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The Effects of Haloperidol, Scopolamine, and MK-801 on Amphetamine-Induced Increases in Ascorbic and Uric Acid as Determined by Voltammetry In Vivo

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SAPONJIC, R. M., K. MUELLER, D. KRUG AND P. M. KUNKO. *The effects of haloperidol, scopolamine, and MK-801 on amphetamine-induced increases in ascorbic and uric acid as determined by voltammetry in vivo.* PHARMACOL BIOCHEM BEHAV **48**(1) 161-168, 1994.—Amphetamine (which enhances dopaminergic, cholinergic, and glutamatergic activity) increases release of ascorbic acid (AA) and uric acid (UA) in the caudate nucleus. In this study, linear sweep voltammetry with carbon past electrodes was used to investigate the effects of haloperidol (a DA receptor blocker), scopolamine (a muscarinic receptor blocker), and MK-801 (an NMDA receptor blocker) alone and in combination on amphetamine-induced increases in AA and UA in the caudate nucleus. Both scopolamine (0.5 mg/kg, IP) and MK-801 (0.5 mg/kg, IP) significantly reduced amphetamine-induced increases in AA. Also, scopolamine did not affect MK-801-induced reductions of amphetamine-induced increases in AA. Unexpectedly, a subthreshold dose of haloperidol (0.1 mg/kg, IP) potentiated the ability of scopolamine to block amphetamine-induced increases in AA. Therefore, the data suggest that acetylcholine release and subsequent binding to cholinergic receptors in the caudate, are components of amphetamine-induced increases in AA. In addition, scopolamine modulated haloperidol-induced reductions of amphetamine-induced increases in release of UA. Thus, our data demonstrate that cholinergic and dopaminergic systems may interact to control release of UA.

Ascorbic acid Haloperidol MK-801 Scopolamine Amphetamine Uric acid Voltammetry

ASCORBIC acid (AA) and uric acid (UA) are present in brain; however, their function (if any) or origin is not yet understood. Amphetamine increases release of AA and UA. If this effect is dependent upon stimulation of dopaminergic, cholinergic, and/or glutamatergic systems, then haloperidol (a dopamine receptor blocker), scopolamine (a muscarinic anticholinergic), and MK-801 (noncompetitive NMDA antagonist) should reduce the effects of amphetamine on AA and UA in caudate. The research described below reports the effects of haloperidol, scopolamine, and MK-801 alone and in combination on amphetamine-induced increases of striatal AA and UA.

AA is found in high concentrations throughout the mammalian brain (16); however, AA is not synthesized in the brain (34). AA is transported to the brain via the circulatory system and transferred from blood plasma to cerebrospinal fluid (CSF) by the choroid plexus via an active transport system (31). Ascorbic acid levels in the brain are carefully regulated (29); thus, AA might be important for proper functioning of the brain. In addition, ascorbic acid is released from synaptosomes (10) and enhances release of neurotransmitters from synaptosomes (14). AA also affects the binding of several neurotransmitters (30,33). Thus, AA may function as a neurotransmitter or neuromodulator in brain.

Three neurotransmitters are under investigation for regulating release of AA: dopamine, glutamate, and acetylcholine. Amphetamine (an indirect dopamine agonist) and pergolide (a dopamine agonist) increase extracellular levels of AA (5,

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9,25,28,35). Therefore, AA initially appeared to be intimately involved with dopaminergic neurotransmission.

However, other studies have failed to support dopaminergic regulation of release of AA. Amphetamine-induced increases in extracellular levels of AA in caudate continue despite depletion of dopamine (12). Furthermore, AA has been found to be released under conditions in which dopamine release could not occur (2). Thus, the association (if any) between dopamine and AA remains tenuous.

Others have suggested that extracellular levels of AA may be modulated by the excitatory amino acids glutamate and aspartate (3,23). For example, administration of subtoxic doses of glutamate increases levels of AA in the caudate nucleus as does administration of aspartate. In addition, blockers of glutamate uptake, homocysteic acid, and D,L-threo-beta-hydroxy-aspartic acid eliminate the ascorbate signal in the hippocampus and thalamus (3). In particular, these authors suggest that extracellular levels of AA are mediated by the high-affinity uptake system of glutamate. Finally, lesions of the corticostriatal pathway, which uses glutamate as a transmitter, blocked amphetamine-induced increases in AA by 74% (8).

However, a recent study has failed to replicate these findings. For example, cortically lesioned animals demonstrated increased release of AA vs. sham and control groups following amphetamine (1). Thus, the role of cortical structures and the release of glutamate in control of extracellular levels of AA warrants further study.

Extracellular AA may also be modulated by the cholinergic system. The cholinergic agonist, pilocarpine, increased striatal AA, while scopolamine reduced the pilocarpine-induced increases in AA (18,20). Indirectly acting dopamine agonists, such as *d*-amphetamine, increase release of acetylcholine in the caudate nucleus (4,6,15). Preliminary investigations reveal that scopolamine blocks amphetamine-induced increases in AA by 50% (13). These data suggest that amphetamine-induced increases in AA are controlled, at least in part, by cholinergic receptor stimulation.

Uric acid, a byproduct of purine metabolism, has not been extensively studied in brain. Previous studies have demonstrated that both scopolamine and amphetamine increased release of UA in the caudate nucleus (20,21). The amphetamine-induced increase in UA is blocked by haloperidol, while the scopolamine-induced increase is blocked by pilocarpine (a cholinergic agonist).

