Advances in Pharmacology and Therapeutics Journal
Shahid Sadoughi University of Medical Sciences, Yazd, Iran

SHAHID SADOUGHI UNIVERSITY OF MEDICAL SCIENCES

Original Article

Scopolamine Induced Non-cognitive Effects in the Radial-arm maze: Alleviation by Physostigmine

Running Title: Non-cognitive Effects of Scopolamine

Gholamreza Poorheidari[1,2], Mahdi Mashhadi Akbar Boojar[1,2*]

[1]Department of Pharmacology and Toxicology, Faculty of Pharmacy, Baqiyatallah University of Medical Sciences, Tehran, Iran.
[2]Chemical Injuries Research Center, Baqiyatallah University of Medical Sciences, Tehran, Iran.

ARTICLEINFO

Received: 11/22/2022
Accepted: 01/16/2023

Corresponding author
Department of Pharmacology and Toxicology, Faculty of Pharmacy, Baqiyatallah University of Medical Sciences, Tehran, Iran.
Tel: +98- 9124401322
E-mail
mahdimashhadi@yahoo.com

Abstract

Purpose: Scopolamine has frequently been reported to induce a "memory deficit" in animals and humans. However, the possible role of the non-cognitive effects of the drug in these impairments is often ignored. In the present study, the effects of scopolamine on various behaviors in the radial arm maze were recorded and the ability of physostigmine to reverse them was examined.

Methods: Male Long-Evans hooded rats were trained on an 8-arm radial maze to consume drops of 0.1 ml of sweetened milk from the end of each arm. Once the asymptomatic performance was achieved, the effects of scopolamine (0.25 mg/kg i.p. 20 min before testing) on(1) the number of errors (re-entries into the arms), (2) the number of rewards drops not consumed, and (3) agitation were examined.

Results: The number of errors, the number of drops left, and agitation scores were increased significantly compared to saline-treated rats. Concomitant administration of scopolamine and physostigmine (0.25 i.p. 15 min before testing) significantly reduced the agitation scores and revealed a trend toward a decrease in the number of drops left compared to scop-treated rats. However, in this experiment, scopolamine did not significantly increase the number of errors compared to the saline-treated rats.

Conclusion: Taken together, scopolamine induced a small but inconsistent increase in the number of errors. In contrast, there were significant effects of scopolamine on agitation and milk consumption in both experiments. These non-cognitive effects of scopolamine (which were attenuated by physostigmine) may indirectly lead to an increase in errors.

Keywords: Memory, Physostigmine, Radial maze, Rats, Scopolamine

Citation: Poorheidari Gh, Mashhadi Akbar Boojar M. Scopolamine Induced Non-cognitive Effects in the Radial-arm maze: Alleviation by Physostigmine. Adv Pharmacol Ther J. 2023;3(1): 9-21.

Introduction

Many pharmacological studies have examined the effect of cholinomimetic drugs and cholinergic receptor antagonists on learning and memory tasks. The most commonly used model is based on the finding that scopolamine, a muscarinic receptor antagonist, produces amnesia in young healthy subjects similar to that in untreated elderly subjects (1). Cholinesterase (ChE) inhibitors may reverse these deficits. Compounds that reverse these scopolamine-induced deficits in experimental animals may be considered potential drugs to treat cognitive impairment (2).

The cholinergic hypothesis of learning and memory has been the basis for many studies. This hypothesis of cognition became even more popular when some scopolamine-induced deficits were reversed by physostigmine, a cholinergic enhancer; and lesions of central cholinergic pathways induced deficits in animals' performance (3).

Thereafter, according to this hypothesis, either pharmacological blockade (using muscarinic antagonists such as scopolamine or nicotinic antagonists like mecamylamine) or lesioning of the anatomical pathways underlying the cholinergic system (such as nucleus basalis which projects to frontal, lateral, and temporal cortex) were used as models of dementia (4). For example, cognitive impairment induced by scopolamine has been extensively used for searching for cognitive enhancers or for studying the cognitive performance in animals and humans (5).

The ability of scopolamine to induce deficits in learning and memory has also been widely employed as a model of amnesia in different tasks, including the radial arm maze (6,7). It was vastly used by David Olton and his colleagues (8). They provided extensive information concerning the neurobehavioral processes involved in radial-maze performance. Several other groups have employed a similar training and testing procedure (8,9,10). They also used scopolamine (in doses ranging from 0.25-1.0 mg/kg, depending on the study) to induce memory deficits in rats.

In addition, many researchers have reported that deficits induced by scopolamine could be reversed by a variety of agents (11). These include cholinergic compounds such as physostigmine and non-cholinergic compounds such as NS-3(CG3703), a thyrotropin-releasing hormone analog (7,12). Researchers have reviewed these studies.

Scopolamine initially appeared to impair specifically working memory and induce similar deficits to those of Alzheimer's disease (13,14). The specific effect of scopolamine on working memory, however, became controversial later on (15). In fact, some reports showed that scopolamine impairs attention (16). Although there is little doubt about the involvement of the cholinergic system in learning and memory (Davies 1985), there is no evidence to suggest that it is solely involved in memory (3). There is some evidence indicating that the cholinergic system is involved in cognitive functions other than memory such as attention and non-cognitive (motor/motivational) functions (16,17,18). Therefore, the exact role of the cholinergic system in learning and memory remains unclear. Hence, we can assume the interaction between the non-

 DOI: https://doi.org/10.18502/aptj.v3i1.12487

cognitive and cognitive effects of scopolamine as a key point in evaluating scopolamine-induced learning and memory impairment.

In this study, we have tried to show that cholinergic system blockade may underlie some non-cognitive behaviors, which could influence its cognitive effects (such as memory impairment). Moreover, we investigated the effects of physostigmine in the alleviation of such effects.

