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Involvement of Central Cholinergic Mechanisms in the Effects of Oxytocin and an Oxytocin Receptor Antagonist on Retention Performance in Mice

Mariano M. Boccia and Carlos M. Baratti

Laboratorio de Neurofarmacología de Procesos de Memoria, Cátedra de Farmacología, Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires, Junín 956-5° Piso, 1113-Buenos Aires, Argentina

Oxytocin (OT, 0.10 μg/kg, sc) impaired retention of a one-trial step-through inhibitory avoidance task when injected into male Swiss mice 10 min after training, as indicated by retention performance 48 h later. In contrast, the immediate post-training administration of the putative oxytocin receptor antagonist $d(CH_2)_5$[Tyr(Me)^2, Thr^4, Thy-NH$_2^9$] OVT (AOT, 0.30 μg/kg, sc) significantly enhanced retention performance. Neither OT nor AOT affected response latencies in mice not given footshock on the training trial, and neither the impairing effects of OT nor the enhancing effects of AOT were seen when the training–treatment interval was 180 min, suggesting that both treatments influenced memory storage. The effects of OT (0.10 μg/kg, sc) on retention were prevented by AOT (0.03 μg/kg, sc) given immediately after training, but 10 min prior to OT treatment. The central acting anticholinesterase physostigmine (35, 70, or 150 μg/kg, ip), but not its quaternary analogue neostigmine (150 μg/kg, ip), reversed the impairment of retention performance induced by OT, whereas low subeffective doses of the centrally active muscarinic cholinergic antagonist atropine (0.5 mg/kg, ip) or the central acting nicotinic cholinergic antagonist mecamylamine (5 mg/kg, ip), but not methylatropine (0.5 mg/kg, ip) or hexamethonium (5 mg/kg, ip), prevented the enhancement of retention performance caused by AOT. We suggest that oxytocin negatively modulates the activity of central cholinergic mechanisms during the posttraining period that follows an aversively motivated learning experience, leading to an impairment of retention performance of the inhibitory avoidance response.

Key Words: oxytocin; oxytocin receptors–memory; oxytocin antagonists–memory; acetylcholine.

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Address correspondence and reprint requests to Carlos M. Baratti, Cátedra de Farmacologia, Universidad de Buenos Aires, Junin 956-5 Piso, Buenos Aires RA-1113, Argentina. Fax: 54-11-4508-3645. E-mail: cbaratti@ffyb.uba.ar.

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INTRODUCTION

The closely related nonapeptides arginine vasopressin (AVP) and oxytocin (OT) were originally identified as hormones secreted from the neurohypophysis into the systemic circulation. The classical hormonal actions of AVP on kidney and blood vessels and those of OT on the uterus and mamma are beyond doubt. In addition to these well-known effects, both peptides also exert a variety of central nervous system (CNS) effects (De Wied, Diamant, & Fodor, 1993), including profound actions on learning and memory processes (Engelmann, Wotjak, Neumann, Ludwing, & Landgraf, 1996; De Wied et al., 1993; Kovacs & De Wied, 1994). Although the mechanisms of action of AVP and OT on behavioral performance are largely unknown, and several questions remain to be answered (Engelmann et al., 1996), there is an increasing body of neuroanatomical, neurochemical, and neuropharmacological evidence supporting the notion that AVP and OT function in the CNS as neuromodulators of the synaptic transmission in classical transmitter pathways known to be closely associated with information processing in limbic and cortical structures (Buijs, 1983; Kovacs & De Wied, 1994; Kovacs & Versteeg, 1993; Rinaman, Sherman, & Stricker, 1995).

In general, AVP and some AVP fragments induce long-lasting facilitation of learned inhibitory behaviors, whereas OT tends to impair the retention of these behaviors (Boccia, Kopf, & Baratti, 1996, 1998; Elands, de Kloet, & De Wied, 1992; De Wied et al., 1993; Faiman, de Erausquin, & Baratti, 1991). Oxytocin facilitated extinction of a bench-jumping active avoidance response in rats (Schulz, Kovacs, & Telegdy, 1974), attenuation of social memory by OT has also been reported (Dantzer, Bluthe, Koob, & Le Moal, 1987; Popik & Vetulani, 1991), and recently we found that OT impaired retention of a “nose-poke” habituation response in mice (Boccia & Baratti, 1999) (for a further review see Engelmann et al., 1996).