The function of UA (if any) in brain remains controversial. Some purines, such as ATP and adenosine, are released from brain tissue and are, therefore, thought to function as neuromodulators or neurotransmitters in brain (27,32). Because UA is a byproduct of adenosine metabolism, UA may provide information about release of purinergic neurotransmitters (22).

Another possibility is that extracellular UA may be a nonspecific index of metabolic activity. Nucleotides such as ATP function as carriers of chemical energy. Because UA is a purine metabolite, increased amounts of UA would be expected during increased energy use (11,20).

The majority of research discussed above suggests that dopamine, glutamate, and acetylcholine may be associated with regulation of release of extracellular AA and UA. In the research described below, voltammetric electrodes were placed in the anterior caudate. The first and second experiments examined the AA and UA response to administration of scopolamine and MK-801. The following four experiments examined the effects of combinations of scopolamine, haloperidol, and MK-801 on amphetamine-induced increases in AA and UA.

GENERAL METHOD

Animals

Male Wistar rats were selected from the breeding colony at Texas Christian University. The rats were housed on a 12 L : 12 D cycle. Food and water were freely available. Preoperative body weights ranged from 370 to 480 g.

Procedure

Carbon paste working electrodes were fabricated by pulling the Teflon coating over the end of a stainless steel wire (Medwire, SS8T) and filling the resultant cavity with carbon paste (21). Carbon paste was made by mixing silicone oil (0.214 g) with CCl_4 (3.5 ml); followed by carbon (0.5 g). The carbon paste mixture was left under a vapor hood to allow the remaining CCl_4 to evaporate.

After pretreatment with atropine, rats were anesthetized with Nembutal (50 mg/kg). Surgery was performed under standard stereotaxic procedures. Working electrodes were implanted in the anterior caudate (2.6 mm anterior to bregma, 2.8 mm lateral to bregma, and 5.0 mm beneath the cortex) (24). An Ag/AgCl reference electrode was also implanted and a silver wire attached to a skull screw provided an auxiliary electrode.

Forty-eight hours after surgery animals were connected to the apparatus, but no drugs were administered. Four days after surgery, animals were connected to the voltammetry apparatus and after a 2 h recording period were injected. Testing was always conducted 1.5 to 2 h after lights on. The percent increase in peak height (over the mean of three scans prior to the first injection) was the dependent variable.

A DCV-5 voltammetry controller (Bioanalytical Systems) controlled via a personal computer performed linear sweep (10 mV/s) recordings from -100 to 500 mV. Electrodes were scanned every 12 min, producing two distinctive peaks. Previous research from this lab has identified the first peak as AA and the second peak as UA (17,21). Animals were connected to the apparatus by a swivel to provide freedom of movement.

At the completion of testing, animals were given a lethal dose of Nembutal and were perfused intracardially with saline followed by a 10% formalin solution. After perfusion, brains were removed and sectioned at 20 μm. Data from rats with misplaced electrodes were discarded.

Experiments

Experiment 1 tested the effects of various doses of scopolamine on release of AA and UA. In experiment 1, all animals (n = 6) received four treatments (IP) in randomized order: 0.1 mg/kg scopolamine, 0.3 mg/kg scopolamine, 0.8 mg/kg scopolamine, or saline. Each treatment session was separated by 48 h. The experiment was designed to determine if scopolamine alone was producing an effect on AA and UA. The data were analyzed with a repeated analysis of variance (ANOVA) with two within groups variables: dose and time.

Experiment 2 tested the effects of various doses of MK-801 on release of AA and UA. In Experiment 2, all animals (n = 7) received four treatments (IP) in randomized order: 0.05 mg/kg, 0.10 mg/kg, 0.50 mg/kg, or saline. Each treatment session was separated by 48 h. The experiment was designed to determine if MK-801 alone was producing an effect on AA and UA. The data were analyzed with a repeated ANOVA with two within-group variables: dose and time.

Experiment 3 tested the effects of scopolamine on amphetamine-induced increases in AA and UA. In Experiment 3, all

animals were randomly assigned to three groups with different rats in each group (n = 5 for each group) and pretreated with either 0.3 mg/kg, 0.8 mg/kg scopolamine, or saline (IP) and 36 min later received 1.5 mg/kg d-amphetamine sulfate (Sigma) subcutaneously (SC). The data were analyzed with a mixed ANOVA with one between-groups variable (treatment) and one within-groups variable (time).

Experiment 4 tested the effects of a combination of haloperidol and scopolamine on amphetamine-induced increases in AA and UA. In Experiment 4, all animals were randomly assigned to four groups with different rats in each group (n = 6 for each group) and subsequently received (IP) one of four pretreatments. A pretreatment consisted of two drugs administered in combination; haloperidol (0.1 mg/kg; a subthreshold dose) plus scopolamine (0.5 mg/kg), haloperidol plus saline, scopolamine plus saline, or saline plus saline. Previous research from this lab has determined that haloperidol (0.2 mg/kg) has no effect on resting and amphetamine-induced increases of AA (19). Thirty-six minutes after pretreatment all animals were injected (SC) with amphetamine (2.0 mg/kg). A higher dose of amphetamine was used in this study to produce larger increases in release of AA. Hopefully, potential differences in the ability of haloperidol and scopolamine to block amphetamine-induced increases in AA would be more apparent. The data were analyzed using a three-way (main effect of haloperidol, main effect of scopolamine, and main effect of time) ANOVA, with time as a repeated measure.