Material and Methods

Subjects

18 male Long-Evans hooded rats, six months old at the start of experiments (320-380 g), were housed in groups of three per cage under a 12-hour light/dark cycle. To be the same, animals were water-deprived with access to water for only two hours immediately after behavioral testing or training, each day. Food was allowed ad libitum.

Apparatus

The protocol employed in the first series of experiments was according to that of, in which all arms were baited and animals were not confined in the central arena between arm entries (8).

The radial maze, elevated 60 cm above the floor, consisted of a central arena, 48 cm in diameter, surrounded by eight equally spaced radial arms. Each arm was 52 cm long and 10 cm wide and contained a food well at the distal end where rewards (0.1 ml of milk containing 5% sugar) were placed. To prevent the falling of rats from the maze, the edges around each arm and the central arena contained a 3cm lip. The maze was surrounded by a variety of extra-maze cues, consisting of posters, shelves, and chairs. The animals were observed via a monitor situated in an adjacent room.

Training procedure

In the first few days of training, drops of reward (0.1 ml sweetened milk) were placed at the beginning, middle, and end (food well) of each arm to familiarize the animals with the maze and the reward. On the sixth day, only the food wells of the arms were baited. Animals received one trial per day; they were placed individually on the central platform and allowed to explore for 8 min or until all the arms had been entered. During each trial, choice accuracy was assessed by measuring the number of errors (any re-entries into the arms, up to a total of 16 arm entries). Animals reached an asymptotic performance (< 0.5 error/rat/day) after 15 days (8).

Experimental design

After reaching asymptotic performance (less than 0.5 error/rat/trial), drug treatment commenced (19). Each experiment (N=8) was conducted using a Latin-Square design such that by completion of the experiment, all animals had been tested under each treatment condition. In the following experiments, the effects of different doses of scopolamine HBr (scopolamine) and scopolamine MeBr on the performance of rats (experiments 1, 2), as well as the effects of scopolamine HBr on some non-cognitive behaviors (agitation and lack of consumption of milk reward drops), were studied (experiment 3). A possible reversal of scopolamine-induced deficits by physostigmine was also examined (experiment 4). Animals were tested twice a week and a drug-free training day was allowed between testing days.

Measurements

In experiments 1 and 2, only the number of errors (re-entries into the arms) was recorded as a measure of working memory (20). In experiments 3 and 4, in addition to the number of errors, the number of left drops and overt behaviors (that will be collectively described as agitation scores) were also recorded. The number of remaining reward drops was simply counted after the completion of the test trial and removal of the animal. An animal was considered to be agitated and scored one if at least 2 out of 3 below symptoms were observed, otherwise, it was scored zero. The three symptoms were hyperreactivity, tremors (which could be felt only by handling), and vocalization in response to handling.

Drugs

Scopolamine hydrobromide (which acts centrally), scopolamine methylbromide (MeBr, mainly peripherally acting salt), and physostigmine hydrochloride were obtained from Sigma Co. All drugs were administered intraperitoneally in a dose volume of 1 ml/kg. Scopolamine HBr and scopolamine MeBr were administered 20 min before testing, whereas physostigmine was administered 15 min prior to testing. All doses of drugs are expressed as the equivalent of the base (9).

Statistical analysis

One-way analysis of variance (ANOVA) was used with drug treatment (as the within-subjects variable) followed by paired t-test. Significant differences were indicated at the 5% level and a Bonferroni's correction was applied for all multiple comparisons, when appropriate.

Results

Lack of dose-response relationship in scopolamine hydrobromide

In this part of the study, the effect of different doses of scopolamine (0.25, 0.5, and 1.0 mg/kg) on the number of errors (re-entries to the arms) was compared with that of the vehicle (saline). Scopolamine treatment (as the within-subjects factor) revealed a significant effect of the drug treatment ($P<0.01$). Subsequently paired t-test with Bonferroni's correction indicated that the groups treated with scopolamine (0.25 and 0.5, but not 1.0 mg/kg) made significantly more errors than the vehicle-treated group **(Figure 1)**. However, there did not appear a dose-response relationship as all the doses of scopolamine produced a similar number of errors.

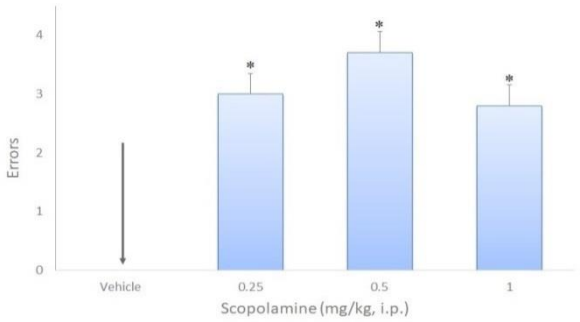

Figure 1. Effect of scopolamine hydrochloride (scopolamine, 0.25-1.0 mg/kg) on the number of errors in the fully baited radial maze.

In this experiment, the control group made no errors (shown by the arrow). *Significantly different from the vehicle-treated control group (Normal saline), $P<0.05$ (After application of Bonferroni's correction the P value was reset to $P<0.05$). Data were expressed as means ± s.e.ms.

In addition, it was noticed that several of the scopolamine-treated animals did not drink some or all of the reward drops despite entering the

 DOI: https://doi.org/10.18502/aptj.v3i1.12487

baited arms and reaching the end of the arms (60-70% of animals). They were also agitated (65-75%) and a few of them also failed not only to complete the task but also to enter a minimum of 8 arms. These animals were excluded from the analysis of the number of errors (3 out of 18 rats). None of these effects was observed in saline-treated animals.

