Pharmacology Biochemistry and Behavior, Vol. 42, pp. 75–83, 1992
Printed in the U.S.A.

0091-3057/92 $5.00 + .00

Ondansetron and Arecoline Prevent Scopolamine-Induced Cognitive Deficits in the Marmoset

G. J. CAREY,* B. COSTALL,* A. M. DOMENEY,*[1] P. A. GERRARD,* D. N. C. JONES,* R. J. NAYLOR* AND M. B. TYERS†

*Postgraduate Studies in Pharmacology, School of Pharmacy, University of Bradford, Bradford BD7 1DP, UK
†Neuropharmacology Department, Glaxo Group Research Ltd., Ware, Herts. SG12 0DP, UK

Received 25 July 1991

CAREY, G. J., B. COSTALL, A. M. DOMENEY, P. A. GERRARD, D. N. C. JONES, R. J. NAYLOR AND M. B. TYERS. *Ondansetron and arecoline prevent scopolamine-induced cognitive deficits in the marmoset.* PHARMACOL BIOCHEM BEHAV 42(1) 75–83, 1992. – The cognitive-enhancing potential of the 5-hydroxytryptamine (5-HT) selective 5-HT_3 receptor antagonist, ondansetron, was investigated in a model of cognitive impairment induced by the muscarinic receptor antagonist, scopolamine. For this purpose, marmosets were trained in an object discrimination task utilizing the Wisconsin General Test Apparatus. Administration of scopolamine (0.01–0.04 mg/kg, SC) caused a dose-dependent impairment in the acquisition of the object discrimination task in that marmosets required more trials to reach criterion, made more errors, and took longer to choose the objects. Administration of arecoline (0.06–0.1 mg/kg, SC) or 1,2,3,9-tetrahydro-9-methyl-3-[(2-methyl-1*H*-imidazol-1-*yl*)methyl]-4*H*-carbazol-4-one,HCl·$2H_2O$ (ondansetron) (0.1–1 μg/kg, SC) prevented the scopolamine-induced impairment in task acquisition in that the performance of marmosets was indistinguishable from that of saline-treated animals and was significantly better than that following scopolamine/saline. From these studies, we conclude that ondansetron prevents impairment in the cognitive performance of marmosets induced by administration of scopolamine.

5-HT_3 receptor antagonist Ondansetron Marmoset Scopolamine Cognition Object discrimination task

BARTUS and coworkers (8) proposed a central role for the cholinergic system in memory and learning when forwarding their "cholinergic hypothesis of geriatric memory dysfunction." This hypothesis has subsequently received support from both clinical and animal research (2,9,20,32,44).

However, despite the well-accepted importance of the cholinergic system in the pathology of dementias, clinical attempts to restore the cholinergic system directly have proved disappointing. Although studies carried out in Alzheimer's disease patients using muscarinic receptor agonists, cholinesterase inhibitors, and cholinergic releasing agents have shown these treatments to have significant effects on cognition, improvements have generally been small, variable, and offered little or no relief in daily living conditions (9). Tacrine or 1,2,3,4-tetrahydro-5-aminoacridine (THA), a cholinesterase inhibitor, has been reported to be a useful, long-term palliative treatment in Alzheimer's disease (41), although other studies have not replicated the beneficial effects of this agent and have indicated a risk of hepatotoxicity (18). Clinical trials with cholinergic precursors have also shown negligible beneficial effects [see reviews, (9,30)]. Thus, the equivocal evidence obtained with such compounds has led many workers to question the value of treating dementias with the presently available cholinergic drugs.

The cholinergic system is by no means the only neurotransmitter system implicated in the impaired cognitive function of aged or demented patients. Indeed, changes in the catecholaminergic, serotonergic, and peptidergic systems have been reported [see reviews, (1,32,39)]. Of particular pertinence to the present study are the changes observed in the serotonergic system (1,4,31,39). In animal studies, there is evidence for 5-hydroxytryptamine (5-HT) receptor-mediated modulation of acetylcholine (ACh) release from the striatal slices (24), while in areas such as the cerebral cortex, striatum, and hippocampus there is a well-documented serotonergic inhibitory control of cholinergic function (10,11,38). Furthermore, Barnes et al. (5) demonstrated a 5-HT_3 receptor-agonist-mediated reduction in [^{3}H]ACh release from rat entorhinal cortex in vitro that could be blocked by selective 5-HT_3 receptor antagonists such as ondansetron. 5-HT_3 receptor antagonists

[1] To whom requests for reprints should be addressed.

have also been demonstrated to possess cognitive-enhancing activity in both rodents (6,14,16) and common marmosets (6,19).

In the present study, we investigated the effects of scopolamine on the acquisition of an object discrimination task in the marmoset and the ability of the 5-HT_3 receptor antagonist 1,2,3,9-tetrahydro-9-methyl-3-[(2-methyl-1*H*-imidazol-1-*yl*) methyl]-4*H*-carbazol-4-one,HCl·$2H_2O$ (ondansetron) to overcome the resulting cognitive impairment.

METHOD

Animals

Six marmosets (male and female; there were no apparent differences in performance between either sex) weighing 310–370 g were housed in single-sex pairs. Animals were given free access to water and food (Mazuri primate diet, SDS Ltd., Essex), which they received in the morning. The remainder of the diet (fruit, brown bread, and malt loaf) was given between 1600 and 1700.

Holding rooms were maintained at 25 ± 1°C at a humidity of 55%. Rooms were illuminated for 12 h, followed by a 12 L : 12 D cycle, lights being on between 0700 and 1900. Simulated dawn and twilight periods were programmed to occur 0.5 h before and after the main lights came on or went off. During the 12-h dark period, a single 60-W red bulb was illuminated to avoid complete darkness.

Assessment of Performance of Marmosets Using the Wisconsin General Test Apparatus

Apparatus. Studies assessed the performance of marmosets in a miniature version of the Wisconsin General Test Apparatus (WGTA). This consisted of a box (45 × 42 × 45 cm) with an opaque shutter at one end to separate the marmoset from the operator. The shutter could be raised and lowered by the operator to reveal a tray containing two food wells located 14 cm apart. The marmoset was able to place its arms between the bars to reach the food wells and displace an object to obtain a reward. During testing, the marmoset was placed behind the shutter in a transport cage large enough to allow the marmoset complete freedom to move during the test session.

The marmoset could be viewed by the operator via a one-way screen. This screen was located above a hinged flap through which the operator could bait the test tray containing the food wells. The interior of the WGTA was illuminated by a 15-W strip light; all testing took place in a darkened room.

