Pharmacology Biochemistry and Behavior, Vol. 44, pp. 815-819, 1993
Printed in the U.S.A.

0091-3057/93 $6.00 + .00

Comparison of the Effects of Scopolamine and Methylscopolamine on the Performance of a Fixed-Ratio Discrimination in Squirrel Monkeys

ERIC D. PAKARINEN AND JOSEPH M. MOERSCHBAECHER[1]

Department of Pharmacology, Louisiana State University Medical Center, 1901 Perdido Street, New Orleans, LA 70112-1393

Received 18 May 1992

PAKARINEN, E. D. AND J. M. MOERSCHBAECHER. *Comparison of the effects of scopolamine and methylscopolamine on the performance of a fixed-ratio discrimination in squirrel monkeys.* PHARMACOL BIOCHEM BEHAV **44**(4) 815-819, 1993.—In the presence of a stimulus behind the center key, squirrel monkeys were required to complete one of two fixed ratios (FRs) on the center key (FR 30 or FR 25). Completion of the ratio turned off the center-key stimulus and produced a stimulus behind each of the two side keys. If the completed ratio was high (e.g., FR 30), a response on the left key produced a food pellet. If the ratio was low (e.g., FR 25) a response on the right key produced food. Errors produced a brief timeout. Dose-effect curves for scopolamine (0.001-0.18 mg/kg) and methscopolamine (0.0032-5.6 mg/kg) were determined under a FR 30 vs. FR 25 discrimination, which controlled both moderate levels of accuracy and high rates of responding. Scopolamine produced a dose-related decrease in overall response rates and increase in percent errors. Methscopolamine decreased response rates and increased percent errors in a dose-related manner much like scopolamine. However, scopolamine was found to be about 10 times more potent on a mg/kg basis than methscopolamine. Scopolamine is in general considered to be centrally acting due in part to its lipid solubility. The results from these studies suggest that methscopolamine, in general considered to be peripherally acting, may also cross the squirrel monkey blood-brain barrier at high doses and produce behavioral effects comparable to those of scopolamine.

Fixed-ratio discrimination Performance Scopolamine Methylscopolamine Operant behavior Squirrel monkeys

SCOPOLAMINE is primarily a centrally acting cholinergic antagonist that readily crosses the blood-brain barrier but also blocks muscarinic receptors in the periphery (4). Methscopolamine is primarily a peripherally acting muscarinic antagonist with a quaternary nitrogen that does not readily cross the blood-brain barrier (4). Methscopolamine has been used as a pharmacological control for the peripheral anticholinergic effects of scopolamine. For example, in monkeys responding under a multiple fixed-ratio fixed-interval schedule of food presentation, scopolamine has been reported to produce dose-related decreases in overall response rate (8). In that same study, it was reported that at high doses methscopolamine also decreased response rate, although it was estimated to be at least 10 times less potent than scopolamine in that regard.

Cholinergic mechanisms have been suggested as playing an important role in the acquisition and performance of complex discriminations in nonhuman primates (1,2,3,5,10). These studies have in general been consistent with the notion that the cholinergic system is critical in regulating learning and memory processes. In such studies, it has in general been found that scopolamine disrupts both the rate and accuracy (percent errors) of responding (1-3,10). In only a few such studies, however, have the effects of the peripherally acting agent methscopolamine also been investigated. For example, in one such study scopolamine (0.2 and 0.4 mg/kg, IM) decreased response rate and disrupted accuracy in a delayed matching-to-sample procedure in monkeys, while similar doses of methscopolamine selectively decreased the rate of responding while having little or no effect on accuracy of responding (3). In another study, it was reported that methscopolamine (0.03-0.09 mg/kg, IM) did not disrupt accuracy on a visual discrimination procedure in monkeys while scopolamine (0.0075-0.06 mg/kg, IM) disrupted accuracy in a dose-related manner (5). The results of these studies would suggest

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

that the effects of scopolamine on accuracy of responding are mediated by central mechanisms while response rate may represent a more peripheral action. It should be noted, however, that in each of these studies methscopolamine was tested only within the effective scopolamine dose range and comparable rate-decreasing effects across a range of doses were not obtained. Based upon the findings of Moerschbaecher et al. (8), one might predict that doses at least 10 times higher should be tested. How methscopolamine might affect accuracy of responding at doses that produce comparable rate-decreasing effects remains unknown. The present study was designed to widen this narrow data base and further investigate the relative potency of these compounds in terms of their effects on rate and accuracy of a discriminative performance. Thus, one aim of this study was to make a quantitative comparison of the relative potency of these compounds to separate their peripheral and central actions. The effects of scopolamine and methscopolamine were therefore compared in squirrel monkeys responding under a fixed-ratio discrimination procedure.