The relevance of these findings to human memory is uncertain, and inconsistent effects of neurohypophyseal peptides on human memory in various experimental paradigms have been reported (Fehm-Wolfsdorf & Born, 1991).

Arginine vasopressin and OT mediate their effects on inhibitory avoidance behavior via the AVP V_{1a} receptor subtype (Boccia et al., 1996; De Wied, Gafffori, Van Ree, & de Jong, 1984; Faiman et al., 1991) and the OT receptor (Boccia et al., 1998), respectively (but also see De Wied, Elands, & Kovacs, 1991). The effects of OT on retention are the opposite of those induced by AVP, and previously we suggested that AVP may exert a positive modulatory influence on central cholinergic mechanisms involved in memory processes (Faiman et al., 1991). A modulatory influence of OT on neuronal events was suggested by Kiraly, Maillard, Dreifuss, and Dolivo (1985) when it was demonstrated that the peptide depresses cholinergic neurotransmission in the rat superior cervical ganglion following its preganglionic stimulation, without affecting the sensitivity of the ganglion cells to acetylcholine. The effect was attributable to a presynaptic reduction of acetylcholine release from the preganglionic cells (Kiraly et al., 1985; Barberis & Tribollet, 1995).

The aim of the present work was to investigate the possible interaction, with regard to retention, of OT and an OT receptor antagonist and drugs which are known to affect the cholinergic system centrally and/or peripherally.

MATERIAL AND METHODS

Subjects

Male Swiss mice furnished by Roux-Ocefa Laboratories (Argentina) were used (age, 60–70 days; weight, 25–30 g). They were housed 10 to 12 in stainless-steel cages (cage size: 50 × 30 × 15 cm) with food and water freely available. In all cases, the mice housed in each single cage comprise an experimental group. The mice were kept in a regulated environment for at least 3 days before they were used, with lights on from 6:00 AM to 6:00 PM, at a room temperature of 23–25°C. All the experiments were carried out in accordance with internationally accepted principles and the local regulations concerning the care and use of laboratory animals.

Behavioral Procedure

Inhibitory avoidance behavior was studied in a one-trial learning, step-through type, inhibitory avoidance test situation (Boccia et al., 1996, 1998), which utilizes the natural preference of mice for a dark environment. The apparatus consists of a dark compartment (20 × 20 × 15 cm) with a stainless-steel grid floor and a small (5 × 5 cm) illuminated, elevated platform attached to its front center. The mice were not habituated to the dark compartment before the learning trial on day 1. During training, each mouse was placed on the platform and received a footshock (0.8 mA, 50 Hz, 1 s) as it stepped into the dark comparment. Forty-eight hours later each mouse was placed on the platform again and the step-through latency was recorded. If a mouse failed to cross within 300 s, the retention test was terminated and the mouse was assigned a score of 300. All experiments were conducted between 8.00 AM and 2.00 PM.

Drugs

The drugs used in these experiments were OT, the oxytocin receptor antagonist $d(CH_2)_5[Tyr(Me)^2, Thr^4, Thy\text{-}NH_2^9]$ OVT (Manning & Sawyer, 1993), which is referred as AOT, the anticholinesterase drugs physostigmine salicilate and neostigmine bromide, and the cholinergic receptor antagonists atropine sulfate, atropine methyl nitrate, mecamylamine hydrochloride, and hexamethonium hydrochloride. All drugs were obtained through Sigma Chemical Co. (St. Louis, Mo) except AOT, which was a gift of Dr. M. Manning, Department of Biochemistry and Molecular Biology, Medical College of Ohio. Oxytocin and AOT were dissolved in 0.05 M acetic acid, then diluted in saline, and given subcutaneously (10 ml/kg). The corresponding control groups received the same volume of acidified saline. Both the anticholinesterase drugs and the cholinergic blockers were dissolved in saline and were given intraperitoneally (10 ml/kg). The doses of both the anticholinesterase and the anticholinergic drugs were calculated as the free base, and the doses of all drugs were chosen on the basis of our previous results (Baratti & Kopf, 1996, Boccia et al., 1996, 1998; Faiman et al., 1991).