Experiment 5 tested the effects of MK-801 on amphetamine-induced increases in AA and UA. In Experiment 5, all animals were randomly assigned to four groups with different rats in each group and pretreated (IP) with either 0.05 mg/kg MK-801 (n = 3), 0.1 mg/kg MK-801 (n = 4), 0.5 mg/kg MK-801 (n = 5), or saline (n = 6). Forty-eight minutes

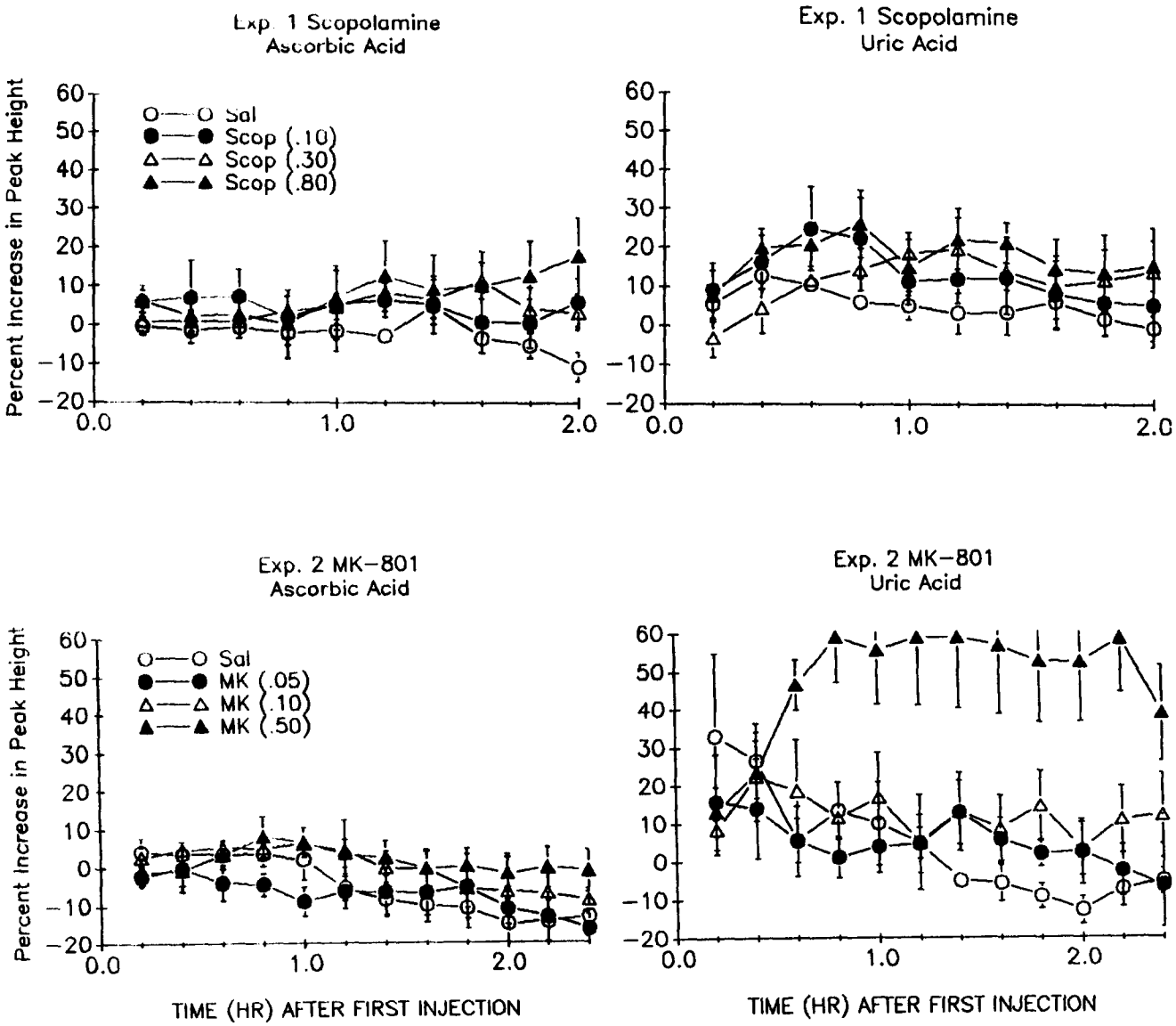

FIG. 1. The effects of various doses of scopolamine (top) and MK-801 (bottom) on release of ascorbic and uric acid. Doses are given in mg/kg. Sal = saline. Scopolamine and MK-801 were administered at time "0" on the graphs.

following pretreatment all animals were injected (SC) with amphetamine (1.5 mg/kg). The data were analyzed with a repeated mixed ANOVA with one between-groups variable (treatment) and one within-groups variable (time).

Experiment 6 tested the effects of a combination of MK-801 and scopolamine on amphetamine-induced increases in AA and UA. In Experiment 6, all animals were randomly assigned to four groups with different rats in each group and subsequently received one of four pretreatments (IP). A pretreatment consisted of two drugs administered in combination; MK-801 (0.1 mg/kg) plus scopolamine (0.3 mg/kg) (n = 7), MK-801 plus saline (n = 6), scopolamine plus saline (n = 6), or saline plus saline (n = 7). Forty-eight minutes after pretreatment, all animals were injected (SC) with amphetamine (2.0 mg/kg). As noted previously, a higher dose of amphetamine was used to help determine potential differences between the ability of MK-801 and scopolamine to block amphetamine-induced increases in AA. The data were analyzed using a three-way (main effect of MK-801, main effect of scopolamine, and main effect of time) ANOVA, with time as a repeated measure.