Lack of dose-response relationship of scopolamine methyl bromide

In this portion of the study, the effect of scopolamine MeBr (0.25, 0.5, and 1.0 mg/kg), which passes through the blood-brain barrier very poorly, on the performance of rats was investigated. None of the doses of scopolamine MeBr had a significant effect on the number of errors **(Figure 2)**. One-way analysis of variance (ANOVA) of the number of errors (with drug treatment as the within-subjects factor) revealed no significant effect of the drug treatment ($P<0.5$).

Scopolamine MeBr, unlike scopolamine HBr, did not appear to induce non-cognitive effects such as agitation and inability to consume the reward drops, suggesting that these effects could be due to the central actions of the drug.

During the dose-response relationship experiment for scopolamine HBr, non-cognitive behavioral effects of scopolamine were noticed in 60-70% of the animals (e.g. agitation and not taking the reward drops). Since the effects of different doses of scopolamine on the number of errors were similar, the 0.25 mg/kg dose (the lowest dose) of the drug was used for further experimentation. The effects of scopolamine (0.25 mg/kg) on the number of errors, agitation scores, and the number of reward drops left by the animals were examined.

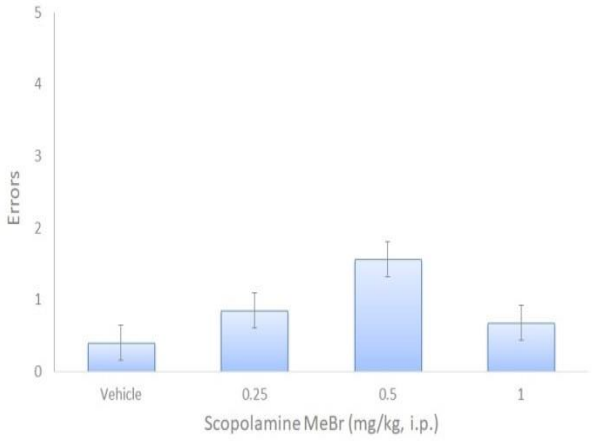

Figure 2. Effect of scopolamine methyl bromide (0.25-1.0 mg/kg) on the number of errors in the fully baited radial maze. Data are expressed as mean±s.e.ms.

Number of errors

Analysis of the number of errors, using a paired t-test revealed that scopolamine significantly increased the number of errors ($P = 0.04$) comparing to the saline control **(Figure 3)**.

Agitation and reward drop consumption

Analysis of the data, using a paired t-test, revealed that scopolamine also significantly increased agitation scores ($P = 0.0001$) and the number of remaining reward drops left ($P<0.001$) compared to the saline control **(Figure 3)**.

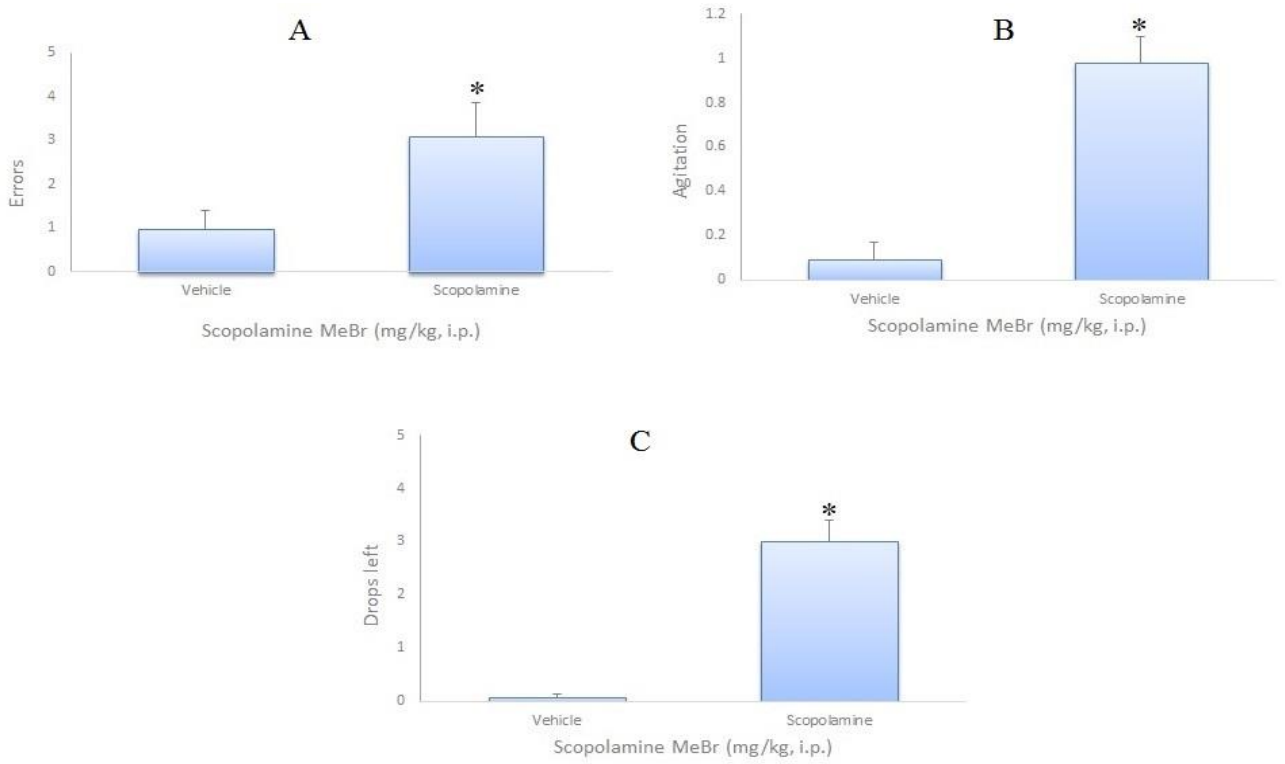

Figure 3. Effects of scopolamine (0.25 mg/kg, i.p.) on the number of errors (A), agitation scores (B), and number of drops (C) in the fully baited radial maze. * Significantly different from the vehicle-treated control group (Normal saline), P<0.05. Data are expressed as means ± s.e.ms

Investigation of the ability of physostigmine to reverse the effects of scopolamine

In this experiment, the ability of physostigmine (0.25 mg/kg), an anticholinesterase agent, to reverse scopolamine effects was investigated. Rats were treated with saline, scopolamine, or scopolamine plus physostigmine. The effects of physostigmine administered alone could not be tested, because of the presence of severe side effects occurring in the absence of scopolamine.