Initial training in object discrimination learning. Throughout initial training, animals were presented with a series of trials in the WGTA in which the shutter was raised to reveal two plastic junk objects (e.g., a rubber bung and a hypodermic needle case) covering two food wells. Selection of only one object (a rubber bung) provided a food reward. Food rewards consisted of syrup-coated cubes (0.5 cm) of brown bread. The left/right position of the rewarded object was varied between trials according to a pseudorandom schedule (23). The intertrial interval was maintained constant at 15 s and each trial lasted until the marmoset made a response (i.e., selected one of the objects).

In an average daily test session, animals generally completed on the order of 40 trials. Animals were trained until they were able to perform the above task to an accuracy of 90% correct responses of 40 trials. Initial object discrimination training to this criterion (i.e., 90 correct responses of 100) took 1–3 weeks. Once animals were achieving 90% accuracy on the above task, they were tested over a smaller number of trials, 20 and then 10, to ensure that this level of performance was maintained (e.g., an animal giving 90 correct responses of 100 should then give correct responses on the order of 18 of 20 and 9 of 10). When consistency of performance was verified, animals could then be utilised to assess the ability of agents to influence object discrimination learning.

Test protocol. In this task, the basic test system was identical to that of simple object discrimination training with the exception that animals were introduced to two novel junk objects that were subject to a changing reward contingency over different test days. The rewarded object was determined using a balanced, crossover design in which individual marmosets received injections with each drug at each dose level with a one-week interval between drug treatments (Table 1).

On the first day of testing, the marmoset was required to displace one of two plastic junk objects covering a food well to collect the reward. The task was completed once the animal had selected the rewarded object in 9 of 10 trials. On the following day (day 2), the previously unrewarded object became rewarded (acquisition task) and this reward contingency was continued on day 3 (retention task). On day 4, the object rewarded on day 1 became, once again the rewarded object (acquisition task) and this was reversed on day 5 (reversal task). In subsequent weeks of testing, the rewarded object on day 1 was the object that had been rewarded on the previous day 5.

To obtain a stable baseline of performance, marmosets were trained using this protocol for at least 2 weeks prior to the commencement of drug studies.

Data Analysis

Data were expressed in several ways:

1. The number of trials required to reach criterion (9 correct responses of 10 trials) for the acquisition of the task and the subsequent 24-h retention and reversal.
2. Choice latency(s) calculated using the formula:

$$\text{choice latency} = \frac{\text{total time to complete test(s)-(no. of trials} - 1) \times 15\text{ s}}{\text{total number of trials}},$$

   where 15 s = intertrial interval and (no. of trials − 1) = number of intertrial intervals.
3. Number of errors: The mean errors per task were calculated.
4. Learning curves: Learning curves were constructed using the method of Ridley et al. (35) by calculating the total number of errors made by all marmosets for each consecutive block of five trials for the acquisition task and its subsequent 24-h retention and reversal. This provided a measure of the accuracy of performance in the trials required before reaching criterion.

It was particularly important that the behavioural status of animals was also recorded as scopolamine has previously been shown to cause behavioural agitation in the marmoset at higher doses [(36) and our preliminary studies].

All testing took place between 1000 and 1300 on weekdays only.

Drugs

Scopolamine HBr (Sigma), arecoline HBr (Sigma), and ondansetron (Glaxo Group Research Ltd.) were dissolved in ster-

TABLE 1

EXPERIMENTAL DESIGN TO ASSESS THE INFLUENCE OF ARECOLINE AND ONDANSETRON ON SCOPOLAMINE-INDUCED IMPAIRMENT IN THE PERFORMANCE OF AN OBJECT DISCRIMINATION TASK

	Week 1					Week 2				
Days	1	2	3	4	5	1	2	3	4	5
Rewarded junk object	1	2	2	1	2	2	1	1	2	1
Relation to previous day	RETn	REV	RETn	REV	REV	RETn	REV	RETn	REV	REV
Drug treatment	–	SCOP (0.01–0.05 mg/kg, SC)	–	VEH	–	–	VEH	–	SCOP (0.01–0.05 mg/kg, SC)	–
Study 1										
Study 2	OND or VEH BID	SCOP or VEH (0.02 mg/kg, SC) + either OND, AREC, or VEH	–	–	–	OND or VEH BID	SCOP or VEH (0.02 mg/kg, SC) + either OND, AREC, or VEH	–	–	–

SCOP, scopolamine; VEH, vehicle; OND, ondansetron (0.01–1 μg/kg, SC); AREC, arecoline (0.06–0.1 mg/kg, SC); RETn, retention of previous day; REV, reversal of previous day; –, no treatment.

ile saline and administered in a volume of 1 ml/kg body weight by the subcutaneous route. Doses are expressed as the free drug.

Statistical Analysis

A matched-pairs *t*-test was utilised to compare the effects of drug treatments on the mean trials to criterion, mean choice latency, and mean errors with those of the corresponding vehicle treatment.

RESULTS

Study 1: Influence of Scopolamine on Object Discrimination and Subsequent 24-h Retention or Reversal

In the first part of this study, scopolamine (0.01–0.05 mg/kg, SC) or saline (1 ml/kg, SC) were administered 30 min before the acquisition of an object discrimination task on days 2 and 4 according to the experimenter-blind, crossover design detailed in Table 1. Throughout the study, the experimenter was unaware of the specific treatment received by animals.

Scopolamine (0.01–0.04 mg/kg, SC) caused a dose-dependent impairment in the performance of this task when compared with that following saline treatment (Figs. 1A–C). Following administration of scopolamine (0.02 and 0.04 mg/kg, SC), these changes reached statistically significant levels such that the mean number of trials required to reach criterion, mean errors per task and, for the higher dose only, mean choice latency were elevated ($p < 0.05$–0.01). For example, scopolamine (0.02 mg/kg, SC) caused an increase in trials required to reach criterion from 7.1 ± 1.6 (saline pretreatment) to 14.3 ± 3.4 and in the mean errors per task from 4.9 ± 0.7 (saline pretreatment) to 9.5 ± 2.1 ($p < 0.05$) (Figs. 1D–F).

