Cognitive Functions Affected by Scopolamine in Alzheimer’s Disease and Normal Aging

F. Jacob Huff, Susan F. Mickel, Suzanne Corkin, and John H. Growdon

Department of Brain and Cognitive Sciences and Clinical Research Center, Massachusetts Institute of Technology (F.J.H., S.F.M., S.C.), and Department of Neurology, Harvard Medical School (F.J.H., S.F.M., J.H.G.), Cambridge; Departments of Psychiatry and Neurology, University of Pittsburgh (F.J.H.)

ABSTRACT


We gave scopolamine to patients with Alzheimer’s disease (AD) and age-matched control subjects in order to identify which cognitive functions are affected by blocking muscarinic receptors for acetylcholine (ACh). Both subject groups showed dose-related impairments in verbal learning, and patients with AD exhibited enhanced vulnerability to effects of scopolamine on attentional vigilance. In the same dose range, scopolamine did not alter retrieval from long-term lexical-semantic memory or performance on a test of visual discrimination, suggesting that cholinergic neurotransmission is not critical for these cognitive functions. The importance of cholinergic transmission in learning and attention is confirmed by this study, and the results indicate that both these abilities should be measured in investigations of potential cholinergic treatments for AD.

Key words: learning, memory, language, attention, acetylcholine

INTRODUCTION

Pharmacological studies using the muscarinic cholinergic antagonist scopolamine indicate that cholinergic neurotransmission is involved in human memory processes [Safer and Allen, 1971; Drachman and Leavitt, 1974]. High doses of scopolamine also impair attentional vigilance in normal subjects [Safer and Allen, 1971; Traub et al., 1987], but whether other...
cognitive functions are mediated cholinergically has not been definitely established. For example, although Drachman and Leavitt [Drachman and Leavitt, 1974] observed decrements on a verbal fluency test of retrieval from semantic categories, they observed that subjects tended to shift categories during the test, suggesting that the decrement in retrieval may have resulted from impairment of attention by scopolamine.

A deficiency of brain acetylcholine (ACh) has been found in Alzheimer's disease (AD), but postsynaptic muscarinic receptors are preserved [Davies and Verth, 1978; Mash et al., 1985]. It has been suggested that cholinergic deficiency may account for some of the cognitive impairments in AD [Corkin, 1981; Bartus et al., 1982]. This hypothesis implies that patients with AD would be more sensitive than healthy, nondemented subjects to an acute challenge with a cholinergic antagonist. In order to test this prediction, we administered several doses of scopolamine and a placebo to AD patients and control subjects, and measured drug effects on cognitive test performance, including measures of learning and recent memory, attention, lexical-semantic language, and visuospatial abilities. It was postulated that cognitive processes dependent upon ACh would be impaired by scopolamine in a dose-dependent fashion in both groups, and that the dose-response effect would be more pronounced in the AD group.

METHODS

The subjects included ten patients with a clinical diagnosis of AD [McKhann et al., 1984; Khachaturian, 1985] and six age-matched healthy control subjects. All subjects gave informed consent before participating in the study. The AD and control groups were closely matched for age, education, and body weight (Table 1). The mean body weight did not differ significantly between the groups ($t = 0.62, P = .55$), even though the proportion of men and women was not the same in both groups. Two patients were receiving treatment for mild hypertension, one with hydrochlorothiazide and a potassium supplement, the other with labetalol. One patient and one control subject used topical corticosteroids, in a skin cream and ophthalmologic solution, respectively. Two patients used aspirin or acetaminophen, and one took a multiple vitamin preparation. No psychoactive drugs other than scopolamine were given during the study. AD cases with dementia of mild to moderate severity were selected; their mean score (sum of all subscales) on the Blessed Dementia Scale [Blessed et al., 1968] was 16.2.

On successive days, subjects were given intramuscular injections of saline and subsequently higher doses of scopolamine, as tolerated, from 0.1 mg up to 0.5 mg. The gradually increasing fixed dosage schedule was used to minimize uncomfortable or dangerous side effects of scopolamine. This procedure was chosen out of concern for subjects' safety, with knowledge that any practice effects that occurred on neuropsychological testing would be confounded with increasing dose of scopolamine. Because previous research has consistently

<table>
<thead>
<tr>
<th>TABLE 1. Description of Control and AD Subjects</th>
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<tbody>
<tr>
<td>Control Group (N = 6)</td>
</tr>
<tr>
<td>Age</td>
</tr>
<tr>
<td>Mean (yr)</td>
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<tr>
<td>Range</td>
</tr>
<tr>
<td>Education (mean No. of yr)</td>
</tr>
<tr>
<td>Sex (males/females)</td>
</tr>
<tr>
<td>Mean weight (kg)</td>
</tr>
<tr>
<td>Males</td>
</tr>
<tr>
<td>Females</td>
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<tr>
<td>Total</td>
</tr>
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shown only decremental effects of scopolamine on cognition, however, any improvement observed across test sessions in the present study was assumed to result from practice effects and not drug effects. The peripherally active cholinergic antagonist methscopolamine was given (0.5 mg p.o.) with the saline injection, and placebo pills were given with scopolamine injections, in order to keep subjects blind to the scopolamine dosage schedule.