Number of errors

There was no significant difference between the number of errors made by the different treatment groups ($P < 0.5$) **(Figure 4)**.

Agitation and reward drop consumption

Scopolamine significantly increased the agitation scores and decreased the consumption of the reward drops. One-way analysis of variance revealed a significant treatment effect on agitation scores ($P < 0.001$) and the number of the remained drops ($P < 0.01$). Subsequent paired t-tests and application of Bonferroni's correction indicated that scopolamine significantly increased the agitation scores and the number of drops left relative to saline control. Physostigmine, when administered in combination with scopolamine, significantly decreased the agitation scores and tended to increase the number of reward drops left relative to scopolamine alone **(Figure 4)**.

 DOI: https://doi.org/10.18502/aptj.v3i1.12487

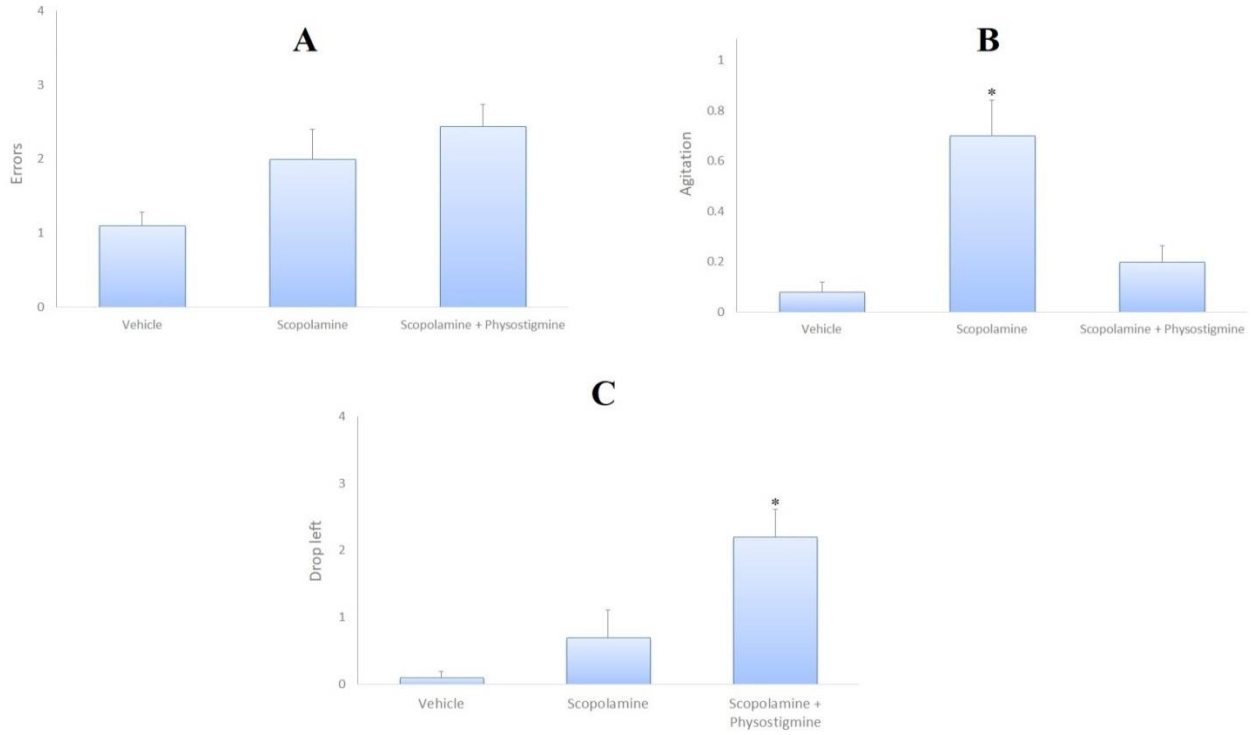

Figure 4. Effects of scopolamine (0.25 mg/kg, i.p.) and combination of scopolamine (0.25 mg/kg, i.p.) with physostigmine (0.25 mg/kg, i.p.) on the number of errors (A), agitation scores (B) and the number of drops (C) in the fully baited radial maze. * Significantly different from vehicle-treated control group (Normal saline) and + significantly different from scopolamine treated group, $P<0.05$. Data are expressed as means ± s.e.ms.

Discussion

Effect on the number of errors: the performance disruptive effects of scopolamine

Different doses of scopolamine (0.25, 0.5, and 1.0 mg/kg) had similar effects on the number of errors (re-entries into the arms). The disruptive effect of scopolamine appeared to be more pronounced the first few times of exposure to scopolamine. But it seemed to be less effective as the animals were more frequently exposed to the drug under similar circumstances. This raised the possibility of behavioral tolerance to the performance disruptive effect of scopolamine (such as adopting a response strategy) and/or pharmacological tolerance. Researchers showed that an uninterrupted radial maze is relatively resistant to pharmacological disruption. These could mainly be due to the resistance of the animals to the effect of the drug by using non-spatial strategies (21). In fact, most of the rats (80-90%) were moving around the maze, clockwise or anti-clockwise, and entering the arms one by one until all baited arms were entered and the task was completed. This sort of response strategy in the radial maze has been called ‘running around in a circle’ (22). Hence, the animals did not need to use the working memory to remember which arms had been entered and which remained to be entered. The only thing they had to remember was the direction, which was almost always the same for each rat. Most of the errors were made by a few of the animals (10-15%) that missed one arm

whilst exploring the maze and moving in one direction, clockwise or anti-clockwise, and making errors (re-entries into the arms) until they reached the missing arm. In other words, a small number of animals made a large number of errors. It seems that they might have learned to cope with the effect of the drug. However, scientists stated that scopolamine-induced deficits were not correlated with the percentage of spatial strategies (23). They also pronounced that, under scopolamine treatment, the performance of rats showing preferences for spatial strategies did not differ significantly from those of rats showing preferences for orientation strategies (24). By looking at the weight of the existing literature, we can assign a deficit to their experiments.