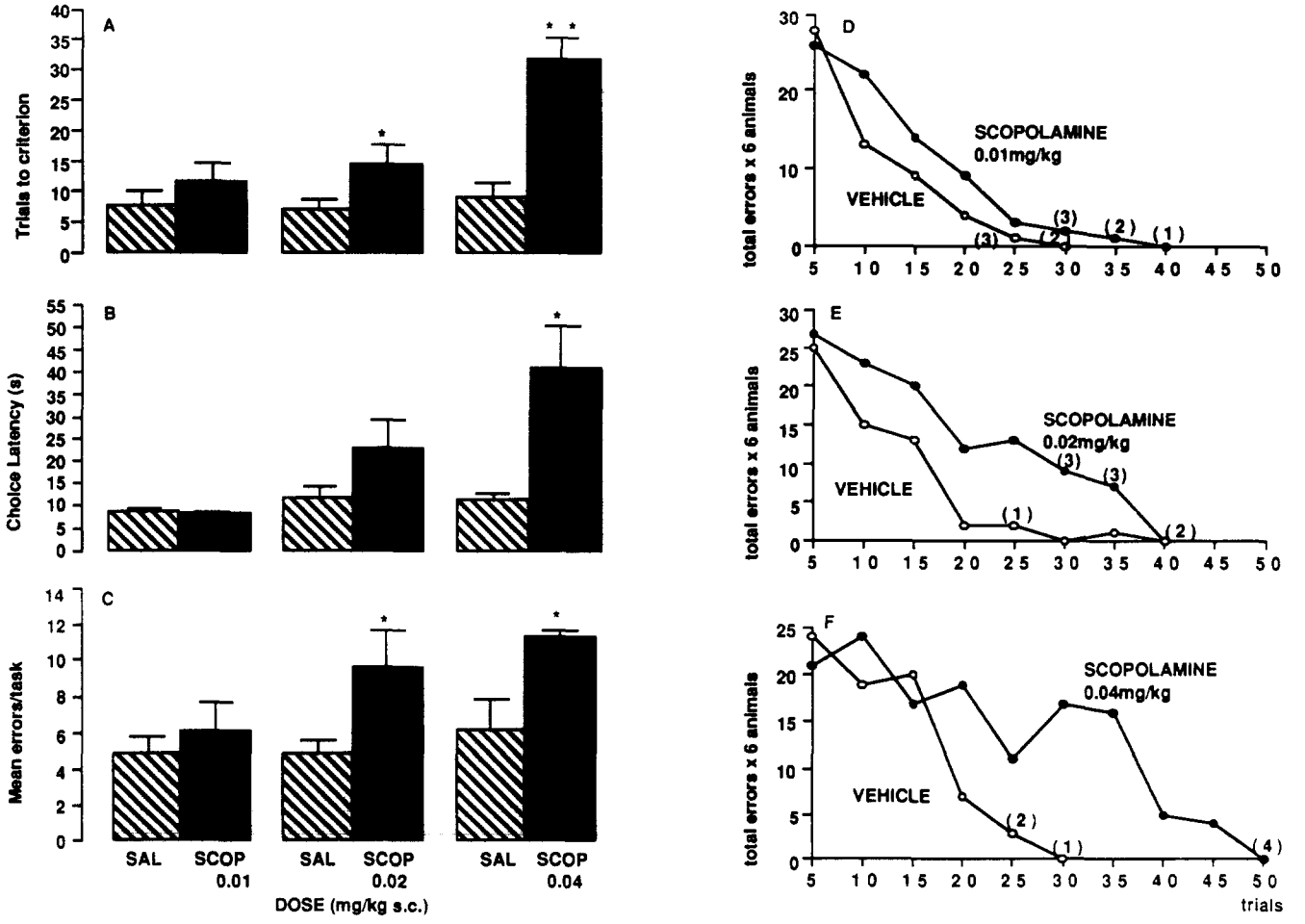

FIG. 1. Ability of scopolamine (SCOP) to impair the acquisition of an object discrimination task. Data are presented as (A) the mean number of trials to reach criterion (9 correct responses of 10 trials), (B) mean choice latency, and (C) mean errors per task. Significant performance impairments (scopolamine compared with appropriate-vehicle treatment; SAL) are indicated by $*p < 0.05$ and $**p < 0.01$ (matched-pairs *t*-test, $n = 6$). D, E, and F represent the learning curves for the completion of the acquisition task. These were obtained by calculating the number of errors in blocks of five trials by all six animals for the first, second, and third, etc. block using the method of Ridley et al. (35). The beginning of each curve represents the total number of errors made by all six animals tested for both the saline or scopolamine treatments in each 2-week crossover period (see Table 1). The numbers in parentheses indicate when fewer animals are involved since some animals may have reached criterion.

Administration of a higher dose of scopolamine (0.05 mg/kg, SC) caused behavioural effects incompatible with performance of this task. These were an unwillingness or inability to choose an object or consume the food reward and behavioural agitation. In contrast, administration of a lower dose of scopolamine (0.02 mg/kg, SC) produced negligible side effects and this dose was selected for use in the subsequent studies to assess the influence of ondansetron and arecoline.

On testing the retention of an object discrimination task that was acquired 24 h previously under the influence of scopolamine (0.02 or 0.04 mg/kg, SC), there was a trend toward an impairment in performance. However, this failed to achieve statistical significance (Figs. 2A and C). There was also a tendency for marmosets to find it easier to reverse from a task acquired 24 h previously under the influence of scopolamine (0.02 and 0.04 mg/kg, SC) such that the mean trials required to reach criterion and the number of errors made were lower than those of saline-treated animals (Figs. 2B and D) although, again, these changes did not reach significance. Choice latency remained unaffected by scopolamine treatment 24 h previously (data not shown).

Study 2: Influence of Arecoline and Ondansetron on Scopolamine-Induced Impairment in the Acquisition of an Object Discrimination Task

Since scopolamine failed to cause statistically significant changes in the retention or reversal of an object discrimination task, the experimental design utilised in Study 1 was modified such that Study 2 assessed only the influence of arecoline and ondansetron upon the scopolamine-induced impairment in the acquisition of an object discrimination task (see Table 1).

Concomitant administration of arecoline (0.06, 0.08, or 0.1 mg/kg, SC) with scopolamine (0.02 mg/kg, SC) attenuated the impairment in task acquisition to a level where the performance of marmosets was not statistically different from that following saline treatment alone. For example, treatment with arecoline (0.1 mg/kg, SC) reduced the number of trials required by scopolamine-treated animals to reach criterion from 21.7 ± 3.9 (saline/scopolamine) to 10.5 ± 3.6 (Fig. 3A) and caused a leftward displacement of the learning curves with respect to those for saline/scopolamine-treated marmosets (Fig. 3E).