Testing of cognitive function began a half-hour after the scopolamine injection, and lasted about 90 min. The following tests were selected to measure cognitive processes that are commonly impaired in AD. The tests were given in the same sequence to each subject and in each drug condition.

**Word-List Learning**

In this variant of a standard auditory verbal learning test [Rey, 1964], subjects were given six learning trials with a list of 10 unrelated words. The total number of correct responses across trials (0–60) was scored for analysis.

**Continuous Performance Test [Rosvold et al., 1956]**

This test measured sustained attention. Subjects were instructed to press a key in response to each occurrence of a specified letter in a random series of letters. Letters were presented at the rate of one per second on a video screen attached to a microcomputer. Six blocks of 70 letters were presented, each block containing seven instances of the target letter. With rest breaks between blocks, the test lasted about 10 min. The total number of target letters correctly detected (0–42) was scored.

**Category Fluency [Newcombe, 1969; Huff et al., 1986a]**

This test measured retrieval from semantic memory. Subjects were given 1 min to list members of each of four semantic categories (vegetables, vehicles, tools, and clothing). The score was the total number of correct responses across all categories.

**Naming to Definition**

This test also measured lexical-semantic retrieval, but placed less demand upon attentional processes than did Category Fluency. Subjects completed definitions, for example, “A tool used to pound nails is called a . . .” The number of correct responses (0–24) was scored.

**Visual Form Discrimination [Huff et al., 1986a]**

Subjects indicated whether pairs of 12-sided polygons were identical or different. The total number of correct responses (0–24) was scored.

### RESULTS

All subjects received 0.1-mg and 0.2-mg doses of scopolamine. The symptoms reported at those doses were thirst (n = 6), lightheadedness or vertigo (n = 4), gait ataxia (n = 2), headache, nausea, dysarthria, and drowsiness (all n = 1). All subjects except one control subject received the 0.3-mg dose, but several were unable to complete testing because of side effects. Additional side effects not observed at lower doses were a drop in heart rate and blood pressure (n = 2) and visual hallucinations and blurred vision (both n = 1). Two control and six AD subjects received 0.4 mg. One AD subject developed delirium and urinary retention after testing had been completed; thirst, vertigo, and ataxia were observed in most subjects. Only one subject proceeded to the 0.5-mg dose level.

Test results (Table 2) were analyzed using repeated measures analysis of variance, examining main effects of subject group and scopolamine dose (0, 0.1, and 0.2 mg), and the interaction between them. A main effect (P < .05) of subject group (the AD group being impaired in relation to the control group) was evident for all tests except the Continuous
TABLE 2. Test Performance in AD and Control groups

<table>
<thead>
<tr>
<th>Test</th>
<th>Scopolamine dose (mg)</th>
<th>Control</th>
<th>AD</th>
<th>Results of ANOVAa</th>
</tr>
</thead>
<tbody>
<tr>
<td>Word list learning</td>
<td>0.0</td>
<td>43.0 (6.0)</td>
<td>21.5 (6.9)</td>
<td>g**, d*</td>
</tr>
<tr>
<td></td>
<td>0.1</td>
<td>39.0 (8.4)</td>
<td>21.4 (6.1)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.2</td>
<td>37.3 (3.9)</td>
<td>17.1 (7.2)</td>
<td></td>
</tr>
<tr>
<td>Continuous performance</td>
<td>0.0</td>
<td>41.4 (1.3)</td>
<td>35.2 (7.3)</td>
<td>g+, d+, i*</td>
</tr>
<tr>
<td></td>
<td>0.1</td>
<td>41.8 (0.4)</td>
<td>35.4 (8.6)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.2</td>
<td>41.8 (0.4)</td>
<td>31.6 (10.5)</td>
<td></td>
</tr>
<tr>
<td>Category fluency</td>
<td>0.0</td>
<td>50.0 (6.5)</td>
<td>25.5 (14.0)</td>
<td>g**, d**</td>
</tr>
<tr>
<td></td>
<td>0.1</td>
<td>54.8 (6.9)</td>
<td>26.3 (14.3)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.2</td>
<td>57.5 (8.2)</td>
<td>28.2 (13.6)</td>
<td></td>
</tr>
<tr>
<td>Naming to definition</td>
<td>0.0</td>
<td>21.3 (2.0)</td>
<td>16.8 (3.7)</td>
<td>g**, d**</td>
</tr>
<tr>
<td></td>
<td>0.1</td>
<td>22.5 (1.0)</td>
<td>16.2 (4.4)</td>
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<tr>
<td></td>
<td>0.2</td>
<td>23.2 (0.7)</td>
<td>17.7 (3.8)</td>
<td></td>
</tr>
<tr>
<td>Form discrimination</td>
<td>0.0</td>
<td>22.4 (0.9)</td>
<td>20.6 (3.2)</td>
<td>g*</td>
</tr>
<tr>
<td></td>
<td>0.1</td>
<td>23.0 (0.7)</td>
<td>20.7 (1.9)</td>
<td></td>
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<tr>
<td></td>
<td>0.2</td>
<td>22.7 (1.6)</td>
<td>21.3 (1.6)</td>
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</tbody>
</table>

