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HALLUCINOGENS POTENTIATE RESPONSES TO SEROTONIN AND NOREPINEPHRINE IN THE FACIAL MOTOR NUCLEUS

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Summary

The present investigation was designed to determine the effect of hallucinogens on the facilitating action of serotonin (5-HT) and norepinephrine (NE) in the facial nucleus. Intravenous administration of d-lysergic acid diethylamide (LSD, 5-10 µg/kg), mescaline (0.5-1.0 mg/kg), or psilocin (0.5-1.0 mg/kg) had no effect by themselves on the glutamate-induced excitation of facial motoneurons. In contrast, the facilitation of facial neuron excitation by iontophoretically applied 5-HT and NE was enhanced 6-10 fold by these hallucinogens. The LSD-enhanced responses to 5-HT and NE continued for at least 4 hours after administration of the hallucinogen. Iontophoretic application of LSD or mescaline (low currents) also markedly potentiated the facilitating effect of 5-HT and NE. Higher currents of LSD (15-40 nA) temporarily antagonized the response to 5-HT. The nonhallucinogen ergot derivatives lisuride and methysergide failed to potentiate the facilitating effects of 5-HT or NE. These observations suggest that hallucinogens potentiate the effect of monoamines on facial motoneurons by increasing the sensitivity of 5-HT and NE receptors. A novel mechanism regarding the psychedelic effects of hallucinogens is discussed.

The facial motor nucleus receives a dense and uniform input from serotonin (5-HT)-containing neurons as well as a lesser norepinephrine (NE) input (1). Recently, it has been found that the excitation of facial motoneurons by various means (e.g., stimulation of the motor cortex, iontophoresis of glutamate) is markedly facilitated by 5-HT and NE (1). As these monoamines do not directly excite facial motoneurons (1), it would appear that they act in a modulatory fashion to enhance the excitability of motoneurons. The hallucinogens d-lysergic acid diethylamide (LSD) and mescaline, as well as 5-HT precursors, have been reported to facilitate certain spinal reflexes (2,3,4). The facilitating effects of these agents on spinal reflexes can be blocked by 5-HT antagonists suggesting that LSD and mescaline may be acting via 5-HT receptors (2,3). In addition, several authors have reported that LSD produces a behavioral motor syndrome in the rat, presumably by stimulating 5-HT receptors in motor nuclei (5,6). Other workers, however, have reported that LSD antagonizes this 5-HT-mediated behavioral syndrome (7) and blocks the excitatory effects of iontophoretically applied 5-HT in the reticular formation and

the cerebral cortex (8,9,10). In the present study low doses of LSD, mescaline, and related hallucinogens were found to markedly enhance the facilitating effects of 5-HT and NE on facial motoneuron excitation.

Methods

Forty-five male albino rats (Charles River) weighing 200-300 g were used. The animals were anesthetized with chloral hydrate (400 mg/kg, intraperitoneally) and mounted in a stereotaxic instrument. Additional doses of chloral hydrate were given as needed throughout the course of the experiment. A burr hole was drilled in the skull over the approximate coordinates of the facial nucleus (2.3 mm posterior to lambda, 2.0 mm lateral to the midline and 8.0-8.5 mm ventral to the surface of the brain). As an aid in locating the nucleus the zygomatic muscle branch of the ipsilateral facial nerve was stimulated to evoke antidromic field potentials. Since facial motoneurons are normally quiescent in the anesthetized rat, iontophoretically applied glutamate was used to locate individual facial motoneurons. For histological localization, the recording sites were marked at the end of each experiment by iontophoretically ejected dye.

Procedures for extracellular unit recording and microiontophoresis were as described previously (1). Five-barrel micropipettes (preloaded with fiberglass filaments) were broken to a tip size of 5-10 μm. The recording (central) barrels were filled with 2M NaCl saturated with Fast Green dye; impedances were 2.5-4.0 MΩ. One side barrel was filled with 4M NaCl for automatic current balancing (11). The remaining barrels were filled with various combinations of the following drug solutions: serotonin creatinine sulfate, 0.04M, pH 4.0 (Regis); l-norepinephrine bitartrate, 0.1M, pH 4.0 (Regis); L-glutamate, 1.0M, pH 8.0 (Sigma); LSD bitartrate, 0.001M in 0.1M NaCl, pH 4.0 (Natl. Inst. Drug Abuse); and mescaline HCl 0.1M, pH 4.0 (Aldrich). A retaining current of 10 nA was applied to all drug barrels between periods of ejection. Previous studies have demonstrated that 5-HT and NE facilitate the excitation of facial motoneurons produced by iontophoretically applied glutamate as well as synaptically mediated excitation (1). Therefore, the sensitivity of facial motoneurons to 5-HT and NE in the presence and absence of hallucinogens was tested during the concurrent ejection of glutamate. In order to standardize the response to 5-HT and NE, 60% of the glutamate current required to activate motoneurons was used to provide a subthreshold excitation to these cells. Only those motoneurons whose threshold for glutamate-induced activation remained constant were selected for further study. In experiments involving the intravenous administration of hallucinogens, only one neuron was studied per animal.