METHOD

Subjects

Three adult, female squirrel monkeys (*Saimiri sciureus*) with a history of responding under a fixed-ratio discrimination procedure served as subjects. Each subject was maintained at approximately 85% of its free-feeding body weight (600–670 g) by a diet consisting of banana-flavored food pellets, Purina Monkey Chow, Zu/Preem Marmoset or Primate Diet, fruit, and vitamins. The banana pellets were earned during the experimental session while the remainder of the diet was fed in the home cage after each session. Each subject was individually housed, maintained on a 12 L : 12 D cycle (light on from 7:00 a.m. to 7:00 p.m.), and water was continuously available.

Apparatus

During each session, each monkey was seated in a Plexiglas chair (STC-300, BRS/LVE, Inc., Laurel, MD) with a water bottle mounted on the left side of the chair positioned such that the subject could easily drink from it. The chair was placed in front of a response panel inside in a ventilated, sound-attenuating chamber (MEC-004 BRS/LVE). The response panel was equipped with three recessed keys (Coulbourn Instruments, Model 21-17) that were mounted 5 cm apart center to center. The reinforcer, a 190-mg Noyes banana-flavored pellet (P.J. Noyes Company, Inc., Lancaster, NH), was delivered into an aperture measuring 4.7 × 4.7 cm located 5 cm below the center key. Events were scheduled and recorded by means of a microprocessor, a printer, and a cumulative recorder (Gerbrands, G3100 Model C-4, Arlington, MA).

Procedure

Each subject responded under a fixed-ratio (FR) discrimination procedure (7). Under this procedure, a stimulus (yellow) behind the center key was illuminated and the subject was required to complete one of two FRs (FR 30 or FR 25) on the center key. Completion of the ratio turned off the stimulus behind the center key and illuminated a stimulus (yellow) behind each side key. If the ratio completed was FR 30, a response on the left key resulted in the delivery of a banana pellet. If, however, the ratio completed was FR 25, only a response on the right key was reinforced. Incorrect responses (e.g., pressing the left key following an FR 25) produced a brief, 5-s timeout during which all stimuli were off and responses had no programmed consequences. After either food delivery or a timeout, the stimulus behind the center key was illuminated and the subsequent ratio was programmed with equal probability, either FR 30 or FR 25. The center key remained illuminated until the ratio was completed. Sessions were conducted 5 days a week (Monday–Friday). Each session lasted 60 min.

Drugs

Dose–effect curves were determined for scopolamine HBr and methylscopolamine bromide (Sigma Chemical Co., St. Louis MO). Both drugs were dissolved in 0.9% sterile saline. Drugs and vehicle control (saline) injections were given IM (gluteus muscle) 15 min prior to the start of the experimental session. The volume of injection for each drug was 0.5 ml/kg of body weight. At least 1 week of baseline (nondrug or saline) sessions intervened between the end of the series of injections with scopolamine and the start of the series with methscopolamine. Drug sessions were conducted on Tuesdays and Fridays with control sessions (saline) on Thursdays. Baseline sessions were conducted on Monday and Wednesday.

Data Analysis

The data for each session were analyzed in terms of: a) the overall FR response rate (total center key responses/time the center key stimulus was on) and b) percentage of errors [incorrect/(incorrect + correct responses)] × 100]. The data for each subject were analyzed by comparing drug sessions with the control range of variability (saline sessions). A drug was considered to have an effect to the extent that the dose data fell outside of the control range. Percent errors were not included in the data analysis when response rate was less than 10 responses/min because of the small number of correct and/or incorrect responses involved.

RESULTS

The effects of scopolamine on overall response rate are shown in the upper panels of Fig. 1. Scopolamine produced dose-related decreases in responding in each of the three subjects. The dose of scopolamine required to decrease response rate to 50% or less of control varied only slightly among subjects: 0.01 mg/kg in SQ A and 0.018 mg/kg in SQ J and SQ W. At higher doses, somewhat greater variability in terms of scopolamine's rate-decreasing effect was seen. For example, in SQ A and SQ J a dose of 0.1 mg/kg scopolamine nearly eliminated response rate. However, in SQ W response rate was never lower than 20 responses/min up to a dose of 0.18 mg/kg scopolamine. The dose of scopolamine was not increased in this subject to prevent untoward antimuscarinic effects.