Experimental Groups

In the first set of experiments we replicated some previous findings about the effects of OT and AOT, and their interaction, on retention. Thus, different groups of 12 mice

each received, immediately after training, saline or AOT (0.03 or 0.30 μg/kg) and 10 min afterward they received saline or OT (0.10 μg/kg). Three additional groups ($N = 12$ mice per group) were injected with saline, OT (0.10 μg/kg), or AOT (0.30 μg/kg) 180 min after training to determine whether the peptide's effect on subsequent retention varied with the training–treatment interval. Finally, to assess a possible nonspecific proactive effect of peptide injections lasting more than 48 h, three other groups of 10 mice each did not receive footshock, but were injected with saline, OT (0.10 μg/kg), or AOT (0.30 μg/kg) immediately after training.

The second experiment was performed to determine whether the memory-impairing effects of OT would be reversed by the anticholinesterase drugs physostigmine and/or neostigmine (Taylor, 1996). For this purpose, 10 different groups of 12 mice each were submitted to the learning trial. Immediately after it, 5 groups of them received a sc injection of saline and 10 min later they were injected again, but by the ip route, with saline, physostigmine (35, 70, or 150 μg/kg), or neostigmine (150 μg/kg). The remaining 5 groups of mice received, immediately after training, OT (0.10 μg/kg, sc) and 10 min afterward were injected with saline, physostigmine (35, 70, or 150 μg/kg), or neostigmine (150 μg/kg).

Finally, we studied the possible interaction with regard to retention between AOT and four cholinergic receptor antagonists. In these experiments, immediately after training 10 different groups of 12 mice each received, in all cases by the ip route, saline, atropine (0.5 mg/kg), methylatropine (0.5 mg/kg), mecamylamine (5.0 mg/kg), or hexamethonium (5.0 mg/kg), followed 10 min apart by a sc injection of AOT (0.30 μg/kg).

Statistical Analysis

Data are expressed as median step-through latencies during the retention test and interquartile ranges, and differences between groups were estimated by individual Mann–Whitney U tests (two-tailed) (Siegel, 1956). In all cases p values less than .05 were considered significant.

RESULTS

Training step-through latency (preshock latency) differences among all the group of mice used in these experiments were not significant (10.3 ± 0.6 s; $F(24, 275) = 0.58$; $p > .05$) (one-way ANOVA) (Winer, 1971).

The main results of the first groups of experiments are shown in Table 1. The step-through latencies of mice given OT 10 min after training were significantly lower than those of saline controls ($U(12, 12) = 15$; $p < .01$), whereas the posttraining effects of AOT were dose-dependent. Thus, the retention latencies of mice injected with 0.03 μg/kg of AOT did not differ from those of the saline control group ($p > .05$), but the retention latencies of mice in the 0.30 μg/kg group were significantly higher than those of the saline controls ($U(12, 12) = 33$; $p < .05$). The impairing effects of OT on retention performance were prevented by AOT ($U(12, 12) = 93$; $p > .05$) in a dose without effect on its own ($p > .05$) (Table 1).

In addition, the retention latencies of mice injected with OT (0.10 μg/kg, sc) or AOT (0.30 μg/kg, sc) 180 min after training did not differ significantly compared with the

TABLE 1
Effect of Posttraining Administration of Oxytocin (OT) and an Oxytocin Receptor Antagonist (AOT) and Their Interaction on Retention

Treatment		Latencies to step-through (seconds)
Immediately after training	10 min after training	
Saline	Saline	117 (60–280)
Saline	OT (0.10 μg/kg)	34 (17–63)*
AOT (0.03 μg/kg)	Saline	123 (50–280)
AOT (0.30 μg/kg)	Saline	300 (196–300)**
AOT (0.03 μg/kg)	OT (0.10 μg/kg)	130 (53–280)

Note. Each entry represents the medians (and interquartile ranges) for 12 mice per group.