aGroup effect: g+ = P < .10; g* = P < .05; g** = P < .01. Dose effect: d+ = P < .10; d* = P < .05; d** = P < .01. Group x dose interaction: i* = P < .05.

Performance Test, for which a trend (P < .10) toward a group effect was observed. A significant (P < .05) negative dose effect was observed only for the Word-list Learning Test, both groups showing worse performance at higher doses. A trend (P < .10) toward a negative dose effect was observed for the Continuous Performance Test. Positive dose effects were observed for the Category Fluency and Naming to Definition Tests, and probably represented practice effects owing to repeated exposure to the same test materials. The group by dose interaction was significant (P < .05) for the Continuous Performance Test, but nonsignificant for all other tests.

For the tests on which a negative dose effect or group by dose interaction was observed, post hoc comparisons between scores at baseline and the 0.2-mg dose were made in each subject group independently using paired t-tests. These comparisons revealed dose effects in both the control (t = 2.49, P = .05) and AD (t = 2.64, P = .03) groups for Word-list Learning, and a dose effect in the AD (t = 3.02, P = .02) but not the control (t = 1.00, P = .37) group for the Continuous Performance Test.

Performance at the 0.4-mg dose level was examined in the two control and six AD subjects who received that dose. The group by dose interaction on the Continuous Performance Test that was demonstrated at lower doses (Table 2) was also evident after the 0.4-mg dose, a further decline occurring in AD patients’ scores (mean 26.0, SD 16.6), but not in those of the control subjects (mean 41.5, SD 0.7). A decrement (compare with Table 2) in Category Fluency performance was observed after the 0.4-mg dose in the AD group (mean 21.2, SD 10.2), but not the control group (mean 58.0, SD 5.7). No decrement was found in performance on the Naming to Definition Test for either the AD (mean 17.7, SD 4.4) or control (mean 23.0, SD 1.4) group.

DISCUSSION

The results of this investigation confirm previous studies indicating the importance of cholinergic neurotransmission in learning, memory, and attentional processes. The failure to find dose-dependent decrements in lexical retrieval and form discrimination with scopolamine
implies that these other processes are less dependent upon cholinergic transmission. It is possible, however, that these negative findings reflect insensitivity of the tests used to measure the cognitive processes in question. Control subjects performed close to the maximal score on some tests, possibly reducing the sensitivity of the tests to drug effects in those subjects. The fact that group differences were detected with the tests nevertheless indicates that their sensitivity was sufficient to detect drug effects comparable in magnitude to the symptoms of AD.

No deterioration in retrieval from long-term lexical-semantic memory was observed in this study at doses of scopolamine that interfered significantly with Word-list Learning. This result suggests that retrieval from semantic memory is less dependent upon cholinergic transmission than is registration of new information into episodic memory. Similar results have been reported by others [Davis et al., 1983; Beatty et al., 1986]. Higher doses of scopolamine may be required to disrupt performance on fluency tests and other measures of retrieval from semantic memory. Several studies of healthy subjects have shown this effect [Drachman and Leavitt, 1974; Caine et al., 1981], and Sunderland et al. [1985] reported that AD patients receiving 0.25 mg scopolamine showed such disruption, whereas control subjects did not. Similarly, among subjects in our study who received 0.4 mg of scopolamine, Category Fluency scores deteriorated in AD patients but not in control subjects, whereas Naming to Definition scores did not deteriorate in either group.