Results

Intravenous administration of LSD (10-100 μg/kg, n=8 animals) had no effect by itself on the glutamate-induced excitation of facial motoneurons. In contrast, the facilitation of facial neuron excitation by iontophoretically applied 5-HT and NE was enhanced approximately 10-fold by low doses (5-10 μg/kg, i.v.) of LSD (n=8 animals, Figure 1A, Table I). The LSD-enhancement of responses to 5-HT and NE continued for at least 4 hours (Figure 1A), but returned to near control 24 hours after administration of the hallucinogen. The enhancement by LSD of responses to iontophoretically applied 5-HT and NE was also seen in two rats pretreated with the 5-HT synthesis inhibitor p-chlorophenylalanine (PCPA, 400 mg/kg, i.p., 24 hours before experiment). Intravenously administered LSD also potentiated the effect of iontophoretically applied 5-methoxy-N,N-dimethyltryptamine (5-MeODMT, a 5-HT agonist) on facial motoneurons (n=4 animals). The iontophoresis of LSD at low currents (2-7 nA), which by itself had no effect on glutamate-induced excitation of moto-

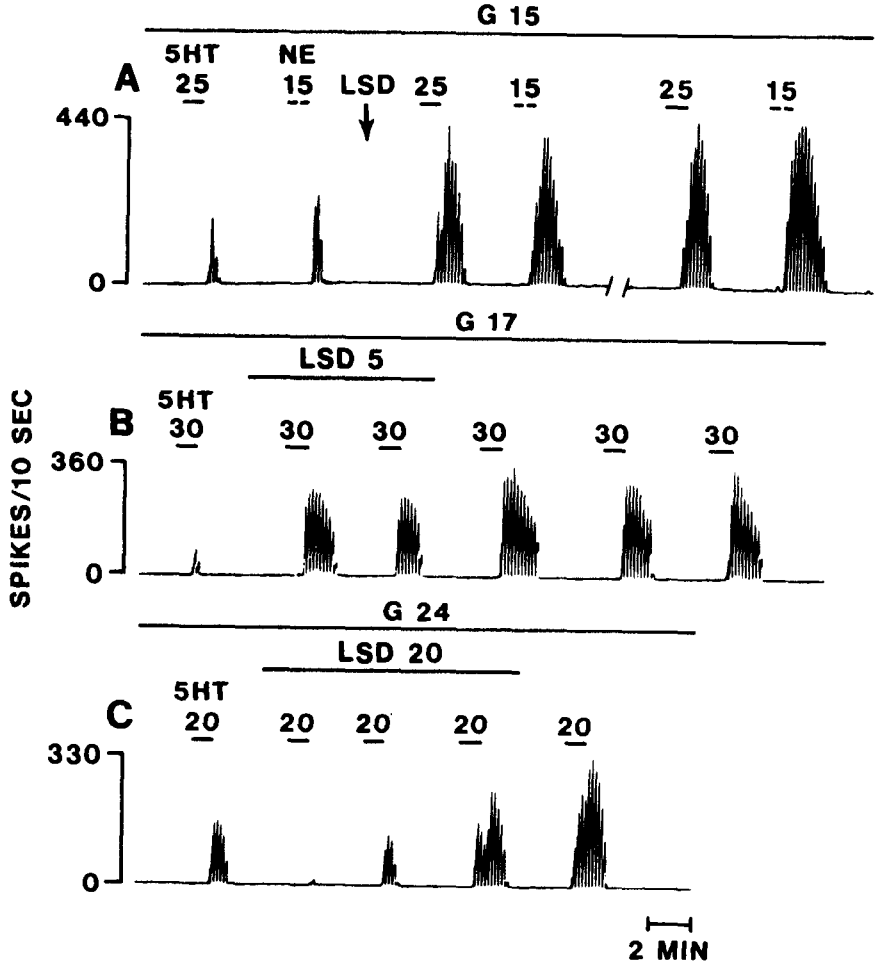

FIG. 1