RESULTS

Scopolamine

In general, scopolamine failed to modulate release of AA (see Fig. 1). Although (0.8 mg/kg) scopolamine seemed to minimally increase release of AA, this effect was not significant.

Scopolamine failed to significantly modulate release of UA (see Fig. 1). Neither the main effect of scopolamine (0.1, 0.3, and 0.8 mg/kg) nor the interaction between dose and time after injection were significant.

MK-801

MK-801 did not affect release of AA (see Fig. 1). Neither the main effect of MK-801 (0.05, 0.10, and 0.50 mg/kg) nor the interaction between dose and time after injection were significant.

The highest dose of MK-801 (0.50 mg/kg) dramatically increased release of UA, $F(39, 234) = 4.28$, $p < 0.0008$, for the interaction between dose and time (see Fig. 1). However, the two lower doses of MK-801 (0.05 and 0.10 mg/kg) did not significantly affect release of UA.

Scopolamine and Amphetamine

Both doses of scopolamine (0.3 and 0.8 mg/kg) reduced amphetamine-induced increases in AA (see Fig. 2). Although the main effects were not significant, the interaction between group and time was significant, $F(1, 12) = 4.32$, $p < 0.01$.

In contrast, scopolamine did not affect amphetamine-induced increases in UA (see Fig. 2). Neither the main effect for scopolamine (0.3 and 0.8 mg/kg) nor the interaction between treatment and time after injection were significant.

Haloperidol, Scopolamine, and Amphetamine

As expected, a low dose of haloperidol (0.1 mg/kg) alone failed to affect the amphetamine-induced increase in AA (see Fig. 2). Scopolamine (0.5 mg/kg) alone moderately reduced (49%) the effect of amphetamine on AA, $F(11, 220) = 5.25$, $p < 0.0001$. However, the combination of haloperidol plus scopolamine almost completely blocked (89%) the effect of amphetamine on AA, $F(11, 220) = 2.41$, $p < 0.03$, for the three-way interaction. Thus, the subthreshold dose of haloperidol potentiated the effects of scopolamine.

Haloperidol alone moderately reduced (37%) amphetamine-induced increases in UA, $F(11, 220) = 2.72$, $p < 0.008$, for the interaction between time and main effect of haloperidol (see Fig. 2). Scopolamine alone failed to affect the amphetamine-induced increases in UA. In addition, the combination of haloperidol and scopolamine failed to significantly affect amphetamine-induced increases in UA. Thus, scopolamine blocked the effect of haloperidol on amphetamine-induced increases in release of UA.

MK-801 and Amphetamine

The highest dose of MK-801 (0.50 mg/kg) reduced (93%) amphetamine-induced increases in AA, $F(39, 182) = 1.84$, $p < 0.03$, for the interaction between time and treatment (see Fig. 3). However, the two lower doses of MK-801 (0.05 and 0.10 mg/kg) did not significantly affect amphetamine-induced increases in AA.

MK-801 failed to significantly affect the amphetamine-induced increases in UA (see Fig. 3). The main effect of MK-801, $F(3, 14) = 0.86$, $p < 0.433$, and the interaction between time after injection and MK-801, $F(39, 182) = 1.55$, $p < 0.08$, were not significant. Although the low dose of MK-801 (0.05 mg/kg) seemed to increase release of UA, this effect was not significant.

MK-801, Scopolamine, and Amphetamine

As expected, MK-801 (0.1 mg/kg) significantly reduced (73%) amphetamine-induced increases in AA, $F(13, 286) = 19.16$, $p < 0.001$, for interaction between time and main effect of MK-801] (see Fig. 3). Scopolamine (0.3 mg/kg) alone failed to affect amphetamine-induced increases in AA. In addition, the combination of MK-801 and scopolamine reduced amphetamine-induced increases in AA; however, the three-way interaction of main effect of MK-801, main effect of scopolamine, and main effect of time, $F(13, 286) = 1.53$, $p < 0.17$, was not statistically significant.

MK-801 slightly affected amphetamine-induced increases in UA, $F(13, 286) = 5.19$, $p < 0.001$, for the interaction between main effect of MK-801 and main effect of time (see Fig. 3). Scopolamine alone failed to affect the amphetamine-induced increases in UA. In addition, the combination of MK-801 and scopolamine did not affect amphetamine-induced increases in UA.

DISCUSSION

The purpose of these experiments was to determine if dopamine, acetylcholine, and/or glutamate neurotransmitter systems interact to regulate release of AA and UA. The most important findings of these experiments are as follows: a) scopolamine reduced amphetamine-induced increases in AA while not affecting resting levels of AA, b) a subthreshold dose of haloperidol enhanced scopolamine-induced reductions of amphetamine-induced increases in AA, c) MK-801 reduced amphetamine-induced increases in AA while not affecting resting levels of AA, d) Scopolamine failed to modulate the effectiveness of MK-801 in reducing amphetamine-induced increases in AA, e) haloperidol reduced amphetamine-induced increases in UA, f) scopolamine blocked the effect of haloperidol on amphetamine-induced increases in UA.