Fluency tests place demands upon attention and ability to suppress habitual responses [Perret, 1974] as well as upon semantic memory, whereas Naming to Definition is a more specific test of retrieval from lexical-semantic memory. Both fluency and attention were more sensitive to disruption by scopolamine in AD patients than control subjects in the present study, and our results are therefore consistent with the hypothesis that decrements on fluency tests with scopolamine are due to impaired attention. The conclusion that retrieval from semantic memory is unaffected by scopolamine must be viewed cautiously, however, because the practice effect on Naming to Definition may have masked a small drug effect. With the knowledge from the present study that scopolamine had no effect on visual discrimination, it will be possible in future investigations to measure lexical-semantic retrieval with tests of visual object naming, for which multiple equivalent forms are available that could be used to minimize practice effects [Huff et al., 1986b]. Alternatively, because the present study suggests that scopolamine at doses of 0.2 mg or lower can be administered safely to AD patients and elderly control subjects, practice effects may be controlled statistically by counterbalancing the sequence of drug doses and placebo across subjects.

The group by dose interaction observed on the Continuous Performance Test indicates that attentional vigilance is more vulnerable to muscarinic blockade in patients with AD than in healthy individuals. This interaction effect suggests the possibility that cholinergic agonists may produce greater enhancement of attentional processes in patients with AD than in control subjects. The enhanced sensitivity of attentional performance to scopolamine in AD may be mediated by cholinergic neurons in the basal forebrain projecting directly to neocortex, or by pedunculopontine neurons projecting to intralaminar neurons in the thalamus, which project to neocortex [Mesulam, 1985]. Decreased cholinergic activity in neocortex has been documented in AD [Bowen et al., 1976; Davies and Maloney, 1976; Perry et al., 1977], and has been attributed to a loss of neurons in the forebrain nucleus basalis [Whitehouse et al., 1982]. Loss of cholinergic neurons is more pronounced in the basal forebrain than in the pedunculopontine [Zweig et al., 1987] and thalamic regions [Davies, 1979; Rossor et al., 1982], suggesting that the nucleus basalis lesion is more likely to account for the attentional deficit produced by cholinergic blockade.

The effects of scopolamine on learning and recent memory are also probably mediated by cholinergic neurons in the ventral forebrain, particularly those arising from the medial septal nuclei or the diagonal band of Broca and projecting to the hippocampus [Mesulam et al., 1983; Van Hoesen, 1985], although nucleus basalis neurons projecting to the neocortex
may also be involved [Kesner, 1985]. The fact that no dose by group interaction was found for Word-list Learning indicates that patients with AD and healthy control subjects have similar dose-response functions, and implies that they may show similar improvements in verbal learning with cholinergic agonists. The absence of a dose by group interaction suggests, however, that patients with AD will not have an enhanced response to such agents relative to healthy subjects. Studies of drugs that increase cholinergic neurotransmission have demonstrated improved learning in some AD patients, but the gains have generally been modest [Peters and Levin, 1979; Mohs et al., 1985; Thal et al., 1983; Summers et al., 1986].

The fact that a large group difference was found on Word-list Learning, whereas the dose-response function was no greater in the AD than in the control group, suggests that a substantial component of the memory deficit in AD is not cholinergically mediated. Deficiencies in other neurotransmitters, in particular glutamate [Hyman et al., 1986; Maragos et al., 1987] and vasopressin [Mazurek et al., 1986] have been associated with the hippocampal pathology of AD, and are probably in part responsible for the memory impairment. Reductions in other neuropeptide [Beal and Martin, 1986] or monoamine [Rossor et al., 1984; Francis et al., 1985; Summers et al., 1986] neurotransmitters may also contribute to the memory deficit in AD.

Because attention as well as verbal learning was impaired by scopolamine in AD patients in our study, we cannot exclude the possibility that the attentional impairment caused the learning impairment in those subjects. However, the fact that control subjects showed similar learning impairments without attentional impairment at the same doses of scopolamine argues against that explanation. Drachman [1977] and Caine et al. [1981] also report evidence in healthy subjects that the effect of scopolamine on memory is independent of its attentional effects.

Our results raise the possibility that cholinergic agonists may improve attentional processes in AD, and indicate that attention as well as learning and memory should be measured in studies of cholinergic drugs proposed as treatments of AD. Simultaneous improvements in attention and learning abilities in patients with AD would be expected to result in better performance in activities of daily living, and thus would be clinically important. Although the cholinergic deficit is not the only neurotransmitter abnormality in AD, an effective cholinergic therapy may prove to be an important palliative treatment for patients suffering with that disease.

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