Thus, the ability of rats to adopt response strategies may influence the sensitivity of rats to scopolamine in radial maze performance (25). There is evidence that rats solve their maze problems by choosing either the adjacent or adjacent-but-one arm in a particular direction (26,27) Scientists showed that animals using non-spatial strategies are relatively resistant to the disruptive effects of scopolamine, compared to those using spatial discrimination involving a complex of intra- and extra-maze cues (27). Therefore, it may be essential to confine the rats in the central platform between arm entries to stop them from using a response strategy (28). Investigators used a modified task to reduce the tendency to use non-spatial strategies (7). They concluded that the modified task, which they used in the radial arm maze, is sensitive to the disruptive effects of scopolamine and can identify the cognitive enhancing effects of drugs. These findings show that an uninterrupted radial maze testing procedure is not highly sensitive to pharmacological treatment (29). Furthermore, we can suppose that the abilities of rats to resist muscarinic blockade depend on the strategy they use in the maze and (also) the task design of the maze.

Taken together, the cognitive impairing effect of the drug (increasing the number of errors) was small and inconsistent throughout our experiments. However, this performance disruptive effect was not seen with either of the doses of scopolamine MeBr (0.25, 0.5, and 1.0 mg/kg) suggesting that this effect is mainly due to the central action of the scopolamine. This is in parallel with the observed effects of scopolamine on cognitive tasks such as memory in other studies carried out by some investigators. They also found that scopolamine HBr affects memory more than scopolamine MeBr (24,30,31). Investigators also used methylscopolamine in the radial arm maze and found that it did not affect correct responses (23,32,33). In another research, investigators elucidated that lower doses of methylscopolamine (0.2 mg/kg) had no effect on memory in radial arm maze and they stated that higher doses (0.63 mg/kg) did cause serious peripheral effects which eventually prevented some animals from completing the task (34). Hence, again we suppose that such scopolamine-induced effects are mediated centrally.

Non-cognitive effects of scopolamine

In our study, non-cognitive effects (agitation and not consuming the reward drops) of scopolamine were also observed. For example, scopolamine-treated animals were agitated and it was a

 DOI: https://doi.org/10.18502/aptj.v3i1.12487

consistent effect of the drug, although it was not observed in all animals. Missing the reward drops was another consistent effect of scopolamine. These non-cognitive effects also appeared to be a result of the central actions of scopolamine, as these effects were not observed with scopolamine MeBr.

It is difficult to separate reliably the scopolamine effects on learning and memory processes from its effects on other behavioral domains. For example, methylscopolamine (which does not cross the blood-brain barrier) is as active as scopolamine in several models of cognitive function, indicating that peripheral changes induced by these compounds indirectly influence performance in cognitive tasks (35,36). It is, therefore, very important to distinguish the central versus peripheral effects of anticholinergic agents. Scopolamine-induced impairment of performance may also be mediated by direct effects on sensorimotor function or motivation deficits (36,37). Further, likely, the scopolamine-induced impairment in the performance of both experimental animals and humans in the delayed matching to position task (a commonly used test of cognitive function) is secondary to attentional deficits that are induced by the drug (38,39).

The non-cognitive effects of scopolamine have often been reported, but their possible contribution to the disruptive effect of the drug on cognition has often been ignored. Thus, described similar non-cognitive effects of scopolamine to those observed in the present study (40). Klinkenberg and Blokland found that rats treated with scopolamine were ‘hyperactive, sensitive to touch, had greater muscle tension, and vocalized more frequently than control animals when placed in the operant chambers or handled during injections’. In addition, also reported that scopolamine-treated mice were difficult to capture, vocalized when handled, and showed a high level of escape-like behavior when initially placed in the maze (41). Furthermore, reported that some of the rats treated with scopolamine (0.5-1.0 mg/kg) were either excluded from the experiments or submitted to supplementary sessions due to a lack of exploration (26). Kay and co-workers also noticed on average that scopolamine-treated rats reacted (screaming) more to handling than the rats from the other groups. Scientists also reported that 6 out of 15 rats treated with scopolamine (0.5 mg/kg) failed to complete the task (34). Moreover, stated that behavior on the radial arm maze is often expressed using unrepeated arm entries as the sole measure of maze performance, which can be misleading since the use of a single behavioral measure can obscure other treatment effects (42). They finally concluded that ‘the observed scopolamine-induced performance deficits on the radial maze may result partly from a reduction in locomotion and exploratory behavior, possibly due to non-specific sedative and/or anxiogenic effects of scopolamine, rather than being solely the consequence of learning impairment’.

The existence of non-cognitive effects of scopolamine in animals, similar to those (hyperactivity and agitation) in humans, is suggested by observing symptoms like hyperactivity and vocalization in response to handling in animals treated with scopolamine (43,44,45). Interestingly, the severe side effects of

scopolamine (~ 0.01 mg/kg), which were observed in human subjects, were alleviated by the administration of physostigmine (46). Therefore, as far as such non-cognitive effects of scopolamine are observed in both humans and animals, it may be difficult to justify the use of scopolamine-induced performance impairments as a model of amnesia. However, there is an enormous number of reports in which scopolamine has been used as a model of amnesia (14,18).