The effects of scopolamine on percent errors are also shown for each subject in the lower panels of Fig. 1. In general, scopolamine produced a dose-related increase in percent errors in all three subjects. In SQ J and SQ W, scopolamine disrupted accuracy in a dose-related manner up to this dose, after which error levels then asymptoted. The dose of scopolamine required to increase percent errors twofold above control varied only slightly for two subjects: 0.018 mg/kg in SQ A and 0.032 mg/kg in SQ J. In the remaining subject (SQ W), percent errors did not double the control levels. In each

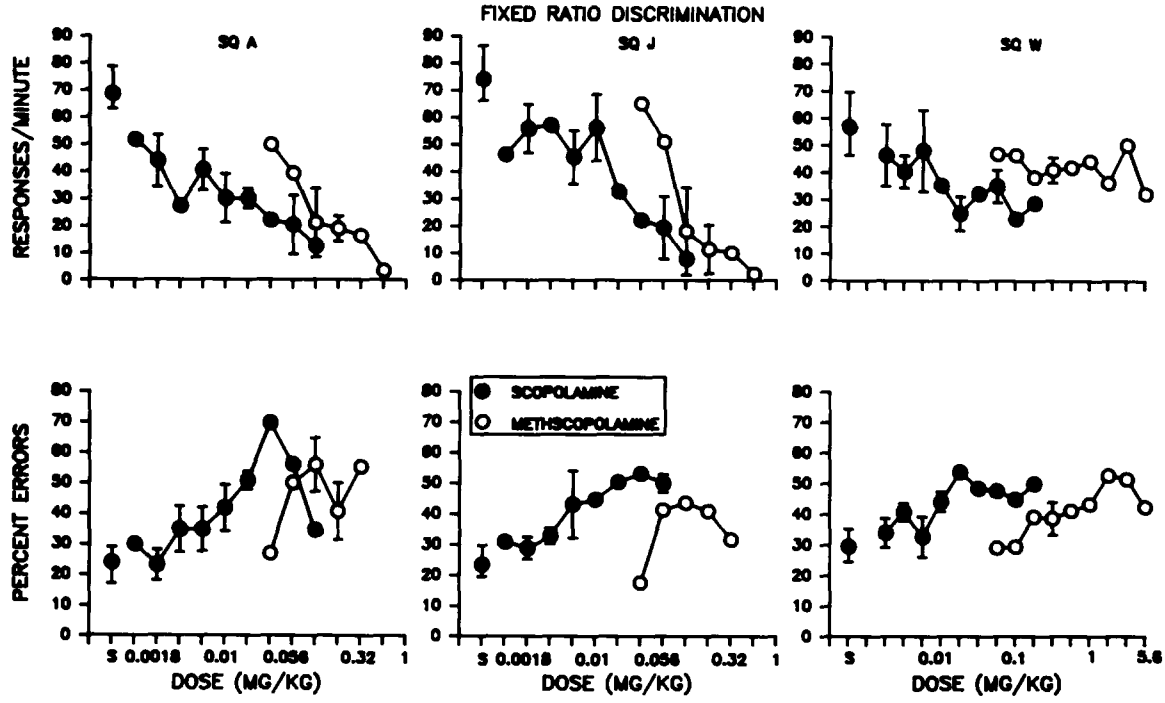

FIG. 1. Effects of varying doses of scopolamine and methscopolamine on the overall response rate and percentage of errors under a fixed-ratio discrimination procedure in each squirrel monkey tested. Points above saline indicate the mean and range of at least 14 sessions that were preceded by a saline injection. The points with vertical lines in the dose–effect curves indicate the mean and range for two determinations. The points without vertical lines indicate either a single determination or an instance in which the range is encompassed by the point.

monkey tested, scopolamine produced its peak disruption in accuracy at a dose of 0.032 mg/kg.

The effects of methscopolamine on response rate are also shown in the upper panels of Fig. 1. Methscopolamine decreased response rate in a dose-related manner, much like scopolamine. However, about 10 times greater doses of methscopolamine were required to produce a comparable decrease in response rate. For example, in SQ A and SQ J it took 0.1 mg/kg methscopolamine to decrease response rate by 50% of control as compared to 0.01 and 0.018 mg/kg scopolamine, respectively. In both SQ A and SQ J, responding was virtually eliminated at a dose of 0.56 mg/kg methscopolamine. In SQ W, response rate never decreased to less than 50% of control, even at 5.6 mg/kg methscopolamine. This is again consistent with the effects of scopolamine in this same subject.