However, two of the six animals tested that received areco-

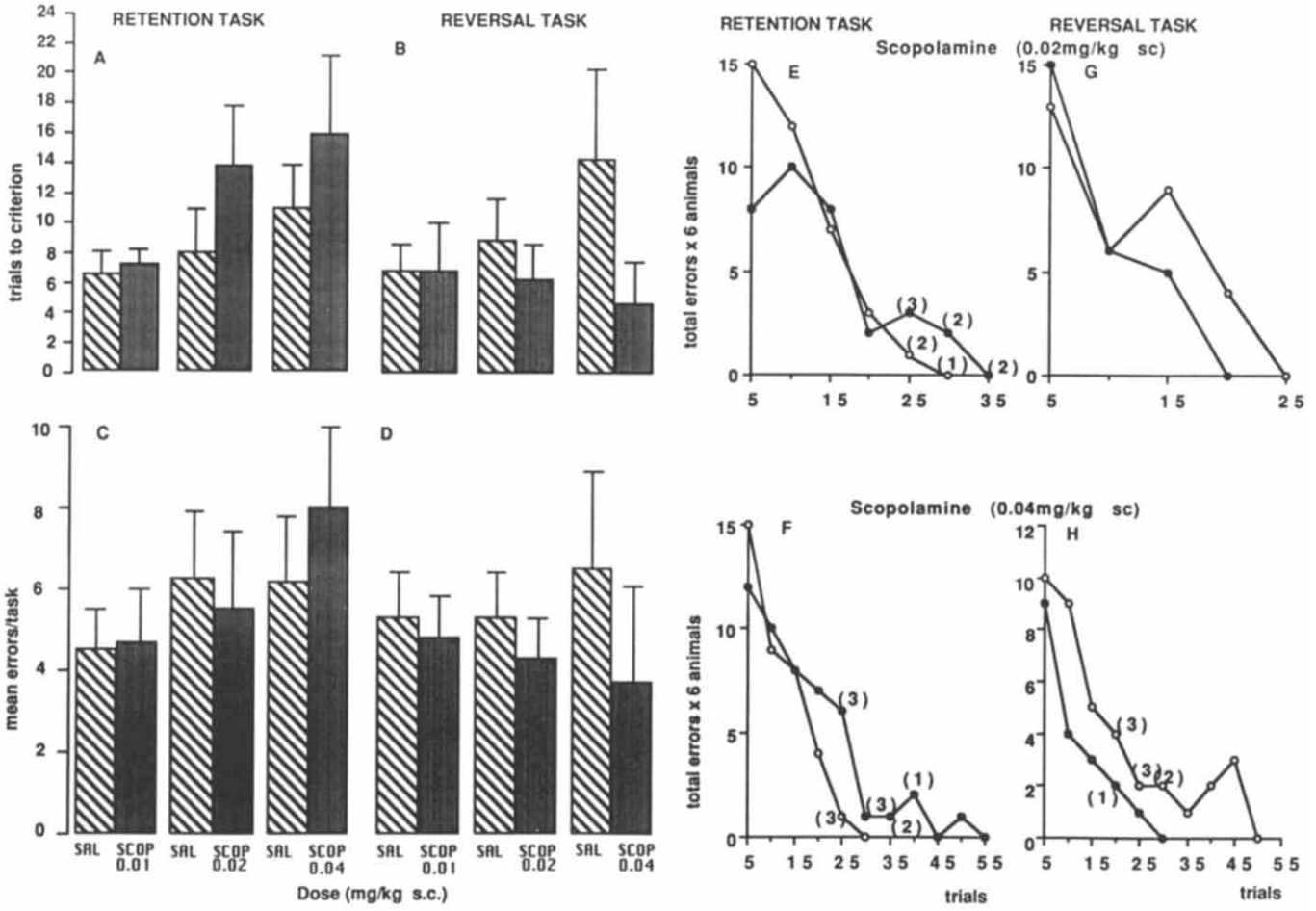

FIG. 2. Influence of scopolamine (SCOP) (administered 24 h previously, 30 min before the acquisition task) on the retention or reversal of an object discrimination task. Data are presented as (A and B) the mean number of trials to reach criterion (9 correct responses of 10 trials) and (C and D) the mean errors per task for the retention and reversal of the task, respectively. No significant differences between the performance of scopolamine and saline-treated (SAL) marmosets were recorded ($p > 0.05$, matched-pairs t-test, $n = 6$). E-H represent the learning curves for the completion of the retention (E and F) and reversal (G and H) tasks. The beginning of each curve represents the total number of errors in blocks of five trials made by all six animals tested. ○—○ represents the performance of saline-treated marmosets, while the performance of scopolamine-treated marmoset is represented by ●—●. The numbers in parentheses indicate when fewer animals are involved since some animals may have reached criterion.