* $p < .01$ and ** $p < .05$, in both cases compared with the saline/saline-injected group (Mann–Whitney *U* test, two-tailed).

saline control group (Saline (180 min) = 120 (37–270) s; OT (180 min) = 130 (26–280) s; and AOT (180 min) = 126 (40–300) s; $U(12, 12) = 81$, $p > .05$; $U(12, 12) = 84$, $p > .05$, in both cases compared with the saline-injected group). Finally, the step-through latencies of mice that did not receive footshock but were injected with saline, OT (0.10 μg/kg, sc), or AOT (0.30 μg/kg, sc) immediately after training did not differ significantly from each other (Saline, 10 (4–13) s; OT, 12 (6–14) s; and AOT, 12 (8–15); $U(12, 12) = 67$, $p > 05$; $U(12, 12) = 69$, $p > .05$, in both cases compared with the saline–injected group).

As shown in Fig. 1, the low dose of physostigmine had a small nonsignificant difference ($p > .05$, compared with the saline/saline control group). However, this dose of physostigmine was able to partially attenuate ($U(12, 12) = 75$, $p > .05$, compared with the OT/saline-injected group) the impairing effects induced by OT on retention performance. The two higher doses of physostigmine significantly enhanced retention on their own ($U(12, 12) = 33$, $p < .05$, and $U(12, 12) = 23$, $p < .02$, respectively, compared with the saline/saline-injected control group). Thus, the retention latencies of the other two OT/physostigmine-injected mice were significantly higher than those of the OT/saline-injected mice ($U(12, 12) = 30$, $p < .02$, and $U(12, 12) = 15$, $p < .002$, respectively). It is worth noting that when physostigmine (70 or 150 μg/kg) was administered 10 min after OT, the enhancement of retention elicited in these conditions was smaller than that after saline ($U(12, 12) = 32$, $p < .02$, and $U(12, 12) = 21$, $p < .01$, compared with its respective control group). Neostigmine neither modified retention performance ($U(12, 12) = 102$, $p > .05$) nor influenced the effect of OT on it ($U(12, 12) = 93$, $p > .05$), compared with the OT/saline-injected group (Fig. 1).

Neither of the anticholinergic drugs affected retention performance by themselves ($p > .05$, in all cases compared with the saline/saline-injected group) (Fig. 2). The oxytocin receptor antagonist enhanced retention when given alone ($U(12, 12) = 19$, $p < .002$) or 10 min after methylatropine ($U(12, 12) = 21$, $p < .02$) or hexamethonium ($U(12, 12) = 19$, $p < .002$) injections, when the performance of each of these groups was compared with that of the saline/saline-injected group. However, the posttraining enhancing effects of AOT on retention performance were prevented by either atropine ($U(12, 12) = 93$,

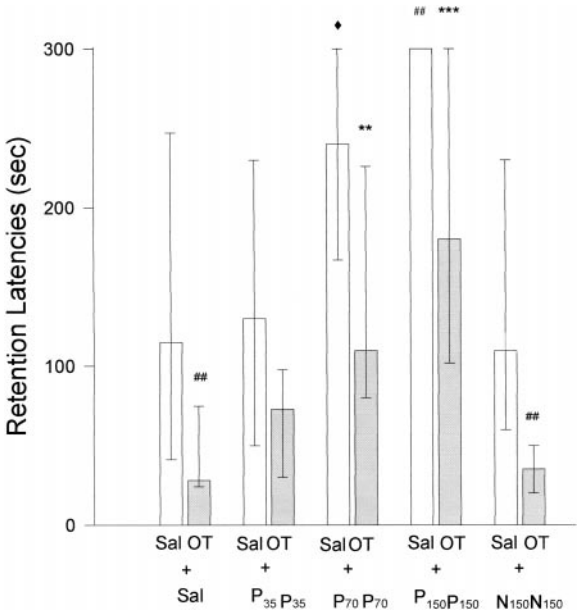

FIG. 1. Effects of posttraining administration of oxytocin (OT), physostigmine (P), or neostigmine (N), and their combinations, on retention test performance. Oxytocin (0.10 μg/kg, sc) was given immediately after training, whereas physostigmine (35, 70, or 150 μg/kg, ip) or neostigmine (150 μg/kg, ip) was given 10 min after saline or oxytocin. Each bar represents the medians and interquartile ranges for 12 mice per group. ##$p < .02$, ♦$p < .05$, compared with the saline/saline-injected control group. **$p < .02$, ***$p < .002$, compared with the OT/saline-injected group.