The effects of methscopolamine of the accuracy of responding (percent errors) are shown in the lower panels of Fig. 1. Methscopolamine produced error-increasing effects in all three subjects. The dose of methscopolamine that was required to produce a twofold increase in percent errors was 0.1 mg/kg in SQ A and SQ J. However, like scopolamine, in SQ W methscopolamine did not increase percent errors twofold above the control level. Methscopolamine was found to be about 10 times less potent on a mg/kg basis than scopolamine in relation to its error-increasing effects.

Cumulative records for SQ A illustrating the within-session pattern of responding are shown in Fig. 2. For each drug, the dose that decreased response rate by at least 50% and the highest dose are shown. Scopolamine at a dose of 0.01 mg/kg produced sporadic prerun pausing, followed by bursts of responding. At 0.1 mg/kg, scopolamine produced a pronounced decrease in the rate of responding. Initially, there were many instances of short pauses through the first third of the session. As the session progressed, response rate decreased dramatically and the ratio pattern of responding was grossly disrupted. Methscopolamine at a dose of 0.1 mg/kg produced short, intermittent pausing throughout the session. When the dose of methscopolamine was increased to 0.56 mg/kg, response rate decreased dramatically due to long periods of pausing.

DISCUSSION

The major finding of the present study was that the putatively peripherally acting agent, methscopolamine, exerted rate-decreasing and error-increasing effects similar to those of scopolamine. Slightly higher doses (1/4 of a log unit) of each drug were required to double percent errors than were required to decrease response rate by 50% in two of three subjects. This suggests that in the performance of a discrimination response rate is slightly more sensitive to the disruptive effects of scopolamine and methscopolamine than is percent errors. The fact that scopolamine decreased response rate in all three subjects is consistent with other reports that scopolamine decreases response rates under FR schedules of food presentation (8,11,15). Methscopolamine was found to be about 10 times less potent on a mg/kg basis than scopolamine at decreasing response rate and also disrupting accuracy. These results would suggest that methscopolamine can cross the blood–brain barrier of the squirrel monkey but that a dose 10 times higher is required to produce comparable effects.