This raises the possibility of a lack of dissociation between the cognitive and non-cognitive effects of the drug. Furthermore, whether the cognitive effect of scopolamine is a secondary effect to the non-cognitive primary effects, or, whether it is a primary effect of the drug per se is not clear.

Alleviation of scopolamine-induced effects by physostigmine

The cholinergic hypothesis predicts that drugs potentiating central cholinergic function should improve cognition and perhaps some of the behavioral problems experienced with Alzheimer's disease (2). But, we found that physostigmine plus scopolamine had no significant effect on the number of errors. Also, in our study, physostigmine (when administered in combination with scopolamine) significantly reduced the agitation scores and tended to reduce the number of reward drops left relative to scopolamine alone.

The possible effects of anticholinesterases on the central nervous system and, in particular, on learning and memory, have generated considerable interest (47). In this way, many investigators used physostigmine or its derivatives and also other kinds of cholinesterase inhibitors. Most of them stated that physostigmine and other cholinesterase inhibitors dose-dependently reverse the cognitive impairment induced by scopolamine. Among these multiple studies, some reported a nonlinear effect of physostigmine in the alleviation of such cognitive effects; however, the doses they used are highly different from each other (the doses used in one were 10 times and more of the doses used in the other study) (48,49).

Most studies of cholinergic agents have assessed cognitive responses rather than the neuropsychiatric impact of these agents, but a few observations relating cholinomimetic treatment to changes in behavior have been made. Physostigmine has been extensively studied as a potential therapeutic agent in AD, but limited information is available on its behavioral effects (50,51).

Besides this, some researchers pointed to the effects of physostigmine upon non-cognitive aspects induced by scopolamine. Non-cognitive behavioral changes such as depression, aggressive behavior, psychosis, and overactivity occur frequently in patients with dementia (in addition to cognitive impairment). Clinical trial data indicate that cholinomimetics improve non-cognitive behavior (52). Also, there is evidence that novel ChE inhibitors like metrifonate produce significant improvements in the 3 domains (cognition, behavior, and function) of Alzheimer's disease (53). The neuropsychological impairments of AD are attributed, at least partially, to the cholinergic disturbance, and current approaches to the treatment of the cognitive abnormalities attempt to enhance cholinergic function.

 DOI: https://doi.org/10.18502/aptj.v3i1.12487

Behavioral changes are common in AD and include psychosis, agitation, depression, anxiety, personality alterations, and neurovegetative changes. The contribution of cholinergic deficiency to behavioral alterations has been little explored, but neurochemical, neuroanatomic, pharmacologic, and clinical observations suggest that cholinergic deficiency contributes importantly to the neuropsychiatric dimension of AD (50).

According to what has been described, we can suppose that both the 'cognitive impairing' effect of scopolamine and the non-cognitive effects of scopolamine (the present study) could become attenuated by physostigmine and this could raise again the possibility of "lack of dissociation between cognitive and non-cognitive effects of scopolamine" (7). Finally, evidence is emerging from clinical trials of cholinomimetic drugs that such drugs may improve the abnormal non-cognitive, behavioral symptoms of Alzheimer's disease. Thus, ChE inhibitors have been reported to significantly improve many manifestations of behavioral disturbance including agitation, apathy, hallucinations, and aberrant motor behavior (54,55). This contention is supported by the serendipitous actions of cholinesterase inhibitors in alleviating non-cognitive behavioral symptoms in patients with AD (2,56).

Conclusion

In summary, the lack of consistent 'cognitive' effects of scopolamine (observed in the fully baited radial arm maze) was likely to be due to several factors including response strategy usage by rats. Our data emphasize the importance of cholinergic mechanisms in behavior, the role of the cholinergic deficit in the behavioral changes of the scopolamine model of AD, and the potential behavioral benefit of cholinergic therapy in this experimental model of AD. Clearly, non-cognitive behavioral effects of scopolamine (at doses similar to those, which are used for induction of cognitive impairments) undermine a specific effect on learning and memory. In other words, scopolamine appears to have a general effect on behavioral performance rather than on learning and memory. The contribution of the cholinergic deficiency of AD to the repertoire of behavioral changes characteristic of the disease has been underestimated and warrants further investigation.

Conflict of interests: No

Funding: No

Acknowledgments: The authors would like to offer special thanks to the Baqiyatallah University of Medical Sciences for their support.

Ethical considerations: This study was conducted with the support of the Faculty of Pharmacy, Baqiyatallah University of Medical Sciences (BMSU), and was ethically approved by the ethics committee and supported by the research deputy of BMSU.

Authors' contribution: MMAB contributed to the conception, formal analysis, methodology, project administration, validation, visualization, and writing – review & editing. GhP contributed to data curation, funding acquisition, investigation, supervision and organized the database, and wrote the original draft of the manuscript.