LSD potentiates the facilitating effect of iontophoretically applied 5-HT and NE on glutamate (G)-induced facial motoneuron excitation. A. Intravenous LSD (10 μg/kg) immediately enhances the response to 5-HT and NE, an effect lasting at least 4 hours (slashed baseline). B. Iontophoretically applied LSD (5 nA) also enhances the effect of 5-HT. C. Higher ejecting currents of LSD (20 nA) temporarily depress the response to 5-HT.

neurons, also enhanced the facilitation of facial motoneuron excitation by 5-HT in 80% of the neurons tested (Figure 1B, n=10 cells). Higher currents of LSD (15-40 nA) temporarily antagonized the response to 5-HT (Figure 1C, n=15 cells). However, if the iontophoresis of LSD was maintained for a longer period of time an enhanced response to 5-HT developed in 73% of the neurons tested (Figure 1C). The enhanced response to 5-HT and NE following iontophoretically applied LSD lasted for at least 2 hours. In the absence of LSD, the response of motoneurons to repeated ejections of 5-HT and/or NE remained constant (Figure 2).

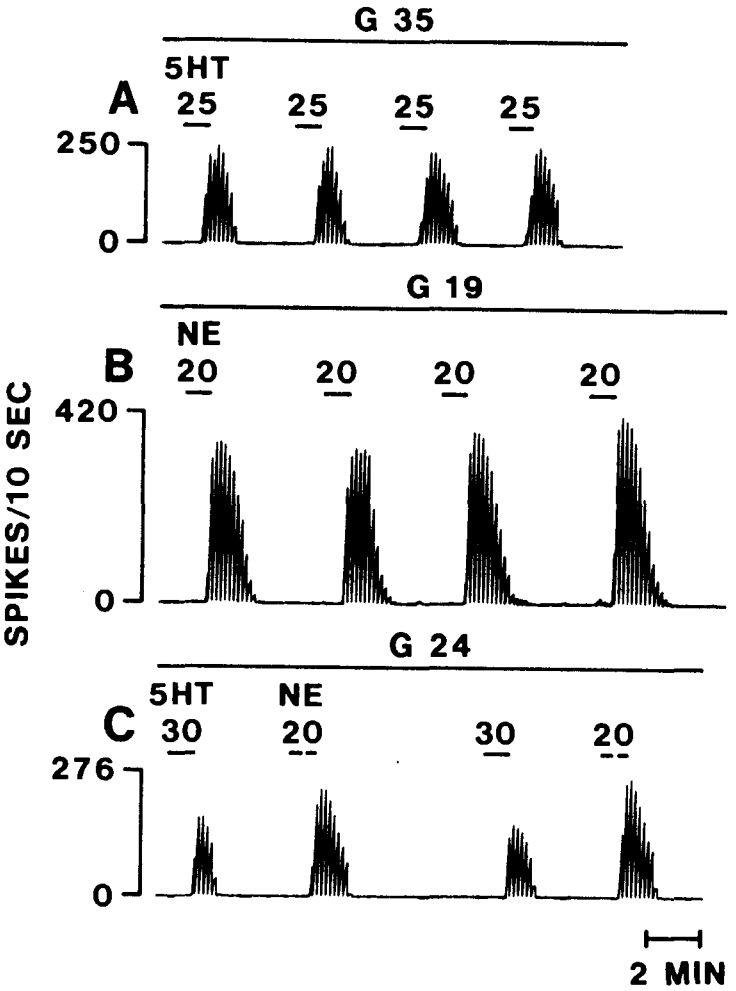

FIG. 2

The response of motoneurons to repeated iontophoretic ejections of 5-HT (A), NE (B), and a combination of the two amines (C).