Ascorbic Acid

In two of three experiments, the amphetamine-induced increase in AA was reduced by the cholinergic receptor blocker,

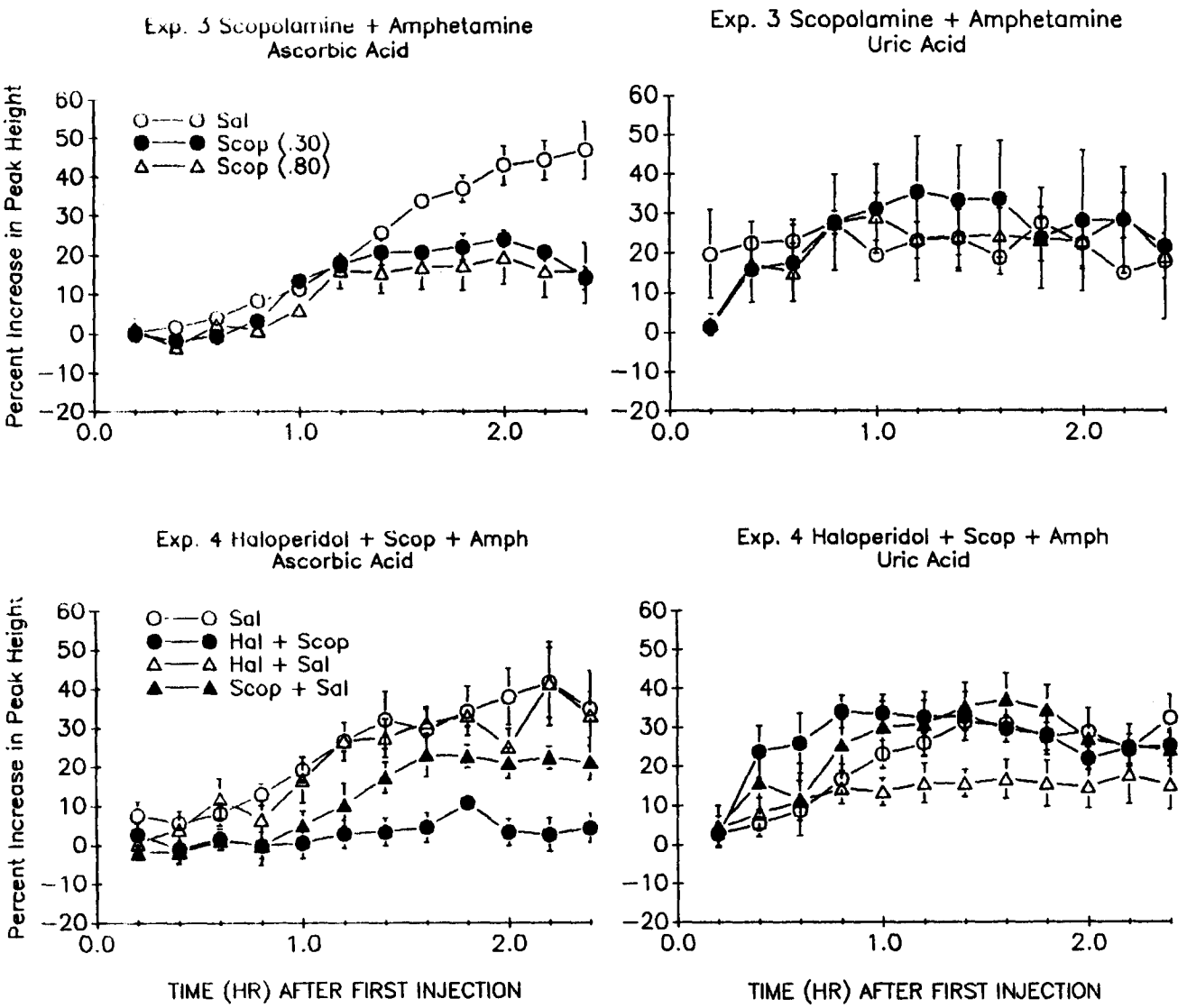

FIG. 2. The effects of scopolamine (top) and haloperidol plus scopolamine (bottom) on amphetamine-induced increases in ascorbic and uric acid. Doses are given in mg/kg. Sal = saline. In Experiment 3, the dose of amphetamine was 1.5 mg/kg, while in Experiment 4, the dose of amphetamine was 2.0 mg/kg. Scopolamine and haloperidol were injected at time "0," while amphetamine was injected 36 min later.

scopolamine. Although scopolamine (0.3 mg/kg) reduced amphetamine-induced increases in AA in Experiment 3 (scopolamine and amphetamine), scopolamine failed to affect amphetamine-induced increases in AA in Experiment 6 (MK-801, scopolamine, and amphetamine). The difference in findings may be attributed to the dose of amphetamine used in Experiment 6. In Experiment 6, a higher dose of amphetamine (2.0 mg/kg) vs. (1.5 mg/kg) was used. The higher dose of amphetamine may have masked any effect of scopolamine on amphetamine-induced increases in AA.

Thus, cholinergic receptor stimulation may mediate release of AA in the caudate nucleus. This hypothesis is supported by findings that amphetamine increases release of acetylcholine in the caudate nucleus (4,15). In accordance with our results, these data demonstrate that acetylcholine release in the caudate nucleus is responsible for a component of amphetamine-induced increases in AA.