The notion has been advanced that centrally mediated

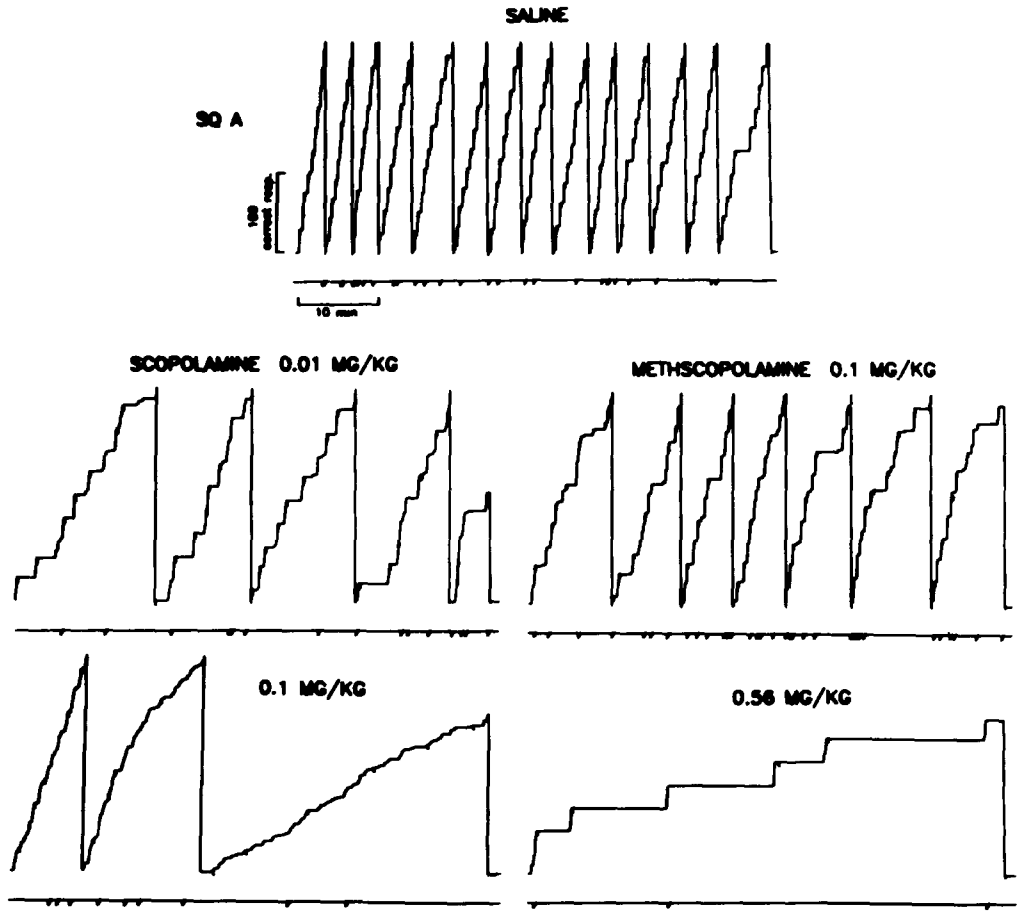

FIG. 2. Cumulative response records for squirrel monkeys showing the pattern of responding during a saline control session that approximated the mean (for both overall response rate and percent errors) and drug sessions as indicated. The response pen stepped up with each correct response and was deflected downward with each reinforcement. Errors are indicated by deflections of the lower event pen.

mechanisms are responsible for the disruptive effects of cholinergic antagonists on accuracy of responding and that peripheral mechanisms are responsible for the rate-decreasing effects (13,14). For example, in one study (3) both scopolamine and methscopolamine were found to decrease the rate of responding in Old World monkeys. Only scopolamine, however, was found to affect accuracy of responding. Such data are in contrast to the present findings, in which both compounds produced decreases in the rate of responding and increases in percent errors. The failure of previous studies to obtain similar results may be due to the fact that inadequate doses were tested. In addition to the present study, evidence that methscopolamine can cross the blood-brain barrier to produce effects like scopolamine is also supported by at least two studies using atropine methylnitrate (9,12). For example, Szerb (12) reported that atropine methylnitrate was approximately 10 times less potent than atropine in producing electroencephalographic synchronization in the curarized cat. Further, it should be pointed out that both scopolamine and methscopolamine have been reported to increase variability of response location rather than producing perseverative responding (8). These data suggest that the error-increasing effects of scopolamine and methscopolamine may result from this action.

An interesting behavioral effect observed after administration of both scopolamine (0.0056–0.18 mg/kg) and methscopolamine (0.056–5.6 mg/kg) was that all three subjects responded for banana pellets but did not consume them. After delivery, the subject would pick up the pellet, rub it in her hands, smell it, and then throw the pellet aside. Subjects continued to respond to the stimuli presented under the procedure but discarded the reinforcers acquired. By the end of the session, there were commonly 20–60 banana pellets discarded on the Plexiglas chair. It should be noted that this effect first occurred at low doses that had no effect on response rate. We have few explanations for this observation. We do not believe that subjects' failure to eat the pellets was a result of dry mouth induced by the drugs. As noted earlier, a water bottle was mounted on the side of the Plexiglas chair, allowing the subject to drink ad lib. If these drugs were causing xerostomia, the monkeys could have drunk at will. This may well have been a drug effect mediated by central mechanisms. The lowest doses at which the discarding of banana pellets was first observed were 0.0056 mg/kg scopolamine and 0.056 mg/kg methscopolamine: a 10-fold shift in potency. Again, this is consistent with the relative potency of these drugs in relation to their rate-decreasing effects.

In a study by Lee and El-Fakahany (6), several muscarinic

receptor antagonists (scopolamine and methscopolamine) were tested for their ability to displace (-)[^{3}H]quinuclidinyl benzilate (QNB) or [^{3}H]*N*-methylscopolamine (NMS) from their specific binding sites in rat brain membranes. Equilibrium dissociation constants for scopolamine (8.94×10^{-10} M) and methscopolamine (6.97×10^{-10} M) were nearly identical for displacing [^{3}H]QNB binding. However, when both drugs were used to displace [^{3}H]NMS binding a slight difference was observed. Methscopolamine (2.55×10^{-10} M) was slightly more potent (4×) than scopolamine (10.35×10^{-10} M) at displacing the bound ligand. From these studies in the rat brain, it can be concluded that scopolamine and its quaternary derivative methscopolamine have nearly equal affinities for muscarinic receptors labelled by [^{3}H]QNB and that methscopolamine is slightly more potent than scopolamine at binding to muscarinic receptors labeled by [^{3}H]NMS. Because these experiments were done on rat membranes after sacrificing the animal, there was no blood-brain barrier for the drug to pass through. Depending upon the ligand used, scopolamine and methscopolamine have nearly equal affinity or methscopolamine has slightly higher affinity than does scopolamine. Given that these two compounds have nearly the same affinity, the difference observed in relative potency must be due to other mechanisms. However, methscopolamine does not readily cross the blood-brain barrier as does the lipophilic scopolamine. The present data are, therefore, consistent with the notion that higher concentrations of methscopolamine are needed to cross the blood-brain barrier than are needed for scopolamine.