$p > .05$) or mecamylamine ($U(12, 12) = 93$, $p > .05$) (in both cases compared with the saline/saline-injected group).

DISCUSSION

The results of the first group of experiments indicate that the posttraining administration of oxytocin and an oxytocin receptor antagonist affected retention performance on an inhibitory avoidance response in mice in an opposite manner. Thus, OT impaired, whereas AOT enhanced, retention performance in mice that had received the aversive stimulus during the training trial. Previously, we demonstrated that the impairing effects of OT, and the enhancing ones of AOT, on retention performance were dose-dependent. As a consequence, the dose–response curve obtained with OT showed a U-shaped form, whereas an inverted-U dose–response curve was observed with AOT (Boccia et al., 1998). In addition, both OT and AOT injections were found to be less effective on retention performance if delayed by 180 min after training, suggesting action on posttraining neural or neurohumoral processes underlying the storage of recently acquired information (McGaugh, 1989). Furthermore, the effects of OT and AOT on retention performance are not attributable to nonspecific influences on response latencies since the peptides did not affect the retention latencies of unshocked control mice when given immediately after training.

The results of the first experiment also indicated that the impairing effects of OT on retention performance were prevented by the compound AOT, which is considered one

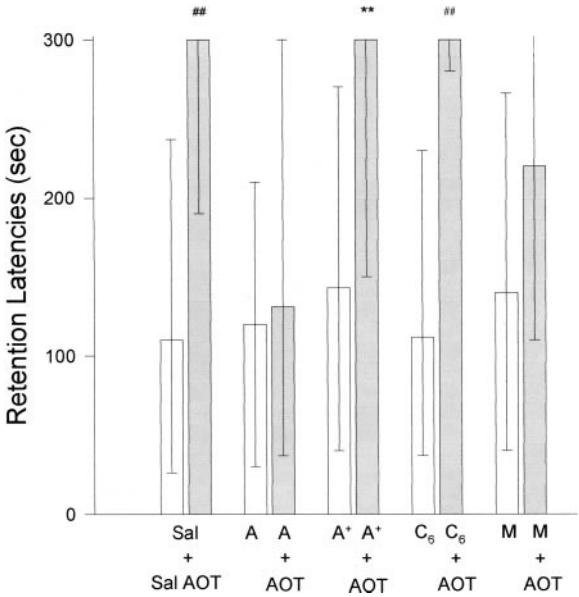

FIG. 2. Effects of an oxytocin receptor antagonist (AOT, 0.30 μg/kg, sc), atropine (A, 0.5 mg/kg, ip), methylatropine (A^+, 0.5 mg/kg, ip), mecamylamine (M 5.0 mg/kg, ip), or hexamethonium (C_6 5.0 mg/kg, ip), and their combinations, on retention test performance. The cholinergic antagonists were given immediately after training, whereas AOT was given 10 min afterward. Each bar represents the medians and interquartile ranges for 12 mice per group. ##$p < .002$, **$p < .02$, compared with saline/saline-injected control group.

of the most potent oxytocin receptor antagonists (Elands et al., 1988), at a dose without effect by itself. In previous unpublished experiments, the same dose of AOT was ineffective when given alone to mice trained with a lower intensity of footshock than that used in this work. This fact suggests that the interaction between OT and AOT cannot be due to a change in baseline avoidance performance.