The origin of amphetamine-induced increases in acetylcholine and AA remains controversial. For example, local application of amphetamine fails to release AA (36) or acetylcholine (7). Thus, these data suggest that the effect of systemic amphetamine on AA and acetylcholine is located outside the striatum. In addition, recent investigations have suggested that amphetamine increases release of acetylcholine indirectly by increasing glutamatergic neurotransmission in the caudate nucleus via the corticostriatal pathway (6). Subsequent increases in acetylcholine may increase release of AA.

The reduction of amphetamine-induced increases in AA by MK-801 in our study and others (26) may be explained by the above model. If amphetamine increases release of acetylcholine by indirectly increasing glutamatergic neurotransmission, then NMDA receptor blockers (e.g., MK-801) should block the cholinergic component of amphetamine-induced increases in AA.

Finally, the similarity of reduction between MK-801 and treatment with MK-801 plus scopolamine suggests that cholin-

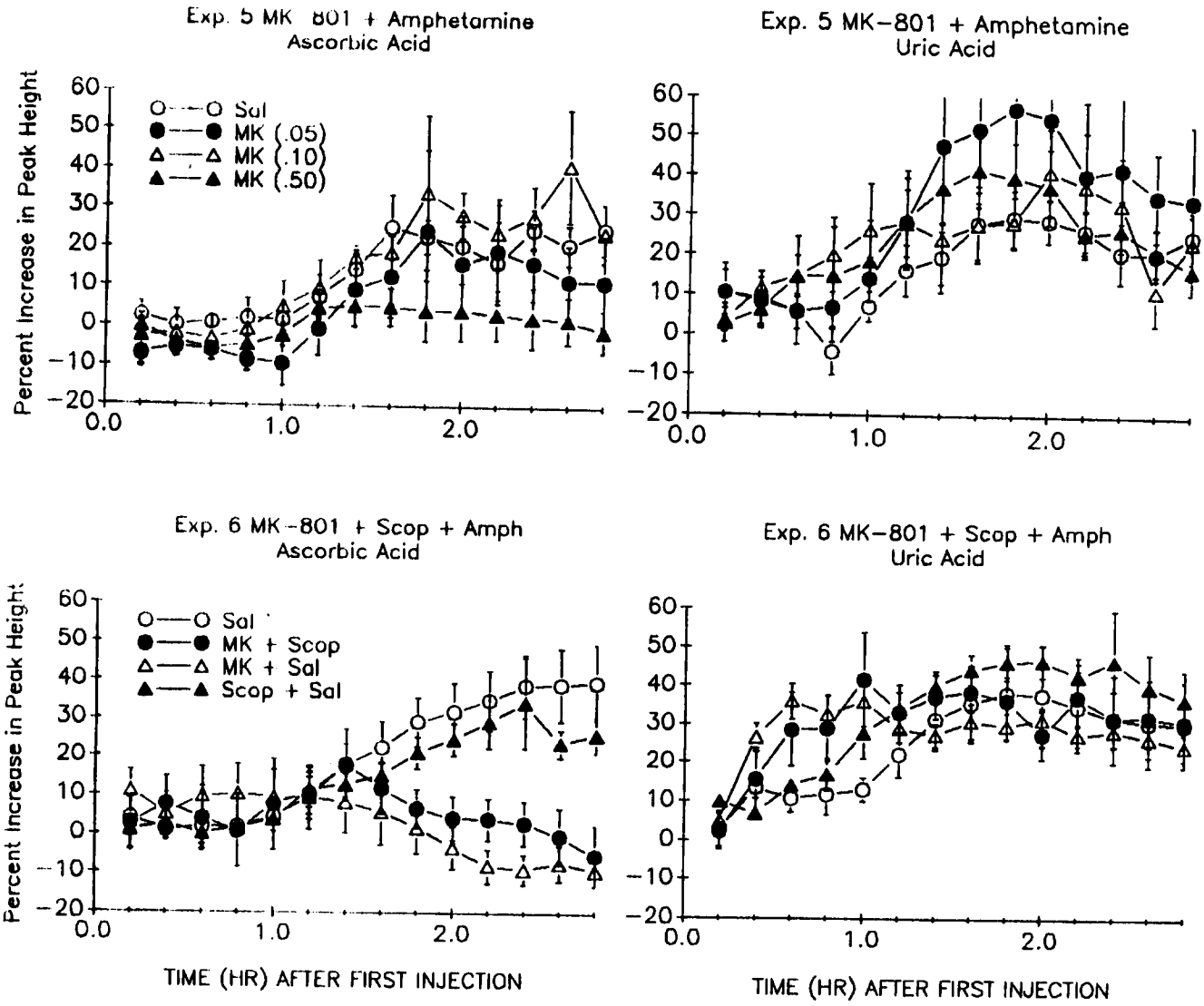

FIG. 3. The effects of MK-801 (top) and MK-801 plus scopolamine (bottom) on amphetamine-induced increases in ascorbic and uric acid. Doses are given in mg/kg. Sal = saline. In Experiment 5 the dose of amphetamine was 1.5 mg/kg, while in Experiment 6 the dose of amphetamine was 2.0 mg/kg. In Experiment 5 (top), MK-801 was administered at time "0," while amphetamine was given 48 min later. In Experiment 6 (bottom), MK-801 was given at time "0," scopolamine 36 min later, and finally amphetamine, 48 min following MK-801 administration.

ergic and glutamatergic systems may regulate the same pool of AA. MK-801 may indirectly block release of acetylcholine by preventing glutamate from stimulating cholinergic interneurons.