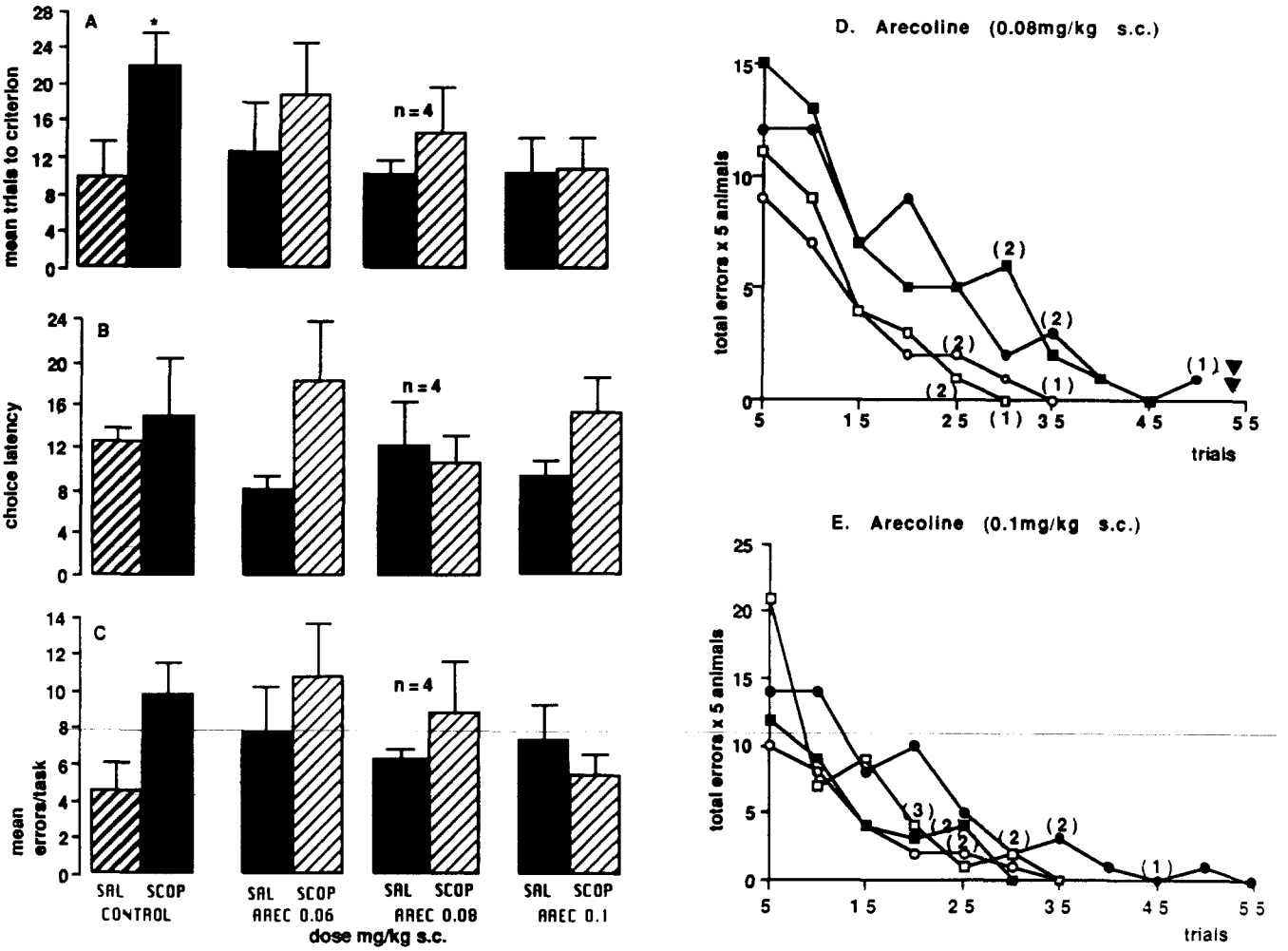

FIG. 3. Influence of arecoline on scopolamine-induced impairment in the acquisition of an object discrimination task. Arecoline was administered concomitantly with saline or scopolamine (0.02 mg/kg, SC) 30 min before testing. Data are presented as (A) the mean number of trials to reach criterion, (B) mean choice latency, and (C) mean errors per task. Significant performance impairments (scopolamine compared with appropriate-vehicle treatment) are indicated by $*p < 0.05$ (matched-pairs t-test, $n = 6$ unless indicated). D and E represent the learning curves for the completion of the acquisition task. The beginning of each curve represents the total number of errors in blocks of five trials made by all six animals (five animals for E). The numbers in parentheses indicate when fewer animals are involved since some animals may have reached criterion. The performance of animals following saline (SAL) control treatment is represented by ○—○, saline/scopolamine (SCOP) treatment by ●—●, arecoline (AREC) saline treatment by □—□ and arecoline/scopolamine treatment by ■—■. Following the combined administration of arecoline (0.08 mg/kg, SC) and scopolamine, one marmoset was unable to complete the task (indicated by ▼ in figure D), while another marmoset failed to make any choices.

line (0.08 mg/kg, SC) and scopolamine (0.02 mg/kg, SC) were unable to complete the task; indeed, one of these animals refused to make any choices. These two marmosets displayed behavioural changes similar to those observed following treatment with the highest dose of scopolamine (0.05 mg/kg, SC).

In a separate study, ondansetron (0.01, 0.1, or 1 μg/kg, SC) or saline were administered three times in the 24 h preceding the acquisition task (i.e., at 0800 and 1700 on day 1 and between 0800 and 0930 on day 2) (Table 1). Pretreatment with ondansetron (0.1 or 1 μg/kg, SC) prevented scopolamine-induced impairment in task acquisition. The performance of ondansetron/scopolamine animals was not significantly different ($p > 0.05$) from that of saline-treated marmosets, but significantly better ($p < 0.05$) than that of saline/scopolamine-treated animals (Figs. 4A-C). For example, the mean trials required to reach criterion were reduced from 17.2 ± 4.0 (saline/scopolamine, $n = 5$) to 5 ± 2.2 (ondansetron 0.1 μg/kg, SC/scopolamine, $p < 0.05$) and from 24.3 ± 6.2 (saline/scopolamine, $n = 6$) to 10.5 ± 3.5 (ondansetron 1 μg/kg, SC/scopolamine, $p < 0.05$), respectively. The ability of ondansetron to prevent scopolamine-induced impairment is further revealed by the examination of the learning curves constructed for this task (Figs. 4D and E). Pretreatment with ondansetron (0.1 or 1 μg/kg, SC) caused a marked leftward displacement of learning curves for scopolamine-treated marmosets with respect to those for the corresponding saline/scopolamine treatments. Indeed, ondansetron (0.1 but not 1.0 μg/kg, SC) produced a significant reduction in the mean errors per task from 10.2 ± 1.8 (saline/scopolamine, $n = 5$) to 3.4 ± 0.9 ($p < 0.05$, Fig. 4C).