The results discussed above are remarkably consistent with those of our previous study (Boccia et al., 1998) and indicate that a single peripheral posttraining administration of OT impairs retention of an inhibitory avoidance response in mice. They also suggest that the effects of OT may be mediated through an interaction with specific receptors. In this sense, we reported (Boccia et al., 1998) that the effects of OT on retention were only prevented by AOT and, on the contrary, OT impaired retention in mice pretreated with the V_{1a} vasopressin receptor antagonist $d(CH_2)_5[Tyr(Me)^2]AVP$ (Manning & Sawyer, 1993). Conversely, the vasopressin receptor antagonist was able to prevent the enhancement of retention induced by posttraining administration of lysine or arginine vasopressin (Boccia et al., 1996; Faiman et al., 1991), as well as the effects of peripheral stimuli that induce a release of endogenous vasopressin (Baratti, Faiman, & de Erausquin, 1989; Boccia et al., 1996). Further, the enhancing effects of AVP on retention were not prevented by AOT (Boccia et al., 1998). The behavioral findings observed after posttraining administration of OT are, in general, in agreement with those obtained by other authors in other experimental conditions (Boccia & Baratti, 1999; Bohus, Kovacs, & De Wied, 1978a, 1978b; Engelmann et al., 1996; Kovacs & De Wied, 1994), while the use of the vasotocin analogue AOT as an oxytocin receptor antagonist, which not only enhanced retention by itself, but also prevented the impairing effects of OT on retention, is a new finding. To

the best of our knowledge, there are no published data concerning this issue, except our own previous studies (Boccia et al., 1998; Boccia & Baratti, 1999).

In any case, the interpretation of data from pharmacological studies using the peripheral route of administration, as reported here, may be in some respects uncertain in a physiological context (Engelman et al., 1996). Thus, in general, peptides, including OT, pass the blood–brain barrier with difficulty (Zaidi & Heller, 1974), and there are indications suggesting that only 0.002% of subcutaneous-administered OT reached the cerebrospinal fluid within the first 10 min postinjection (Landgraf, 1995; Mens, Witter, & van Wimersma Greidanus, 1983). Further, since the concentration of this type of neuropeptide in the extracellular fluid of discrete brain areas is several orders of magnitude higher than that in plasma (Landgraf, Neumann, & Schwarzberg, 1988), the central content of OT, and likely also of AOT, should not be affected significantly by the alteration of peripheral peptide concentration following its subcutaneous administration (Engelman et al., 1996). If so, these findings suggest that both OT and AOT treatments may alter retention performance of the inhibitory avoidance task without directly influencing memory processes at the brain level. Accordingly, the memory hypothesis of the neurohypophyseal peptides (De Wied et al., 1993; Kovacs & Telegdy, 1982) has been modified and explained by effects on arousal and autonomic peripheral mechanisms (Le Moal et al., 1984; Sahgal, 1984). Briefly, the existence of a chain of events was suggested beginning with endocrinological–physiological effects, as a consequence of hormone release in the periphery (Packard, Williams, Cahill, & McGaugh, 1995), through aversive effects and arousal alertness, finally to altered behavioral performance in a specific learning task (Le Moal et al., 1984).

The main aim of the present study was to examine a possible interaction, with respect to retention, between oxytocin or its putative receptor antagonist and drugs which are known to influence the activity of cholinergic mechanisms both centrally and/or peripherally, in order to delineate their implication in the behavioral action of OT.

From the results of the second experiment it is clear that the memory-impairing effects of OT were reversed by the central acting anticholinesterase physostigmine (Taylor, 1996) administered 10 min after the peptide. The low dose of physostigmine, which did not affect retention when given alone, was partially effective; however, the higher doses of the anticholinesterase, which were effective on their own, completely reversed the impairing effects of OT on retention. However, in these conditions the anticholinesterase did not fully elicit its usual memory-enhancing actions (Baratti & Kopf, 1996; Flood, Landry, & Jarvik, 1981). These findings argue against the interpretation that the effects of physostigmine are due to a generalized memory facilitation caused by the anticholinesterase (Martinez, Jensen, & McGaugh, 1981) and that physostigmine might mask the effects of OT on memory. In general, the characteristic pharmacological effects of physosotigmine are due primarily to the prevention of hydrolysis of acetylcholine by acetylcholinesterase at sites of cholinergic neurotransmission (Taylor, 1996); the resulting increase in extracellular acetylcholine concentration could restore a central cholinergic hypofunction and, eventually, improve memory. In addition during the posttraining period that follows a learning experience and that remains susceptible to modulatory influences (McGaugh, 1989), there is an increase in the brain cholinergic tone (Baratti & Kopf, 1996; Matthies, Rauca, & Liebman, 1974; Ragozzino, Unick, & Gold, 1996; Rauca, Kammerer, & Matthies, 1980; Toumane, Durking, Marighetto, Galey, & Jaffard, 1988) which correlates well with an enhanced retention performance in several learning tasks. All the findings discussed