However, the dramatic reduction (89%) of amphetamine-induced increases in AA by pretreatment with haloperidol plus scopolamine is not easily explained by the above model. One possible explanation is that dopaminergic and cholinergic systems may interact to control release of AA. For example, recent investigations have demonstrated that stimulation of D_1 receptors (in the frontal cortex) was necessary for amphetamine-induced increases in release of acetylcholine (6). Thus, amphetamine may indirectly increase release of acetylcholine and AA by the following mechanism: stimulation of cortical D_1 receptors causes corticostriatal neurons to release excitatory amino acids (EAAs); these EAAs increase release of acetylcholine; stimulation of cholinergic receptors then increases release of AA.

On the other hand, the glutamate uptake model (3,23) suggests that the high affinity uptake system for glutamate exchanges glutamate for AA. In support of this hypothesis, infusion of EAAs increase release of AA; glutamate uptake blockers (e.g., homocysteic acid) eliminate the baseline ascorbate signal; glutamate uptake blockers prevent the glutamate-induced increases in release of AA. The acetylcholine-model proposed above cannot account for the effects of the glutamate uptake blockers.

Note, however, that the two models rely on data obtained from different areas of the brain. The acetylcholine model describes release of AA in caudate nucleus; the glutamate up-

take model describes release of AA in thalamus and hippocampus. Release of AA may be controlled by different mechanisms in these areas of the brain.

Furthermore, the two models describe different facets of the release of AA. The acetylcholine model describes pharmacologically evoked release of AA; the glutamate uptake model describes basal release of AA. Different mechanisms may control "resting" levels of AA and evoked release of AA. Thus, the two models are not necessarily incompatible.

In summary, much of the research concerning AA release in the caudate nucleus has focused on three neurotransmitter systems: dopaminergic, cholinergic, and glutamatergic. The results of our studies suggest that a particular neuronal system, specifically the cholinergic system, is primarily responsible for the evoked release of AA in the caudate nucleus.

Uric Acid

In general, the amphetamine-induced increases in UA were less than in the increases in AA. In support of previous research, amphetamine increased release of UA, while low doses of haloperidol reduced amphetamine-induced increases in UA (19).

Interestingly, scopolamine blocked the effect of haloperidol on amphetamine-induced increases in UA. Thus, scopolamine modulated haloperidol-induced reductions of amphetamine-induced increases in release of UA. Therefore, our data demonstrate that cholinergic and dopaminergic systems may interact to control release of UA from the same pool. In support of this hypothesis, the effects of amphetamine are identical to the effects of the combination of pilocarpine plus amphetamine on release of UA (20). In addition, pilocarpine seems to potentiate the effects of amphetamine on release of UA (20).

Thus, the UA data are not consistent with the hypothesis that extracellular levels of UA provide an index of purinergic activity. Rather, cholinergic and dopaminergic systems may regulate the release of UA.

