann–Whitney *U*-test, two-tailed, $p < 0.05$; $p > 0.10$ for groups not different from controls). *N* per group was 10, 9–13, and 10, respectively.

Effects of Infusion Into Anterior Fr2 (CI) Given 90–270 min After Training

Muscimol (0.1 or 0.5 μg) was amnesic when given into CI 180 min after training (Mann–Whitney *U*-test, two tailed, $p < 0.05$), but it was amnesic when given 90 min after training only at the higher dose (Mann–Whitney *U*-test, two tailed, $p < 0.05$) and was not effective when given 270 min after training (Mann–Whitney *U*-test, two tailed, $p > 0.10$) (Fig. 5). A trend toward an amnesic effect was seen when muscimol at 0.1 μg was given at 90 min posttraining (Mann–Whitney *U*-test, two tailed, $p < 0.10$). *N* per group was 8–11. SCH (0.5 μg) was amnesic when given 90 or 180 min (Mann–Whitney *U*-test, two tailed, $p < 0.05$), but not 270 min after training (Mann–Whitney *U*-test, two tailed, $p > 0.10$) (Fig. 6). A trend toward an amnesic effect was seen when AP5 (0.5, but not 0.34 or 5.0 μg) was given at 90 min posttraining (Mann–Whitney *U*-test, two tailed, $p < 0.10$) (Fig. 7a). AP5 (0.5, but not 5.0 μg) was amnesic when given 180 min (Mann–Whitney *U*-test, two tailed, $p < 0.05$), but not when given 270 min after training (Mann–Whitney *U*-test, two tailed, $p > 0.10$) (Fig. 7b). *N* per group was 9–11.

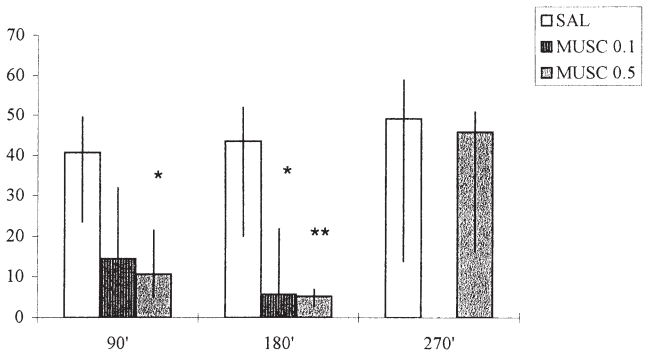

FIG. 5. Median (interquartile range) test session latency in groups infused bilaterally into coordinate I of Fr2 with saline (0.5 μl, pH 7.4) or muscimol (0.1 or 0.5 μg) (see legend), 90, 180, or 270 min posttraining. Double and single asterisk indicates statistical significance in Mann–Whitney *U*-tests, two tailed, at $p < 0.01$ and at $p < 0.05$, respectively, from the control group. *N* per group was 8–11.

Effects of Infusion Into the Junction of Fr1–Fr2 (CII) Given 0–180 min After Training

Muscimol was amnesic at the higher concentration (Mann–Whitney *U*-test, two tailed, $p < 0.05$), but not at the lower concentration (Mann–Whitney *U*-test, two tailed, $p > 0.10$), when given 90 min after training into CII. Muscimol was not effective when given 0 or 180 min after training into CII (Mann–Whitney *U*-test, two tailed, $p > 0.10$) (Fig. 8). *N* per group was 10–11.

AP5 (0.5 or 5.0 μg) was not effective when given into CII 0, 90, and 180 min after training (Mann–Whitney *U*-test, two tailed, $p > 0.10$) (Fig. 9). *N* per group was 8–11.

Infusion of SCH into CII was amnesic when given 90 min after training (Mann–Whitney *U*-test, two tailed, $p < 0.05$), but not when given immediately or 180 min after training (Mann–Whitney *U*-test, two tailed, $p > 0.10$) (Fig. 10). *N* per group was 10–11.

DISCUSSION

The present results suggest that 1) the glutamatergic, GABAergic, and dopaminergic systems in Fr2 are involved in the consolidation of memory for inhibitory avoidance learning, either directly or as parts of modulatory systems; and 2) the most anterior part of Fr2 is involved in this for a longer period than the most posterior part.