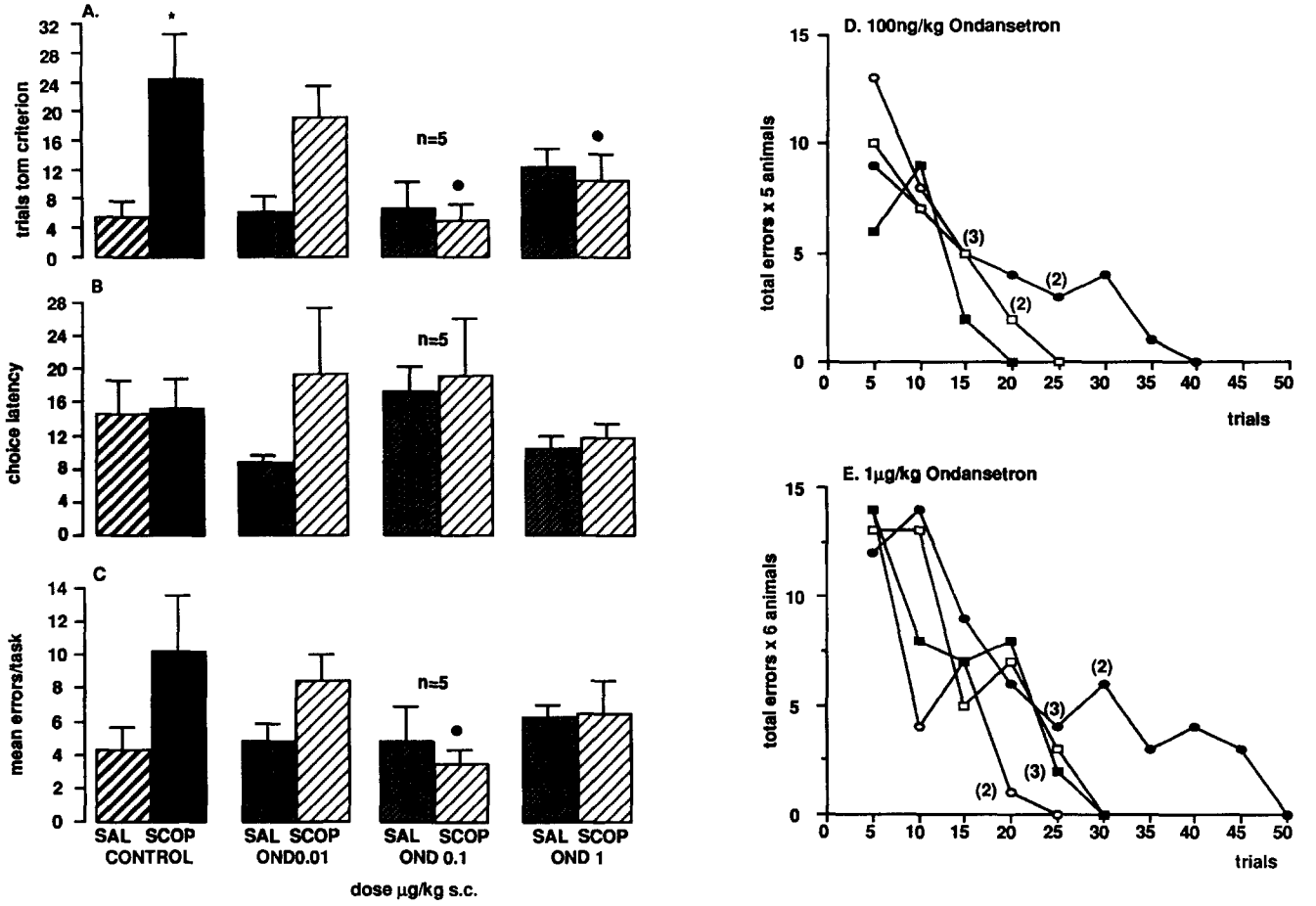

FIG. 4. Influence of ondansetron on scopolamine-induced impairment in the acquisition of an object discrimination task. Ondansetron (OND) was administered three times in the 24 h preceding saline (SAL) or scopolamine (SCOP; 0.02 mg/kg, SC) treatment. Data are presented as (A) the mean number of trials to reach criterion, (B) mean choice latency, and (C) mean errors per task. Significant performance impairments (scopolamine compared with appropriate-vehicle treatment) are indicated by $*p < 0.05$, while significant prevention of impairments (ondansetron/scopolamine compared with saline/scopolamine) are indicated by $\bullet p < 0.05$ (matched-pairs t-test, $n = 6$ unless indicated). D and E represent the learning curves for the completion of the acquisition task. The beginning of each curve represents the total number of errors in blocks of five trials made by all six animals (five animals for E). The numbers in parentheses indicate when fewer animals are involved since animals may have reached criterion. The performance of animals following saline (SAL) control treatment is represented by ○—○, saline/scopolamine treatment by ●—●, ondansetron (OND)/saline treatment by □—□ and ondansetron/scopolamine treatment by ■—■.

Administration of either arecoline or ondansetron failed to influence the performance of saline-treated marmosets in the paradigm described.

DISCUSSION

The major findings of the present studies are that a specific cholinergic cognitive impairment in the marmoset may be reversed by both a muscarinic agonist, arecoline, and by a selective 5-HT_3 receptor antagonist, ondansetron. The administration of scopolamine before the acquisition of an object discrimination task in doses similar to those previously reported for the marmoset (35,36) and for other primates (8) produced a dose-dependent increase in the number of trials required to reach criterion, mean task errors, and mean choice latency. One hypothesis under test was that animals would show deficits in the retention of a task acquired under the influence of scopolamine and would, consequently, also find it easier to reverse from that task when compared with the case following saline treatment. However, no statistically significant effects were seen. It might be suggested that the performance of marmosets was near maximal in all tasks assessed and that any slight differences in the memory of the marmosets between the acquisition of a task and its subsequent retention or reversal were only revealed following scopolamine treatment. This is particularly apparent when a comparison is made between the performance of the retention and reversal tasks of the object discrimination that had been acquired under the influence of scopolamine (0.04 mg/kg, SC). Ridley and colleagues (35) reported less equivocal results, for example, marmosets found the reversal of a position discrimination acquired under the influence of saline to be substantially more difficult than its retention, while the situation was reversed when the position discrimination was acquired under the influence of scopolamine. However, it should be noted that, in an attempt to strengthen position memories, the protocol utilised

involved a degree of overtraining, that is, marmosets were offered 10 more trials upon reaching criterion. These workers concluded from other studies (36) that the retention of an object discrimination was a poor measure of memory in the protocol utilised, although marmosets found it significantly easier to reverse from a task acquired under the influence of scopolamine treatment.

The difference in results obtained in the present and previous studies can perhaps most easily be explained by the differences in procedures used. In the study by Ridley et al. (36), animals were required to acquire an object discrimination using novel objects. However, in the present study, to avoid the element of object novelty and the idiosyncratic aversion/preferences that sometimes occurred, the same object pair was used throughout the present study. This change in procedure is likely to influence the relative importance of the brain areas associated with this task (33).

The combined findings of the present and previous (36) studies indicate that scopolamine acts to impair the acquisition of the object-reward association [see also (27)]. The detailed work of Ridley et al. (36) counters arguments that the poor performance of scopolamine-treated marmosets is a product of an inability to perceive the stimulus, motor deficits, or other nonspecific behavioural changes such as sedation.

Pretreatment of marmosets with the muscarinic receptor agonist, arecoline, attenuated the scopolamine-induced impairment. These findings complement those of Ridley et al. (34,37), who demonstrated attenuation by arecoline of the learning impairments in marmosets induced by lesion of the nucleus basalis of Meynert (nBM) and by intracerebroventricular (ICV) administration of hemicholinium-3 (HC-3), presumably via a direct action at the postsynaptic muscarinic receptors. Arecoline has also been reported to reverse the cognitive deficits induced by scopolamine in the rodent (6,14,16) and improve age-related deficits in a delayed-recall task in primates (7). However, this compound has not proved to be particularly effective in the treatment of dementia/Alzheimer's disease (42), although some improvements have been reported (13).

The problems associated with the administration of a nonselective muscarinic agonist, such as arecoline, are illustrated by the effects observed in two of six marmosets tested following treatment with arecoline (0.08 mg/kg) combined with scopolamine. These animals were unable to perform the task and displayed behavioural changes associated with agitation that were similar to those observed subsequent to the administration of the highest dose of scopolamine. The ability of this dose of arecoline to exacerbate the scopolamine effects may be via an action on presynaptic muscarinic autoreceptors, the existence of which has been well documented (40,43). It is proposed that this autoreceptor exerts an inhibitory control on ACh release. Indeed, Wilson and coworkers (45) demonstrated an arecoline-induced reduction of approximately 50–60% in the release of [^{3}H]ACh in the hippocampus of conscious rats utilising in vivo microdialysis. The performance of marmosets remained unaltered following combined arecoline and saline treatment, which reflects the findings of others (6,37).