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REFERENCES

1. Basse-Tomusk, A.; Rebec, G. V. Corticostriatal and thalamic regulation of amphetamine-induced ascorbate release in the neostriatum. Pharmacol. Biochem. Behav. 35:55–60; 1990.
2. Bigelow, J.; Brown, D.; Wightman, R. Gamma-aminobutyric acid stimulates the release of endogenous ascorbic acid from rat striatal tissue. J. Neurosci. 42:412–419; 1984.
3. Cammack, J.; Ghasemzadeh, B.; Adams, R. N. The pharmacological profile of glutamate-evoked ascorbic acid efflux measured by in vivo electrochemistry. Brain Res. 565:17–22; 1991.
4. Casamenti, F.; Giovannini, M. G.; Pepeu, G. Acetylcholine release in freely moving rats by means of transversal dialysis coupled to an HPLC with electrochemical detection. J. Neurosci. Methods 29:291; 1989.
5. Clemens, J. A.; Phebus, L. A. Brain dialysis in conscious rats confirms in vivo electrochemical evidence that dopaminergic stimulation releases ascorbate. Life Sci. 35:671–678; 1984.
6. Damsma, G.; Robertson, G. S.; Tham, C.; Fibiger, H. C. Dopaminergic regulation of striatal acetylcholine release: Importance of D_1 and *N*-Methyl-D-Aspartate receptors. J. Exp. Pharmacol. Exp. Ther. 259:1064–1072; 1991.
7. DeBoer, P.; Damsma, G.; Fibiger, H. C.; Timmerman, W.; DeVries, J. B.; Westerink, B. H. C. Dopaminergic-cholinergic interactions in the striatum: The critical significance of calcium concentrations in brain microdialysis. Naunyn Schmiedebergs Arch. Pharmacol. 342:528–534; 1990.
8. Fillenz, M.; O'Neill, R. D.; Grunewald, R. A. Changes in extracellular brain ascorbate as an index of excitatory amino acid release. In: Joseph, M. H., ed. Measuring peripheral and central neurotransmitter release during behavior. Chichester: Ellis Horwood; 1986:144–163.
9. Gonon, F.; Buda, R.; Cespuglio, R.; Jouvet, M.; Pujol, J. F. Voltammetry in the striatum of freely moving rats: Detection of catechols and ascorbic acid. Brain Res. 223:69–70; 1981.
10. Grunewald, R.; Fillenz, M. Release of ascorbate from synaptosomal fraction of rat brain. Neurochem. Int. 6:491–500; 1984.
11. Joseph, M. H.; Hodges, H.; Gray, J. A. Lever pressing for food reward and in vivo voltammetry: Evidence for increases in extracellular homovanillic acid, the dopamine metabolite, and uric acid in the rat caudate nucleus. Neuroscience 32:195–201; 1989.
12. Kamata, K.; Wilson, R. L.; Alloway, K. D.; Rebec, G. V. Multiple amphetamine injections reduce the release of ascorbic acid in the neostriatum of the rat. Brain Res. 362:331–338; 1986.
13. Kunko, P. M.; Mueller, K.; Saponjic, R. M. Cholinergic and glutamatergic modulation of amphetamine-induced increases in ascorbic acid in the rat caudate nucleus. Soc. Neurosci. Abstr. 17:271; 1991.
14. Kuo, C.; Hata, H.; Yoshida, A.; Yamatodani, A.; Wada, H. Effect of ascorbic acid on release of acetylcholine from synaptic vesicles prepared from different species of animals and release of noradrenaline from synaptic vesicles of rat brain. Life Sci. 24: 911–915; 1979.
15. Mandel, R. J.; Nilsson, E.; Rosengren, E.; Bjorklund, A. Amphetamine-induced increases in striatal acetylcholine release as measured by microdialysis are not dependent upon nigrostriatal neurons. Soc. Neurosci. Abstr. 16:1306; 1990.
16. Mefford, I. N.; Oke, A. F.; Adams, R. N. Regional distribution of ascorbate in human brain. Brain Res. 212:223–226; 1981.
17. Mueller, K. In vivo voltammetric recording with nafion-coated carbon paste electrodes: Additional evidence that ascorbic acid release is monitored. Pharmacol. Biochem. Behav. 25:325–328; 1986.
18. Mueller, K. Voltammetric evidence in vivo of cholinergic modulation of extracellular ascorbic acid and uric acid in rat striatum. Brain Res. 408:313–316; 1987.
19. Mueller, K.; Haskett, C. Effects of haloperidol on amphetamine-induced increases in ascorbic and uric acid as determined by voltammetry in vivo. Pharmacol. Biochem. Behav. 27:231–234; 1987.
20. Mueller, K.; Kunko, P. M. The effects of amphetamine and pilocarpine on the release of ascorbic acid and uric acid in several rat brain areas. Pharmacol. Biochem. Behav. 35:871–876; 1990.
21. Mueller, K.; Palmour, R.; Andrews, C. D.; Knott, P. J. In vivo voltammetric evidence of production of uric acid by rat caudate. Brain Res. 335:231–235; 1985.
22. O'Neill, R. D. Adenosine modulation of striatal neurotransmitter release monitored in vivo using voltammetry. Neurosci. Lett. 63: 11–16; 1986.
23. O'Neill, R. D.; Fillenz, M.; Sundstrom, L.; Rawlins, J. Voltammetrically monitored brain ascorbate as an index of excitatory amino acid release in the unrestrained rat. Neuroscience 52:227–233; 1984.
24. Pellegrino, L. J.; Pellegrino, A. S.; Cushman, A. J. A stereotaxic atlas of the rat brain. New York: Plenum; 1979.
25. Phebus, L. A.; Rousch, M. E.; Clemens, J. A. Effect of direct and indirect dopamine agonists on brain extracellular ascorbate levels in the striatum and nucleus accumbens of awake rats. Life Sci. 47:1317–1323; 1990.

26. Pierce, R. C.; Rebec, G. V. Dopamine, NMDA and Sigma receptor antagonists exert differential effects on basal and amphetamine-induced changes in neostriatal ascorbate and dopac in awake, behaving rats. Brain Res. 579:59–66; 1992.
27. Ribeiro, J. Purinergic modulation of neurotransmitter release. J. Theor. Biol. 80:259–270; 1979.
28. Salamone, J. D.; Hamby, L. S.; Neill, D. B.; Justice, J. B. Extracellular ascorbic acid increases in striatum following systemic amphetamine. Pharmacol. Biochem. Behav. 20:609–612; 1984.
29. Schenk, J. O.; Miller, E.; Gaddis, R.; Adams, R. N. Homeostatic control of ascorbate concentrations in CNS extracellular fluid. Brain Res. 253:353–356; 1982.
30. Schultz, D. W.; Lewis, M. H.; Petitto, J.; Mailman, R. B. Ascorbic acid decreases [H] dopamine binding in striatum without inhibiting dopamine sensitive adenylate cyclase. Neurochem. Int. 6:117–122; 1984.
31. Spector, R.; Lorenzo, A. V. Specificity of ascorbic acid transport system of the central nervous system. Am. J. Physiol. 226:1468–1473; 1974.
32. Stone, T. Purinergic transmission in the CNS. Trends Pharmacol. Sci. 1:273–275; 1980.
33. Todd, R. D.; Bauer, P. A. Ascorbate modulates binding to central 5-HT-3 sites in bovine frontal cortex. J. Neurochem. 50: 1505–1512; 1988.
34. Vitler, R. Pharmacology. In: Sebrell, W., ed. The vitamins. New York: Academic Press; 1967:487.
35. Wilson, R. L.; Wightman, R. M. Systemic and nigral application of amphetamine both cause an increase in extracellular concentrations of ascorbate in the caudate nucleus of the rat. Brain Res. 339:219–226; 1985.
36. Wilson, R. L.; Kamata, K.; Wightman, R. M.; Rebec, G. V. Unilateral infusions of amphetamine produce differential, bilateral changes in unit activity and extracellular levels of ascorbate in the neostriatum of the rat. Brain Res. 384:342–347; 1986.