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BRIEF COMMUNICATION

Amnesic Effect of the Novel Anticonvulsant MK-801

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BENVENGA, M. J. AND T. C. SPAULDING. *Amnesic effect of the novel anticonvulsant MK-801.* PHARMACOL BIOCHEM BEHAV **30**(1) 205–207, 1988.—MK-801, a reported N-methyl-D-aspartate (NMDA) antagonist with affinity for the phencyclidine (PCP) receptor, injected intravenously in mice before a training trial in a passive avoidance procedure, produced a similar amnesic effect to that produced by the standard amnesic agent scopolamine. Compared to vehicle-treated mice, each drug produced significant amnesia, yet the potency of MK-801 was 40 times that of scopolamine. This result with the MK-801 is consistent with previous reports that drugs which act at PCP recognition sites within the brain produce memory impairing effects in rodents.

Amnesia MK-801 Phencyclidine Scopolamine

MK-801, [(+)-5-methyl-10,11-dihydro-5H-dibenzo[a,d]cyclohepten-5,10-imine maleate], a novel anticonvulsant [3] has been reported as a receptor antagonist of N-methyl-D-aspartate (NMDA) in rat brain [10]. MK-801 has also been reported to produce its antagonistic effects on NMDA-type receptors via a direct interaction with phencyclidine (PCP) binding sites associated with NMDA receptors [7]. Along with this, glutamate has the ability to enhance the affinity of MK-801 for the PCP binding site similar to that observed with the dissociative anesthetics phencyclidine and ketamine [7]. Since it has recently been reported that phencyclidine and ketamine can inhibit NMDA-induced neuronal responses [1] and that phencyclidine impairs memory in both rats [5] and mice [6,9], it was of interest to investigate the possible memory impairing effects of the novel NMDA antagonist, MK-801.

METHOD

Male mice (Swiss-Webster; Hilltop Farms, PA) were housed 10 per cage in a temperature (70±2°F) and humidity (50+5%) controlled vivarium with a light-dark cycle of 12 hours (0600–1800 lights on). Prior to testing, the animals were allowed ten minutes to adapt to laboratory conditions.

For behavioral assessment, a one-trial passive avoidance task was employed. The test chambers (Coulbourn Instruments) had two compartments with dimensions 16×17.5×21 cm with a metal grid floor through which electric shock could be delivered. The compartments were connected by a guillotine door measuring 8×9 cm which could be raised to give access to the compartment beyond. The test chamber was located within a large sound and light-attenuating enclosure. For experimental purposes, either side of the test chamber could be lighted and a naive mouse when placed in the lighted compartment would enter the dark chamber with a short latency when given the opportunity.

To test for memory-impairing effects, MK-801 was compared to the standard amnesic drug, scopolamine. Drugs were dissolved in distilled water and injected 0.1 ml/kg body weight as a bolus into the tail vein.

On the first experimental day, the training trial was run. Twenty minutes following injections of MK-801 (0.025, 0.05, 0.10 or 0.15 mg/kg; N=8/dose), scopolamine (0.5, 1.0, 2.0, 4.0 mg/kg, N=8/dose), or vehicle (N=8), mice were placed into the lit chamber and given access to the dark chamber by raising the guillotine door. Upon crossing over into the dark chamber, the guillotine door was closed and the mice received inescapable foot-shock (1 mA) for one second after which they were immediately removed from the apparatus and returned to their home cage. The conditioned inhibition to re-enter the dark chamber was then tested 24 hours later comparing the drugged mice to the vehicle-treated mice. A maximum of 300 seconds was used for cut-off on the second day if the mice failed to enter the dark chamber at that time. The within drug group medians were first analyzed with the Kruskal-Wallis H statistic and if appropriate, the median latency for each drug group was compared to the vehicle group using a Mann-Whitney U test.

MK-801 was a generous gift of Merck, Sharp and Dohme (Westpoint, PA) while scopolamine hydrochloride was obtained commercially (Sigma, St. Louis, MO).