References

1. Baradaran A, Rabiei Z, Rafieian M, Shirzad H. A review study on medicinal plants affecting amnesia through cholinergic system. J Herbmed Pharmacol 2012;1(1):3-9.
2. Tannenbaum C, Paquette A, Hilmer S, Holroyd-Leduc J, Carnahan R. A systematic review of amnestic and non-amnestic mild cognitive impairment induced by anticholinergic, antihistamine, GABAergic and opioid drugs. Drugs Aging 2012;29(8):639-58.
3. Bentley P, Husain M, Dolan RJ. Effects of cholinergic enhancement on visual stimulation, spatial attention, and spatial working memory. Neuron 2004;41(6):969-82.
4. Newman LA, Gold PE. Attenuation in rats of impairments of memory by scopolamine, a muscarinic receptor antagonist, by mecamylamine, a nicotinic receptor antagonist. Psychopharmacology. 2016 Mar 1;233(5):925-32.
5. Wu Q, Cao Y, Liu M, Liu F, Brantner AH, Yang Y, Wei Y, Zhou Y, Wang Z, Ma L, Wang F. Traditional Chinese Medicine Shenmayizhi Decoction Ameliorates Memory And Cognitive Impairment Induced By Scopolamine Via Preventing Hippocampal Cholinergic Dysfunction In Rats. Neuropsychiatr Dis Treat 2019;15:3167.
6. Ionita R, Postu PA, Mihasan M, Gorgan DL, Hancianu M, Cioanca O, Hritcu L. Ameliorative effects of Matricaria chamomilla L. hydroalcoholic extract on scopolamine-induced memory impairment in rats: A behavioral and molecular study. Phytomedicine 2018;47:113-20.
7. Moisescu RE, Ghiță IC, Sorescu A, Coman OA, Coman LA, Fulga I. Cholinergic modulation of memory. Farmacia 2018;66:938-47.
8. Olton DS, Samuelson RJ. Rememberance of place passed: spatial memory in rats. J Exp Psychol 1976;2,97-116.
9. Van Can M, Tran AH, Pham DM, Dinh BQ, Van Le Q, Van Nguyen B, Nguyen MT, Nguyen HX, Nguyen NT, Nishijo H. Willughbeia cochinchinensis prevents scopolamine-induced deficits in memory, spatial learning, and object recognition in rodents. J Ethnopharmacol 2018;214:99-105.
10. Pahwa P, Goel RK. Asparagus adscendens root extract enhances cognition and protects against scopolamine induced amnesia: An in-silico and in-vivo studies. Chem Biol Interact 2016;260:208-18.
11. Lenz RA, Baker JD, Locke C, Rueter LE, Mohler EG, Wesnes K, Abi-Saab W, Saltarelli MD. The scopolamine model as a pharmacodynamic marker in early drug development. Psychopharmacology 2012;220(1):97-107.
12. Ogasawara T, Nakagawa Y, Ukai Y, Tamura M & Kimura K (1995). NS-3(CG3703), a TRH analog, ameliorates scopolamine-induced memory disruption in rats. Pharmacol Biochem Behav 1995;51: 929-934.
13. Sherman SJ, Atri A, Hasselmo ME, Stern CE, Howard MW. Scopolamine impairs human recognition memory: data and modeling. Behav Neurosci 2003;117(3):526.
14. Riedel G, Kang SH, Choi DY, Platt B. Scopolamine-induced deficits in social memory in mice: reversal by donepezil. Behav Brain Res 2009;204(1):217-25.
15. Araujo JA, Chan AD, Winka LL, Seymour PA, Milgram NW. Dose-specific effects of scopolamine on canine cognition: impairment of visuospatial memory, but not visuospatial discrimination. Psychopharmacology 2004;175(1):92-8.
16. Demeter E, Sarter M. Leveraging the cortical cholinergic system to enhance attention. Neuropharmacology 2013;64:294-304.
17. Garcia-Alloza M, Gil-Bea FJ, Diez-Ariza M, Chen CH, Francis PT, Lasheras B, Ramirez MJ. Cholinergic–serotonergic imbalance contributes to cognitive and behavioral symptoms in Alzheimer's disease. Neuropsychologia 2005;43(3):442-9.
18. Bjokland A. Acetylcholine: A neurotransmitter for learning and memory?. Brain ResS Rev 1996;21: 285-300.
19. Ott, RL. An introduction to statistical methods and data analysis. 4th ed. pp: 859, Wadsworth, Inc., Belmont, California 1993.
20. Olton DS & Papas BC.(1979).Spatial memory and hippocampal function. Neuropsychologia 1979;17: 69-682.
21. Buresova O, Bures J. Radial maze as a tool for assessing the effect of drugs on the working memory of rats. Psychopharmacology 1982;77(3):268-71.
22. Tonneau F, Cabrera F, Corujo A. Hamsters'(Mesocricetus auratus) memory in a radial maze analog: The role of spatial versus olfactory cues. J Comp Psychol 2012;126(1):82.
23. van der Staay FJ, Bouger PC. Effects of the cholinesterase inhibitors donepezil and metrifonate on scopolamine-induced impairments in the spatial cone field orientation task in rats. Behav Brain Res 2005;156(1):1-0.
24. Ormerod BK, Beninger RJ. Water maze versus radial maze: differential performance of rats in a spatial delayed match-to-position task and response to scopolamine. Behav Brain Res 2002;128(2):139-52.
25. Fujioka T, Fujioka A, Tan N, Chowdhury GM, Mouri H, Sakata Y, Nakamura S. Mild prenatal stress enhances learning performance in the non-adopted rat offspring. Neuroscience 2001;103(2):301-7.
26. Kay C, Harper DN, Hunt M. Differential effects of MDMA and scopolamine on working versus reference memory in the radial arm maze task. Neurobiol Learn Mem 2010;93(2):151-6.
27. Watts J, Stevens R, Robinson C. Effects of scopolamine on radial maze performance in rats. Physiol Behav 1981;26:845-851.
28. Peltonen I, Jalkanen AJ, Sinervä V, Puttonen KA, Männistö PT. Different effects of scopolamine and inhibition of prolyl oligopeptidase on mnemonic and motility functions of young and 8- to 9- month- old