above, when considered together, suggest that an enhancement of the posttraining central cholinergic tone elicited by physostigmine may be responsible for the observed interaction between the anticholinesterase and OT on retention. Accordingly, it is also tempting to suggest that OT impairs retention by decreasing central cholinergic activity. The fact that the peripheral acting anticholinesterase neostigmine (Taylor, 1996) did not prevent the actions of OT on retention performance add support to that suggestion. However, the nature of the mechanism(s) for the possible actions of OT on central cholinergic activity cannot definitively be elucidated from the behavioral data reported here. Thus, a direct estimation of the release of endogenous OT concomitantly with behavioral training and/or testing was not performed. Therefore, essential information about neuropeptide involvement in the behavioral performance being studied is lacking.

Notwithstanding the limitations discussed above, the results obtained with the oxytocin receptor antagonist AOT provide another indication of the possible participation of central cholinergic mechanisms in the behavioral actions of OT. In fact, the effects of AOT on retention were not only the opposite of those induced by OT, but also were prevented by two central acting cholinergic receptor antagonists, atropine (Brown & Taylor, 1996) and mecamylamine (Milne, Rowe, Sommers, Muehrcke, & Crawford, 1957), when given immediately after training, but 10 min prior to AOT treatment, and in a dose that did not affect retention performance by itself. Atropine, like scopolamine, is a nonselective central acting cholinergic muscarinic receptor antagonist (Brown & Taylor, 1996), whereas mecamylamine is a centrally active cholinergic antagonist (Milne et al., 1957) which, however, exhibits both competitive and noncompetitive actions on nicotinic receptors (Martin, Suchocki, May, & Martin, 1990) and also possesses other pharmacological properties, probably related to the NMDA–glutamate receptor subtype (O'Dell & Christiensen, 1988). As antagonists of cholinergic neurotransmission, both atropine and mecamylamine, when given in an appropiate dose immediately after a learning trial, reduce posttraining cholinergic tone and impair retention (Bammer, 1982; Levin, 1992). It is also possible that at doses which have no effect on retention when given alone, atropine and/or mecamylamine prevent the memory-enhancing effects of some treatments that directly or indirectly influence the activity of cholinergic mechanisms. Thus, we previously demonstrated that atropine prevents the enhancement of retention induced by the muscarinic agonist oxotremorine and the anticholinesterase physosotigmine (Baratti, Huygens, Miño, Merlo, & Gardella, 1979), the opioid receptor antagonist naloxone (Baratti, Introini, & Huygens, 1984), the muscarinic M_2 presynaptic receptor antagonist AF-DX 116 (Baratti, Opezzo, & Kopf, 1993), and glucose (Kopf & Baratti, 1994), whereas mecamylamine prevents the memory-enhancing effects of nicotine (Faiman et al., 1991), a central nicotinic cholinergic agonist (Levin, 1992). It is worth pointing out that these effects were only observed following the administration of atropine or mecamylamine but not its pharmacological quaternary analogues methylatropine and hexamethonium, respectively, which cross the blood–brain barrier poorly after systemic administration (Asghar & Roth, 1971; Breezenoff, Xiao, & Vargas, 1988). All together, the facts discussed above support the hypothesis that the oxytocin receptor antagonist may exert a desinhibitory effect on central cholinergic activity.

In summary, the results of the present experiments provide pharmacological evidence suggesting that the activity of both central muscarinic and nicotinic cholinergic mechanisms are negatively modulated by oxytocin during the posttraining period that follows an

aversively motivated learning task in mice. Although the present work reveals an emerging mechanism through which oxytocin may influence memory storage, the role of interactions between acetylcholine and other neurotransmitters affecting memory (Decker & McGaugh, 1991) cannot be ignored in order to explain the present results. Further, additional investigations should use other tasks, making possible more general conclusions. In this light, we recently reported an impairing effect of OT on retention of a nose-poke habituation response in mice (Boccia & Baratti, 1999).

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