The administration of ondansetron before scopolamine reversed scopolamine-induced impairment in task acquisition. The greatest effect was obtained at the 0.1-μg/kg dose, which is 10–100 times greater than that required to improve the basal performance of marmosets in an object discrimination reversal learning task (6,19). In a water maze (16) or T-maze reinforced alternation task in the rat and habituation test in the mouse (6), ondansetron was also able to attenuate a deficit in performance induced by scopolamine. While there are species and task variations in doses required to inhibit a scopolamine-induced deficit, there is a consistent action of ondansetron to inhibit cognitive impairment.

The mechanism(s) via which ondansetron is able to prevent a scopolamine-induced deficit in the marmoset remains to be elucidated. However, the findings of Barnes et al. (5) and Bianchi et al. (11) of a 5-HT$_3$ receptor-mediated modulation of ACh release and the ability of ondansetron to attenuate a scopolamine-induced cognitive deficit in the rat, but not one induced by ICV infusion of HC-3 (6,17), provides some evidence for a possible mechanism of action that involves the modulation of the presynaptic cholinergic system. Functional studies have established a role for 5-HT$_3$ receptors in the modulation of other neurotransmitter systems (12,15,22,26). Furthermore, 5-HT$_3$ receptors are reported to mediate an inhibition of cell firing in the prefrontal cortex (3), an area believed to be essential for "working memory" (25). The cognitive-enhancing properties of the 5-HT$_3$ receptor antagonists revealed in this and previous studies may not simply result from an action on any one neurotransmitter system.

The location of 5-HT$_3$ receptors has recently been determined in the brain of the common marmoset utilising quantitative receptor autoradiography (29). Relatively low levels of [^{3}H]GR65630 binding were found in the amygdala, an area implicated in the acquisition of object-reward associations (21,28), while higher binding levels were detected in the pyramidal cell layer of the hippocampus. In the marmoset, the hippocampus has been associated with "rule learning," that is, learning what is required to obtain a reward (i.e., the rule of the task) rather than building up an object-reward association per se [see (33)]. Studies are under way to determine the relative importance of these two structures and associated brain areas in the cognitive-enhancing properties of agents such as the 5-HT$_3$ receptor antagonists.

In conclusion, this study provides further evidence for a role for 5-HT in the modulation of cognitive processes but, more specifically, a potential role for the 5-HT$_3$ receptor antagonist, ondansetron, in the treatment of cognitive deficits.

REFERENCES

1. Altman, H.; Normile, H. J.; Gershon, S. Non-cholinergic pharmacology in human cognitive disorders. In: Stahl, S. M.; Iversen, S. D.; Goodman, E. C., eds. Cognitive neurochemistry. Oxford: Oxford Science Publications; 1987:346–371.
2. Araujo, D. M.; Lapchak, P. A.; Robitaille, Y.; Gauthier, S.; Quirion, R. Differential alteration of various cholinergic markers in subcortical regions of human brain in Alzheimer's disease. J. Neurochem. 50:1914–1923; 1988.
3. Ashby, C. R.; Edwards, E.; Harkins, K. L.; Wang, R. Y. Characterisation of 5-hydroxytryptamine$_3$ receptors in the medial prefrontal cortex: A microiontophoretic study. Eur. J. Pharmacol. 173:193–196; 1989.
4. Baker, G. B.; Reynolds, G. P. Biogenic amines and their metabolites in Alzheimer's disease: Noradrenaline, 5-hydroxytryptamine and 5-hydroxyindole-3-acetic acid depleted in hippocampus but not substantia innominata. Neurosci. Lett. 100:335–339; 1989.