In summary, the muscarinic antagonists scopolamine and methscopolamine both decreased response rate and increased percent errors in a dose-related manner in all three squirrel monkeys responding under the fixed-ratio discrimination procedure. Scopolamine was found to be about 10 times more potent (on a mg/kg basis) than methscopolamine. The data indicate that methscopolamine can cross the squirrel monkey blood-brain barrier to exert its behavioral effects but to a lesser degree than scopolamine.

ACKNOWLEDGEMENTS

This research was supported by U.S. Public Health Service Grants DA 03573 and DA 04775. The authors thank C.P. France for helpful comments on this manuscript.

REFERENCES

1. Aigner, T. G.; Mishkin, M. The effects of physostigmine and scopolamine on recognition memory in monkeys. Behav. Neural Biol. 45:81–87; 1986.
2. Bartus, R.; Johnson, H. Short-term memory in the rhesus monkey: Disruption from the anticholinergic scopolamine. Pharmacol. Biochem. Behav. 5:39–46; 1976.
3. Bohdanecky, Z.; Jarvik, M. E.; Carley, J. L. Differential impairment of delayed matching in monkeys by scopolamine and scopolamine methylbromide. Psychopharmacologia 11:293–299; 1967.
4. Brown, J. H. Atropine, scopolamine, and related antimuscarinic drugs. In: Gilman, A. G.; Rall, T. W.; Nies, A. S.; Taylor, P., eds. The pharmacological basis of therapeutics. 8th ed. New York: Macmillan; 1990:150–165.
5. Evans, H. L. Scopolamine effects on visual discrimination: Modifications related to stimulus control. J. Pharmacol. Exp. Ther. 195:105–113; 1975.
6. Lee, J. H.; El-Fakahany, E. E. Heterogeneity of binding of muscarinic receptor antagonists in rat brain homogenates. J. Pharmacol. Exp. Ther. 233:707–714; 1985.
7. Moerschbaecher, J. M.; Mastropaolo, J.; Winsauer, P. J.; Thompson, D. M. Effects of opioids on accuracy of a fixed-ratio discrimination in monkeys and rats. J. Pharmacol. Exp. Ther. 230:541–549; 1984.
8. Moerschbaecher, J. M.; Thompson, D. M.; Thomas, J. R. Effects of methamphetamine and scopolamine on variability of response location. J. Exp. Anal. Behav. 32:255–263; 1979.
9. Paul-David, J.; Riehl, J. L.; Unna, K. R. Quantification of effects of depressant drugs on EEG activation response. J. Pharmacol. Exp. Ther. 129:69–74; 1960.
10. Rupniak, N. M. J.; Samson, N. A.; Steventon, M. J.; Iversen, S. D. Induction of cognitive impairment by scopolamine and noncholinergic agents in rhesus monkeys. Life Sci. 48:893–899; 1991.
11. Shannon, H. E.; Bemis, K. G.; Hart, J. C. Assessment of working memory in rats using spatial alternation behavior with variable retention intervals: Effects of fixed-ratio size and scopolamine. Psychopharmacology 100:491–497; 1990.
12. Szerb, J. C. The effect of tertiary and quaternary atropine on cortical acetylcholine output and on the electroencephalogram in cats. Can. J. Physiol. 42:303–314; 1964.
13. van Haaren, F.; van Hest, A.; van Hattum, T. Scopolamine and methylscopolamine differentially affect fixed-consecutive-number performance of male and female Wistar rats. Pharmacol. Biochem. Behav. 33:361–365; 1989.
14. Viscardi, A. P.; Heise, G. A. Effects of scopolamine on components of delayed response performance in the rat. Pharmacol. Biochem. Behav. 25:633–639; 1986.
15. Wenger, G. R. Effects of physostigmine, atropine, and scopolamine on behavior maintained by a multiple schedule of food presentation in the mouse. J. Pharmacol. Exp. Ther. 209:137–143; 1979.