Previous studies from our laboratory showed that the hippocampus, the amygdala, the entorhinal, parietal, and posterior cingulate cortices are involved in the consolidation of memory for a step-down inhibitory avoidance learning task by muscimol-dependent mechanisms (16–18,27). The amnesic effect of muscimol (0.5 μg) into coordinate I when given 0–180 min after training suggests two explanations: 1) the most anterior part of Fr2 is involved in the consolidation of memory for inhibitory avoidance learning by mechanisms sensitive to muscimol during a period of time equivalent to that of the hippocampus (0 min) and the entorhinal (30–180 min), parietal (60–180 min), and posterior cingulate (90 min) cortices combined; or (2) alterations in the activity of the most anterior part of

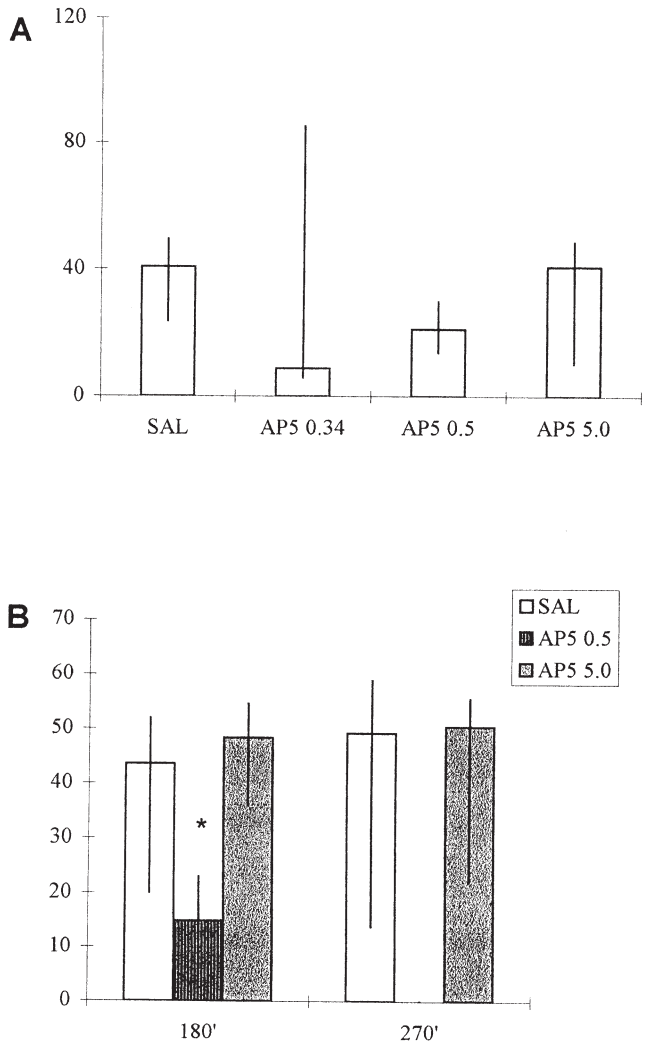

FIG. 7. (A) Median (interquartile range) test session latency in groups infused bilaterally into coordinate I of Fr2 with saline (SAL) or AP5 (0.34, 0.5, or 5.0 μg) 90 min after training. (B) Same of A but for infusions of saline (0.5 μl, pH 7.4) or AP5 (0.5 or 5.0 μg) (see legend) 180 or 270 min posttraining. Single asterisk indicates statistical significance in Mann–Whitney *U*-tests, two tailed, at $p < 0.05$, from the control group. *N* per group was 9–11.

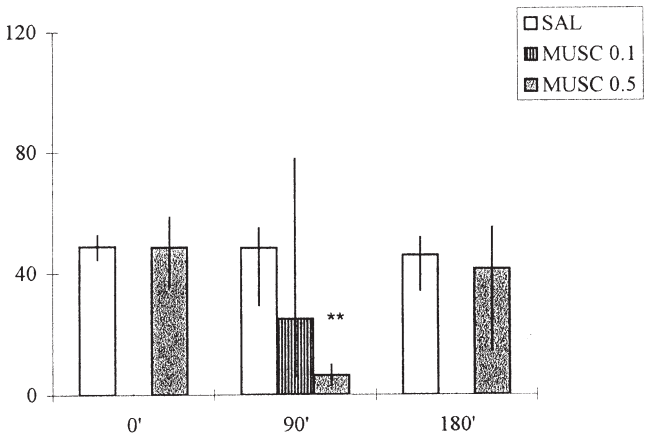

FIG. 8. Median (interquartile range) test session latency in groups infused bilaterally into coordinate II with saline (SAL) or muscimol (0.1 or 0.5 μg) immediately, or 90 or 180 min after training. Double asterisks indicates statistical significance in Mann–Whitney *U*-tests, two tailed, at $p < 0.01$ and at $p < 0.05$, respectively, from the respective control group. *N* per group was 10–11.

Fr2 merely affect the transactional processes involved in consolidation 0–180 min posttraining. In the case of the most posterior part of Fr2, its time window seems to occur between that of the hippocampus (0 min), and those of the entorhinal (30–180 min) and parietal cortex (60–180 min) (19), because muscimol (0.5 μg) infusion was effective only at 90 min posttraining.