5. Barnes, J. M.; Barnes, N. M.; Costall, B.; Naylor, R. J.; Tyers, M. B. 5-HT_3 receptors mediate inhibition of acetylcholine release in cortical tissue. Nature 338:762–763; 1989.
6. Barnes, J. M.; Costall, B.; Coughlan, J.; Domeney, A. M.; Gerrard, P. A.; Kelly, M. E.; Naylor, R. J.; Onaivi, E. S.; Tomkins, D. M.; Tyers, M. B. The effects of ondansetron, a 5-HT_3 receptor antagonist on cognition in rodents and primates. Pharmacol. Biochem. Behav. 35:955–962; 1990.
7. Bartus, R. T.; Dean, R. L.; Beer, B. Memory deficits in aged cebus monkeys and facilitation with central cholinomimetics. Neurobiol. Aging 1:145–152; 1980.
8. Bartus, R. T.; Dean, R. L.; Beer, B.; Lippa, A. S. The cholinergic hypothesis of geriatric memory dysfunction. Science 217:408–417; 1982.
9. Bartus, R. T.; Dean, R. L.; Flicker, C. Cholinergic psychopharmacology: An integration of human and animal research on memory. In: Meltzer, H. Y., ed. Psychopharmacology: The third generation of progress. New York: Raven Press; 1987:219–232.
10. Bianchi, C.; Siniscalchi, A.; Beani, L. Effect of 5-hydroxytryptamine on [^{3}H]acetylcholine release from guinea-pig striatal slices. Br. J. Pharmacol. 97:213–221; 1989.
11. Bianchi, C.; Siniscalchi, A.; Beani, L. 5-HT_{1A} agonists increase and 5-HT_3 agonists decrease acetylcholine efflux from the cerebral cortex of freely moving guinea-pigs. Br. J. Pharmacol. 101: 448–452; 1990.
12. Blandina, P.; Goldfarb, J., Green, J. P. Activation of a 5-HT_3 receptor release dopamine from rat striatal slices. Eur. J. Pharmacol. 155:349–350; 1988.
13. Christie, J. E.; Shering, A.; Ferguson, J.; Glen, A. I. M. Physostigmine and arecoline: Effects of intravenous infusions in Alzheimer presenile dementia. Br. J. Psychiatry 138:46–50; 1981.
14. Costall, B.; Coughlan, J.; Naylor, R. J.; Tyers, M. B. Attenuation of scopolamine-induced deficits in a T-maze reinforced alternation task by 5-HT_3 receptor antagonists, cholinomimetics and known nootropic agents. J. Psychopharmacol. 3(4):51P; 1989.
15. Costall, B.; Domeney, A. M.; Naylor, R. J.; Tyers, M. B. Effect of the 5-HT_3 receptor antagonist, GR38032F, on raised, dopaminergic activity in the mesolimbic system of the rat and marmoset brain. Br. J. Pharmacol. 92:881–894; 1987.
16. Coughlan, J.; Costall, B.; Kelly, M. E.; Naylor, R. J.; Tyers, M. B. Ondansetron attenuates the scopolamine deficit in a water maze task. Br. J. Pharmacol. 101:562P; 1990.
17. Coughlan, J.; Costall, B.; Kelly, M. E.; Naylor, R. J.; Tyers, M. B. Ondansetron does not affect hemicholinium-3 induced disruptions in a T-maze reinforced alternation task. Br. J. Pharmacol. 100:561P; 1990.
18. Davis, S. A controlled trial of THA in Alzheimer's disease. J. Psychopharmacol. 3(4):45P; 1989.
19. Domeney, A. M.; Costall, B.; Gerrard, P. A.; Jones, D. N. C.; Naylor, R. J.; Tyers, M. B. The effect of ondansetron on cognitive performance in the marmoset. Pharmacol. Biochem. Behav. 38:169–175; 1991.
20. Drachman, D. A.; Leavitt, J. Human memory and cholinergic system: A relationship to aging? Arch. Neurol. 30:113–121; 1974.
21. Gaffan, D.; Harrison, S. Amygdalectomy and disconnection in visual learning for auditory secondary reinforcement by monkeys. J. Neurosci. 7:2285–2292; 1987.
22. Galzin, A. M.; Poncet, V.; Langer, S. Z. 5-HT_3 receptor agonists enhance the electrically-evoked release of [^{3}H]5HT in guinea-pig frontal cortex slices. Br. J. Pharmacol. 100:307P; 1990.
23. Gellerman, C. W. Chance orders of alternating stimuli in visual discrimination experiments. J. Gen. Psychol. 42:206–208; 1933.
24. Gillet, G.; Ammor, S.; Tullion, G. Serotonin inhibits acetylcholine release from rat striatum slices: Evidence for a presynaptic receptor-mediated effect. J. Neurochem. 42:1687–1691; 1985.
25. Goldman-Rakic, P. S.; Fundahashi, S.; Friedman, H.; Sawaguchi, T. Distributed circuits and distributed functions in primate cerebral cortex. Eur. J. Neurosci. 2(Suppl. 3):1; 1990.
26. Hagan, R. M.; Butler, A.; Hill, J. M.; Jordan, C. C.; Ireland, S. J.; Tyers, M. B. Effect of the 5-HT_3 receptor antagonist GR38032F on responses to injection of a neurokinin agonist into the ventral tegmental areas of the rat brain. Eur. J. Pharmacol. 138:303–305; 1987.
27. Izquierdo, I. Mechanism of action of scopolamine as an amnestic. Trend Pharmacol. Sci. 10:175–177; 1989.
28. Jones, B.; Mishkin, M. Limbic lesions and the problem of stimulus reinforcement associations. Exp. Neurol. 36:362–377; 1972.
29. Jones, D. N. C. Actions of psychotropic drugs in the common marmoset (*Callithrix jacchus*). Ph.D. thesis, University of Bradford, Bradford, U.K.; 1990.
30. Moos, W. H.; Davis, R. E.; Schwarz, R. D.; Gamzu, E. R. Cognitive activators. Med. Res. Rev. 8(3):353–391; 1988.
31. Palmer, A. M.; Francis, P. T.; Benton, J. S.; Sims, N. R.; Marvin, D. M. A.; Neary, D.; Snowden, J. S.; Bowen, D. M. Presynaptic serotonergic dysfunction in patients with Alzheimer's disease. J. Neurochem. 48:8–15; 1987.
32. Perry, E. K. Cortical neurotransmitter chemistry in Alzheimer's disease. In: Meltzer, H. Y., ed. Psychopharmacology: The third generation of progress. New York: Raven Press; 1987:887–895.
33. Ridley, R. M.; Aitkin, D. M.; Baker, H. F. Learning about rules but not about reward is impaired following lesions of the cholinergic projection to the hippocampus. Brain Res. 502:306–318; 1989.
34. Ridley, R. M.; Baker, H. F.; Drewett, B. Effect of arecoline and pilocarpine on learning ability in marmosets pretreated with hemicholinium-3. Psychopharmacology (Berl.) 91:512–514; 1987.
35. Ridley, R. M.; Barratt, N. G.; Baker, H. F. Cholinergic learning deficits in the marmoset produced by scopolamine and ICV hemicholinium. Psychopharmacology (Berl.) 83:340–345; 1984.
36. Ridley, R. M.; Bowes, P. M.; Baker, H. F.; Crow, T. J. An involvement of acetylcholine in object discrimination learning and memory in the marmoset. Neuropsychologia 22(3):253–263; 1984.
37. Ridley, R. M.; Murrary, T. K.; Johnson, J. A.; Baker, H. F. Learning impairment following lesion of the basal nucleus of Meynert in the marmoset: Modification by cholinergic drugs. Brain Res. 376:108–116; 1986.
38. Robinson, S. E. Effect of specific serotonergic lesions of cholinergic neurons in the hippocampus, cortex and striatum. Life Sci. 32:345–353; 1983.
39. Rossor, M.; Iversen, L. L. Non-cholinergic neurotransmitter abnormalities in Alzheimer's disease. Br. Med. Bull. 42(1):70–74; 1986.
40. Sethy, V. H.; Hyslop, D. K. Effect of irreversible loss of muscarinic receptors on [^{3}H]acetylcholine release from the hippocampus. Neuropharmacology 29(2):185–188; 1990.
41. Summers, W. K.; Tachiki, K. H.; King, A. Tacrine in the treatment of Alzheimer's disease: A clinical update and recent pharmacologic studies. Eur. Neurol. 29(Suppl. 3):28–32; 1989.
42. Sunderland, T.; Tanot, P. N.; Newhouse, P. A. Differential responsibility of mood, behaviour and cognition to cholinergic agents in elderly neuropsychiatric populations. Brain Res. Rev. 13:371–389; 1988.
43. Szerb, J. C.; Hadhazy, P.; Dudar, J. D. Release of ^{3}H-acetylcholine from rat hippocampus slices: Effects of septal lesions and of graded concentrations of muscarinic agonists and antagonists. Brain Res. 128:285–291; 1987.
44. Vogels, O. J. M.; Broere, C. A. J.; Ter Laak, H. J.; Ten Donkelaar, H. J.; Nieuwenhys, R.; Schulte, B. P. M. Cell loss and shrinkage in the nucleus basalis Meynert complex in Alzheimer's disease. Neurobiol. Aging 11:3–13; 1989.
45. Wilson, J. M.; Barnes, N. M.; Costall, B. Methodological development and pharmacological characterisation of 'in vivo' [^{3}H]acetylcholine release from rat hippocampus. J. Psychopharmacol. 4:267; 1990.