RESULTS

The results of the behavioral assessment for each drug are indicated in Table 1. For each drug, a vehicle control group

TABLE 1

MEDIAN AND RANGE OF DAY-1 AND DAY-2 ENTRANCE LATENCIES FOR SCOPOLAMINE AND MK-801

Dose	Day-1 Median	Range	Day-2 Median	Range
		Scopolamine (N=8/dose)		
Control	22.8	12.5- 79.0	300.0	0
0.5	21.2	9.4- 61.3	287.9	14.8-300.0
1.0	32.2	14.6-135.1	203.9	107.4-300.0
2.0	26.3	5.7-100.7	222.2	15.4-300.0
4.0	21.3	6.7-106.1	30.0*	1.9-300.0
		MK-801 (N=8/dose)		
Control	32.3	9.1-281.9	300.0	0
0.025	30.5	1.0- 87.5	300.0	156.2-300.0
0.050	10.8	1.0- 50.8	224.9	7.7-300.0
0.100	11.0	1.7- 42.5	78.9*	30.8-300.0
0.150	17.2	1.0- 44.8	64.7*	15.6-300.0

*Significantly different from control $p<0.05$.

was used for comparison of amnesic effect. In all cases, mice treated with vehicle prior to the initial test did not respond within the 300 second cut-off time when processed 24 hours later in the test session.

For both drugs, the Day-1 latencies comparison revealed no significant differences for either scopolamine, $H(4)=2.09$, $p<0.05$, or MK-801, $H(4)=8.07$, $p<0.05$. This comparison became necessary when it was noted that both scopolamine (2.0 and 4.0 mg/kg) and MK-801 (0.10 and 0.15 mg/kg) produced an increase in locomotor activity at doses which caused an amnesic effect. A subsequent study in our laboratory (to be reported elsewhere) revealed that this increase in locomotor activity was not significant between drugs and was not a causative factor in the amnesic effect.

Analysis of the Day-2 latencies revealed a significant effect for both scopolamine, $H(4)=11.11$, $p<0.05$, and MK-801, $H(4)=16.75$, $p<0.05$. A subsequent Mann-Whitney U-test revealed that scopolamine produced an amnesic effect only at the highest dose tested (4.0 mg/kg). Lower doses (1.0 and 2.0 mg/kg) produced median latencies which were shorter than the control latency, however, these effects did not reach statistical significance.

MK-801, on the other hand, produced an amnesic effect at both the 0.10 and 0.15 mg/kg doses which were statistically significant from the control group. The 0.05 mg/kg dose did produce an effect, but it was not significant.

RESULTS AND DISCUSSION

In this study, both scopolamine and MK-801 were shown to produce an amnesic effect in mice. However, the potency to produce these effects was quite different. While scopolamine produced amnesia at 4.0 mg/kg, MK-801 was able to produce a significant effect as low as 0.1 mg/kg. This result is in contrast to results reported previously by Glick and Zimmerberg [6] who found that PCP was less potent than scopolamine in causing an amnesic effect when administered before the training trial in a one-trial passive avoidance paradigm. Similarly, Duncan [5] reported PCP to be less potent than scopolamine in affecting memory in rats. In our study, MK-801, a drug which acts through PCP binding sites [7], was found to be much more potent than scopolamine in mice.

Both scopolamine and MK-801 produced an increase in locomotor activity at doses which coincided with their amnesic effect. Subsequently, we investigated the locomotor effects of these drugs and determined that this effect did not contribute to the amnesic effect.

Another factor which may contribute to our finding is that MK-801 may have some analgesic effects which could cause a sensory deficit rather than a memory deficit. Since it has previously been reported that naloxone, an opiate antagonist, can inhibit PCP-induced amnesia [9] when it is given before the training trial, this possibility certainly exists. We are preparing to investigate this possibility in the future, but we believe that the dose-range of MK-801 used in our study, relative to the PCP dose-range reported by Nabeshima *et al.* [9], precludes this possibility.

These authors [9] have also reported that PCP causes a retrograde amnesia when administered after the training trial. This type of effect was not considered in our experimental design and clearly needs to be addressed in the future.

It has been reported previously that scopolamine, an anticholinergic, produces profound amnesic effects in rodents [2,8], presumably as a result of an interaction with acetylcholine receptors in the brain. MK-801, on the other hand, previously reported to not possess any affinity for receptors of acetylcholine [4], produces amnesic effects through a yet unknown mechanism. Since MK-801 has been shown to interact with NMDA receptors [2,10], PCP receptors [3,7] and to a lesser extent, benzodiazepine receptors [10], many possible mechanisms of its memory-impairing effects exist. A likely mechanism is that MK-801 produces an antagonism at the NMDA receptor site complex through a direct interaction with PCP binding sites, and like phencyclidine, causes a memory-impairing effect in this way.

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