The differentiation of Brodmann's areas 6 (premotor area) and 8, which belong to the posterior dorsolateral frontal cortex, and area 10 (frontopolar cortex) in humans, but not in rats (5,6,13,39), may be related to further evolutionary advances of the human brain, prominent in the neocortex. Therefore, we think that the different intervals of involvement between the most anterior and the most posterior parts of Fr2 by mechanisms sensitive to muscimol might be related to functional rather than anatomical differences of Fr2 along the anterior–posterior axis.

The fact that muscimol was amnesic when given into CI 90 min after training only at the higher concentration suggests that Fr2 is less sensitive to muscimol infusion at 90 min posttraining relative to other posttraining times, 0 and 180 min; i.e., muscimol might have two peaks of action, at 0 and 180 min. The reasons for this are unknown. Increasing evidence of simultaneous and coordinated activity of different brain regions in the posttraining period suggests a "multiple consolidation of memory" (4). We might speculate that the processes of consolidation are widely distributed at this time, being mediated by several cortical areas, such as the entorhinal, parietal, and cingulate cortices (17,18,27), and the precentral area. Therefore, the network would be less sensitive to alterations in the precentral area.

The amnesic effect of SCH infused into both coordinates might be related to the role of dopamine in modulating pre-

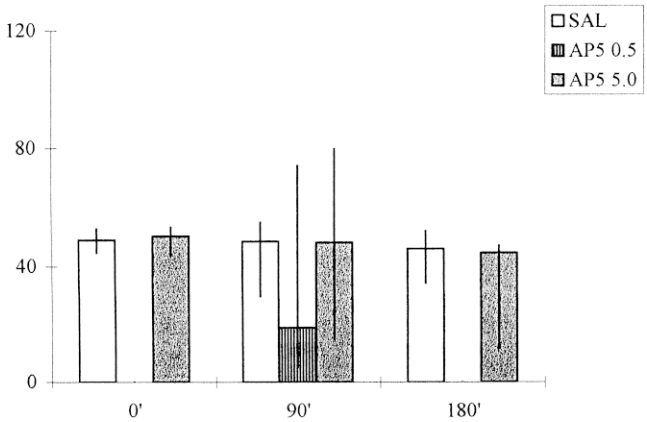

FIG. 9. Same of the previous figure but for infusions of AP5 (0.5 or 5.0 μg). *N* per group was 8–11.

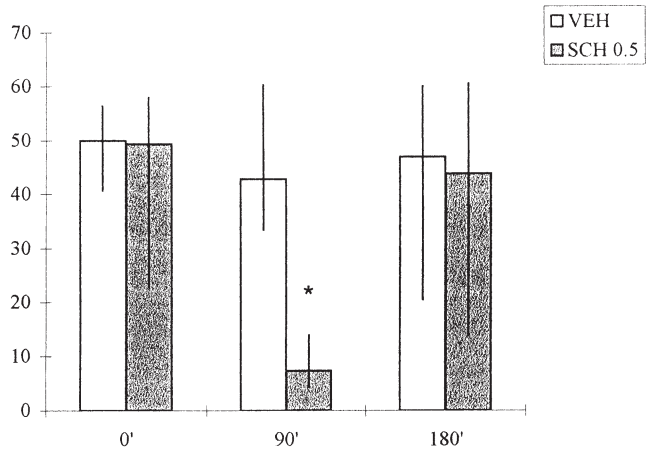

FIG. 10. Same of the previous figure but for infusions of SCH (5.0 μg). *N* per group was 10–11.

frontal pyramidal cell excitability, which is, for instance, essential for working memory for a oculomotor task, which requires memory-guided saccades (10,34). Primates have increased levels of extracellular dopamine in the prefrontal cortex during performance of a delayed alternation task, and in the premotor area, during performance of both this task and a sensory-guided paradigm (41). In addition, spatial working memory performance exhibits an inverted U dose–response curve to D_1 receptor stimulation levels in the prefrontal cortex; i.e., intermediate levels of D_1 receptor stimulation optimize working memory performance (42). A previous study showed that SCH (0.5 μg) impaired immediate memory for inhibitory avoidance learning when applied prior to training, and impaired consolidation when applied immediately after training (19). In this study, the dose–response curve shows that SCH is amnesic when infused immediately after training at doses higher than a threshold, value of which is higher than 0.34 μg. It is worth pointing out that 1) the highest dose almost reachs the saturation point of SCH in vehicle (DMSO 20% in saline), and 2) the dose of 0.5 μg is enough to significantly bind SCH to D_1 receptors, but not to serotonergic receptors (3,9,25). Therefore, low levels of D_1 receptor stimulation in Fr2 may disrupt consolidation of memory for inhibitory avoidance learning.