The intravenous administration of low doses of mescaline (0.5-1.0 mg/kg) also potentiated the effect of 5-HT and NE (n=6 animals, Figure 2A, Table I), but did not alter the glutamate-induced excitation of facial motoneurons by itself. In higher doses (6-10 mg/kg, i.v.) mescaline facilitated glutamate-induced excitation of facial motoneurons even without the addition of 5-HT (n=5 animals). This effect was blocked by the 5-HT antagonist metergoline (100 µg/kg, i.v.) (12), indicating that mescaline at high doses can act as a 5-HT agonist in the facial nucleus (Figure 2B). In rats pretreated with metergoline, iontophoretically applied mescaline potentiated the facilitating effect of NE despite the fact that responses to 5-HT were blocked (Figure 2C, n=14 cells). Taken together, these observations indicate that low doses of LSD and mescaline, in the absence of an effect of their own, markedly increase sensitivity to both 5-HT and NE in the facial nucleus.

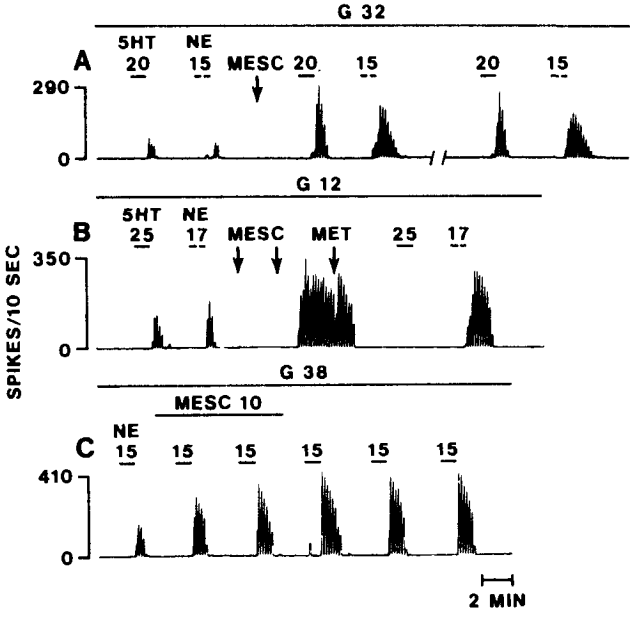

FIG. 3

Mescaline potentiates the facilitating effect of iontophoretically applied 5-HT and NE on facial motoneurons. A. A low dose of mescaline (1 mg/kg, i.v.) enhances the effects of 5-HT and NE without directly activating the cell. The action of mescaline lasts at least 2 hours. B. Metergoline (100 µg/kg, i.v.) blocks the enhancement of glutamate excitation which is produced by higher doses of mescaline (6 mg/kg, i.v.); metergoline also blocks 5-HT but the response to NE is enhanced. C. Iontophoretically applied mescaline (10 nA) potentiates the facilitating effect of NE on facial motoneurons.

To determine if the sensitizing action of LSD and mescaline on facial motoneurons is specific for hallucinogens, we studied the effects of two simple indoleamine hallucinogens, psilocin (13) and N,N-dimethyltryptamine (DMT) (14), and two nonhallucinogenic ergot derivates, lisuride (15) and methysergide. As in the case of LSD, psilocin (0.5-2.0 mg/kg, i.v.) markedly potentiated the effect of 5-HT and NE on motoneurons, but had no direct action when given alone (Table 1). In contrast, DMT (0.5-2.0 mg/kg, i.v.) facilitated the glutamate-induced excitation of motoneurons by itself; this effect was blocked by metergoline. DMT was unlike mescaline in that a 5-HT sensitizing action could not be differentiated even at low doses from the 5-HT agonist activity. Nevertheless, in animals pretreated with metergoline, DMT (like mescaline) potentiated the facilitating effect of NE (Table I). The nonhallucinogen lisuride (10-40 µg/kg) had no effect on glutamate-induced excitation of motoneurons or on the facilitating action of 5-HT and NE (Table I). Finally, the peripheral 5-HT antagonist methysergide antagonized the action of 5-HT and failed to potentiate the effect of NE on facial motoneurons (Table 1).

TABLE I

The Influence of Intravenous Hallucinogens and Nonhallucinogens on the Facilitating Effect of 5-HT and NE on Facial Motoneurons

Drug	# Animals[1]	Dose	% Control 5-HT	% Control NE
LSD	8	10 µg/kg	1026±93*	976±109*
mescaline	6	1 mg/kg	763±120*	913±168*
psilocin	5	1 mg/kg	497±46*	491±72*
DMT[2]	5	1 mg/kg	-	469±48*
lisuride	5	40 µg/kg	102±4	102±2
methysergide	5	1 mg/kg	0	98±6

*P <.01, Student's t-test.
[1]One neuron was tested per animal.
[2]Animals were pretreated with metergoline (100 µg/kg, i.v.)