 DOI: https://doi.org/10.18502/aptj.v3i1.12487

rats in the radial-arm maze. Basic Clin Pharmacol Toxicol 2010;106(4):280-7.
29. Lacroix L, White I, Feldon J. Effect of excitotoxic lesions of rat medial prefrontal cortex on spatial memory. Behav Brain Res 2002;133(1):69-81.
30. Szczodry O, van der Staay FJ, Arndt SS. Modelling Alzheimer-like cognitive deficits in rats using biperiden as putative cognition impairer. Behav Brain Res 2014;274:307-11.
31. de Bruin N, Pouzet B. Beneficial effects of galantamine on performance in the object recognition task in Swiss mice: deficits induced by scopolamine and by prolonging the retention interval. Pharmacol Biochem Behav 2006;85(1):253-60.
32. Abe H, Ishida Y, Iwasaki T. Perirhinal N-methyl-D-aspartate and muscarinic systems participate in object recognition in rats. Neurosci Lett 2004;356(3):191-4.
33. Angelucci ME, Cesario C, Hiroi RH, Rosalen PL, Cunha CD. Effects of caffeine on learning and memory in rats tested in the Morris water maze. Braz J Med Biol Res 2002;35(10):1201-8.
34. Magnani M, Pozzi O, Biagetti R, Banfi S & Dorigotti L (). Oxiracetam antagonises the disruptive effects of scopolamine on memory in the radial maze. Psychopharmacology 1992;106(2):175-178.
35.Klinkenberg I, Blokland A. A comparison of scopolamine and biperiden as a rodent model for cholinergic cognitive impairment. Psychopharmacology 2011;215(3):549-66.
36. Poorheidari G, Stanhope KJ, Pratt JA. Effects of the potassium channel blockers, apamin and 4-aminopyridine, on scopolamine-induced deficits in the delayed matching to position task in rats: a comparison with the cholinesterase inhibitor E2020. Psychopharmacology 1998;135:242-55.
37. Blake MG, Boccia MM, Krawczyk MD, Delorenzi A, Baratti CM. Choline reverses scopolamine-induced memory impairment by improving memory reconsolidation. Neurobiol Learn Mem 2012;98(2):112-21.
38. Ellis JR, Ellis KA, Bartholomeusz CF, Harrison BJ, Wesnes KA, Erskine FF, Vitetta L, Nathan PJ. Muscarinic and nicotinic receptors synergistically modulate working memory and attention in humans. Int J Neuropsychopharmacol 2006;9(2):175-89.
39. Warburton DM, Rusted JM.Cholinergic control of cognitive resources. Neuropsychobiology 1993;28:43-6.
40. Klinkenberg I, Blokland A. The validity of scopolamine as a pharmacological model for cognitive impairment: a review of animal behavioral studies. Neurosci Biobehav Rev 2010;34(8):1307-50.
41. Bazalakova MH, Wright J, Schneble EJ, McDonald MP, Heilman CJ, Levey AI, Blakely RD. Deficits in acetylcholine homeostasis, receptors and behaviors in choline transporter heterozygous mice. Genes Brain Behav 2007;6(5):411-24.
42. Watson CD, Hewitt MJ, Fone KCF, Dickinson SL & Bennett GW. (). Behavioural effects of scopolamine and the TRH analogue RX77368 on radial arm maze performance in the rat. J Psychopharmacol 1994;8:88-93.
43. Araujo JA, Studzinski CM, Milgram NW. Further evidence for the cholinergic hypothesis of aging and dementia from the canine model of aging. Prog Neuropsychopharmacol Biol Psychiatry 2005;29(3):411-22.
44. Poorheidari G, Pratt JA, Dehghani N. Effects of low-dose scopolamine on locomotor activity: No dissociation between cognitive and non- effects. Neurosci Res Commun 2002;31(3):165-74.
45. Blokland A. Scopolamine-induced deficits in cognitive performance: A review of animal studies. Scopolamine Rev 2005;1:1-76.
46. Serby M, Corwin J, Jordan B, Novatt A & Rotrosen J . Side effects of scopolamine administration. Am J Psychiatry 1984;141: 1010-1011.
47. Koelle GB. Cholinesterases and anticholinesterase agents. Springer Science & Business Media; 2013.
48. Braida D. Paladini E. Griffini P. Lamperti M. Maggi A & Sala M.(1996).An inverted U-shaped curve for heptylphysostigmine on radial maze performance in rats: comparison with other cholinesterase inhibitors. Eur J Pharmacol 1996;302(1-3):13-20.
49. Kirrane RM, Mitropoulou V, Nunn M, Silverman J, Siever LJ. Physostigmine and cognition in schizotypal personality disorder. Schizophr Res 2001;48(1):1-5.
50. Cummings JL, Kaufer DI. Neuropsychiatric aspects of Alzheimer's disease: the cholinergic hypothesis revisited. Neurology 1996;47(4):876-883.
51. Mach M, Grubbs RD, Price WA, Paton SJ, Lucot JB. Behavioral changes after acetylcholinesterase inhibition with physostigmine in mice. Pharmacol Biochem Behav 2004;79(3):533-40.
52. Minger SL, Esiri MM, McDonald B, Keene J, Carter J, Hope T, Francis PT. Cholinergic deficits contribute to behavioral disturbance in patients with dementia. Neurology 200055(10):1460-7.
53. Ormrod D, Spencer C. Metrifonate: A Review of its Use in Alzheimer's Disease. CNS Drugs 2000;13(6):443-467.
54. Wynn ZJ, Cummings JL. Cholinesterase inhibitor therapies and neuropsychiatric manifestations of Alzheimer's disease. Dement Geriatr Cogn Disord 2004;17(1-2):100-8.
55. Pinto T, Lanctôt KL, Herrmann N. Revisiting the cholinergic hypothesis of behavioral and psychological symptoms in dementia of the Alzheimer's type. Ageing Res Rev 2011;10(4):404-12.
56. Cummings JL, Black C.(1998). The cholinergic hypothesis of neuropsychiatric symptoms in Alzheimer's disease. Am J Geriatr Psychiatry 6:S64-S78.