Because AP5 was amnesic when given into anterior CI immediately after training only at intermediate doses, this drug presents a U dose–response curve. In addition, AP5 is effective in a small range of concentrations. The reasons for this are unknown, and deserve further studies. Some cues raises from other studies: 1) a presynaptic modulation of dopamine release from NMDA and non-NMDA glutamate receptors, as occurs in the striatum (40); 2) a stimulation of GABA release by NMDA receptor activation, which might inhibit the release of dopamine, as probably occurs in the medial prefrontal cortex (21); 3) glutamatergic neurotransmission at non-NMDA receptors in the prefrontal cortex is increased or decreased by low or high doses, respectively, of the NMDA receptor antagonist ketamine, being intermediate doses ineffective (28); 4) in the striatum, high levels of NMDA receptor activation increase dopamine release, an effect that has been attributed to general excitation (24). However, it is worth pointing out that the amnesic effect of AP5 when given into anterior Fr2 (CI) immediately and 180 min after training suggests that NMDA receptors in this area are essential for memory consolidation in inhibitory avoidance learning at these times. In addition, our data do not definitively rule out the possibility that NMDA receptors are involved in this process at 90-min posttraining, because a trend toward an amnesic effect of AP5 (0.5 μg) was found at this time. It is also worth pointing out that muscimol is amnesic when infused immediately after training in a larger range of concentrations than SCH or AP5, even though it has not been necessary to cover the whole dose–response curve of this drug.

The timing of involvement of NMDA, $GABA_A$, and D_1 receptors in anterior Fr2 is probably the same (0–180 min), despite the decrease of sensitivity for muscimol and AP5 at 90 min posttraining. This similarity might be interpreted as an indirect evidence for either an interaction of these systems in Fr2, as occurs in the striatum and in the medial prefrontal cortex (21,28,40), or an interaction that emerges from the recycling of information in the network formed by the prefrontal cortex, the motor and premotor areas, the basal ganglia, and the ventral tegmental area: the motor loop (2). For instance, a contralateral infusion of bicuculline, a $GABA_A$ antagonist, into the medial prefrontal cortex increases the release of dopamine in the striatum, an effect that is reverted by an infusion of an excitatory amino acid receptor antagonist into the ventral tegmental area, but not directly into the striatum, which receives afferents from both the medial prefrontal cortex and the ventral tegmental area (23). Alterations in the glutamatergic system in Fr2 might indirectly interfere with its dopaminergic system via other structures. The fact that anterior Fr2 is less sensitive to muscimol and AP5 infusion at 90 min posttraining might also be explained by one of these putative interactions among the neurotransmitter systems in Fr2. For instance, different mechanisms may be responsible for the activation of dopamine release in the striatum following potent stressors and in the prefrontal cortex following relatively lower stressors (20). The set of mechanisms involved in consolidation at 90 min posttraining might differ from that at 0 or 180 min posttraining. Further studies may clarify these issues and their relevance to memory for inhibitory avoidance learning.

The lack of effect or even a trend toward an effect when AP5 at both concentrations was given into CII suggests that NMDA receptors in this region are not essential for memory consolidation in inhibitory avoidance learning. However, our results suggest that D_1 receptors, not only in anterior Fr2, but also in posterior Fr2, may be involved in consolidation of memory for inhibitory avoidance learning. However, D_1 receptors in posterior Fr2 may be relevant only at 90 min posttraining.

We should point out that another explanation of our results might be that other structures than Fr2 were also reached by our infusions; i.e., each infusion might reach a larger area than showed by our histological procedure. In the case of infusions into CI, our histology showed it was restricted to Fr2 in its majority, but some of them reached the sulcal cortex, an area that corresponds to the primate orbitofrontal cortex (Fig. 1). Therefore, infusions into CI might reach the sulcal cortex and, less probably, the medial prefrontal cortex, which are involved in memory for inhibitory avoidance learning (33). In the case of CII, infusions might reach the dorsal and, less probably, the ventral medial prefrontal cortex. However, the possibility of this explanation is unlikely, because 1) 1.0-μl infusions of muscimol, twice larger in volume than our infusions, maximally reduce glucose uptake in a restricted region of 1 mm^3 (26); and 2) muscimol infusion

into ventral medial prefrontal cortex at the coronal plane + 2.2 cm was not effective in altering retention for inhibitory avoidance learning measured 24 h after training (27).

In conclusion, further studies are necessary to clarify the function of the medial precentral area (Fr2) in memory and the interaction of its neurotransmitter systems. The present contribution focuses on the time window in which memory consolidation for inhibitory avoidance learning in rats is sensitive to posttraining infusion of muscimol, AP5, or SCH, at different doses, into the medial precentral area (Fr2).

ACKNOWLEDGEMENTS

This study was supported by CNPq and PRONEX, Brazil. We are grateful to Silvia Alejandra Minetti, Marcelo Madruga, and Márcia Kauer Sant'Anna for their contribution in the present work.

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