Discussion

The present study indicates that LSD (an indoleamine) and mescaline (a phenethylamine) potentiate the facilitating effect of both 5-HT and NE on facial motoneuron excitation. The similarity between the effects of these two hallucinogens on motoneurons supports the contention that these indoleamine and phenethylamine derivatives are structurally similar in certain respects (16,17,18). This contrasts with the action of these agents on serotonergic neurons in the dorsal raphe, where LSD acts as a 5-HT agonist and mescaline has no direct effect (19). In the facial motor nucleus the effects of these drugs did not result from a 5-HT agonist property since LSD and mescaline (in a low dose) did not facilitate the glutamate-induced excitation of motoneurons in the absence of 5-HT and NE. These observations suggest that LSD did not potentiate the actions of 5-HT and NE via an interaction with glutamate. However, the possibility exists that the hallucinogens decreased the threshold for glutamate-induced excitation of motoneurons. In addition, it appears unlikely that the hallucinogens produced their effects by reuptake blockade since LSD enhanced the facilitating effect of 5-MeODMT (a 5-HT agonist which is not readily taken up by 5-HT terminals) (20). Finally, it is unlikely that LSD potentiated the effect of 5-HT and NE by causing the release of endogenous 5-HT since the LSD-enhanced response was observed in animals pretreated with PCPA. In contrast, PCPA pretreatment has been shown to block the facilitating effect of the 5-HT releasing agent p-chloroamphetamine on facial motoneurons (1). It appears that the ability to potentiate the effects of 5-HT and NE in the facial nucleus may be common to all of the psychedelic hallucinogens since psilocin and DMT were found to act in a similar manner to LSD and mescaline. The observation that two nonhallucinogenic ergot derivatives, lisuride and methysergide, did not enhance the facilitating effect of 5-HT and NE suggests that the ability to potentiate is specific to the hallucinogens.

The hallucinogens appear to potentiate the effects of monoamines on facial motoneurons by increasing the sensitivity of 5-HT and NE receptors. A precedent for such a sensitizing action can be found in the observation that low doses of LSD and mescaline facilitate uterine contractions produced by 5-HT (21). A further parallel with our present observations is the fact that high doses of LSD antagonize the 5-HT induced uterine contractions, while in high doses mescaline acts as a 5-HT agonist (21). The mechanism by which hallucinogens could sensitize 5-HT and NE receptors is not known. However, the fact that LSD and 5-HT appear to bind to different 5-HT receptor sites (22) suggests the possibility that drug-induced changes in receptor sensi-

tivity could occur, in part, through interactions between 5-HT and LSD binding sites.

Lose doses of LSD and mescaline have been shown to enhance certain spinal reflexes (2,3,4). In addition, 5-HT and NE facilitate the excitation of spinal motoneurons (23). Taken together with the present data, these observations suggest that a sensitization of 5-HT and NE receptors by LSD and mescaline could account for the enhancement of spinal reflexes by these hallucinogens. In contrast to the facilitating effects of LSD on spinal reflexes, LSD appears to antagonize the excitatory effects of 5-HT on certain brain stem and cortical neurons (8,9,10). Based on the facial nucleus studies, we have suggested that the apparent "excitations" induced by 5-HT in the latter areas may actually represent a facilitation of tonic excitatory inputs (1). In the present investigation, LSD applied at high iontophoretic currents temporarily antagonized the facilitating effect of 5-HT. In contrast, LSD did not alter the glutamate-induced discharge of facial motoneurons. This suggests that large amounts of LSD may antagonize and/or nonspecifically depress the facilitating actions of 5-HT.

If the sensitizing effects of low doses of hallucinogenic drugs observed in the facial nucleus occurs in nonmotor areas of the central nervous system, then a mechanism of receptor sensitization might contribute to the psychedelic effects of these drugs. In this regard, the time course of the LSD-enhanced effect of 5-HT and NE parallels certain prolonged behavioral effects of this hallucinogen in cats (24). This raises the interesting possibility that such long-term changes in noradrenergic and serotonergic systems may be involved in the psychedelic actions of the hallucinogens.

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