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The Effect of LSD, Mescaline, and D-Amphetamine on the Evoked "Secondary Discharge"*

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The relevance of animal studies to the investigation of hallucinogenic drugs is always questionable because the specific response to them is a human mental state. Any argument that a neurological or behavioral effect in animals is pertinent to their hallucinogenic properties must be that the effect is observed solely with drugs of this class and not with related ones, with which they may have non-specific actions in common, such as vasopression. Furthermore, the analogy will be stronger if the effective doses of different drugs in the sub-human experimental system are proportional to their relative human potency, in ranges not hundreds of times higher than the human ones, and if their time course of action is comparable. A system that responded in the analogous way outlined might be used to predict the properties of new compounds or to suggest a neurological hallucinogenic mechanism.

The Forbes secondary discharge was noted by PURPURA (1956) to be sensitive to as little as 10 µg/kg LSD in cats under moderate barbiturate anesthesia. This was in accord with his theory that non-specific pathways with axodendritic synapses are more easily inhibited by LSD than specific pathways with axosomatic synapses. Although the unanesthetized animal might seem to be the most informative subject, the effect of LSD-25 in such experiments is quite variable, depending partly on slight noises or movements in the environment (BRADLEY and ELKES, 1957). The purpose of the experiments described below was to quantitate the analysis of this evoked response and to compare the effects of different doses of LSD, mescaline, and amphetamine on it.

Methods

Adult cats were anesthetized by intraperitoneal injections of Dial with Urethane (CIBA). After introducing a tracheal cannula, a catheter was placed in the right femoral vein for injection of further anesthetic and the test drugs. The right femoral artery was cannulated for the

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monitoring of blood pressure. A mechanical transducer pressing against the rib cage served to record respiratory rate, and give a rough estimate of its depth.

The left sigmoid and anterior lateral gyri were exposed to allow recording directly from the cortex. Amplifier input came from an active nichrome wire electrode on the cortex and an indifferent electrode attached to the bone overlying the frontal sinus. Amplifier output led to an oscilloscope whose sweeps were triggered by the stimuli. A Grass camera recorded each evoked response. Respiration, blood pressure, and the EEG were simultaneously recorded on a polygraph.

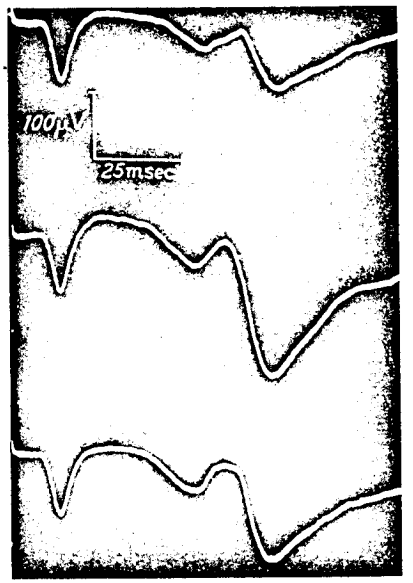

Fig. 1. Film tracings of successive evoked responses. Negativity is upwards

Monophasic pulses were delivered to the right sciatic nerve with a frequency of 0.2/sec, a duration of 0.5 msec, and a voltage 0.1 v. above that necessary to evoke a maximum secondary discharge. Anesthesia was deepened, and the position of the cortical electrode was varied along the lateral anterior gyrus until optimum recordings were obtained. Often both a primary evoked response and a secondary discharge were present, the latter being identified by its latency of 40 to 100 msec, its duration of the same range, and its inhibition at frequencies of stimulation greater than 0.5/sec. Each stimulation and recording period lasted 1 to $1^1/_2$ min. Between these periods no stimuli were delivered. Three or more control stimulation periods were recorded over a 20 min period prior to drug administration, Then a dose of LSD-25 (Sandoz), psilocybin (Sandoz), mescaline hemi-sulfate (Sigma) or d-amphetamine was injected intravenously. After the drug was given, stimulation periods were recorded at approximately $+ {}^1/_2$, 2, and 5 min, and then every 5 or 10 min until the end of the experiment.

Fig. 1 shows part of a typical film tracing, Here the primary evoked response has a latency of about 12 msec and the secondary discharge (or secondary response) a latency of about 70 msec. Because the amplitude of successive secondary responses is quite variable, it is necessary to average them. The film was projected and the secondary discharge was

measured with a ruler from the peak of the initial small negative wave to the large positive peak. The primary evoked response was measured from the initial positive to the following negative peak.

Results

The primary evoked response was not visible in all the tracings and was not systematically studied. In four experiments with LSD in which the amplitude of both primary and secondary responses were measured,

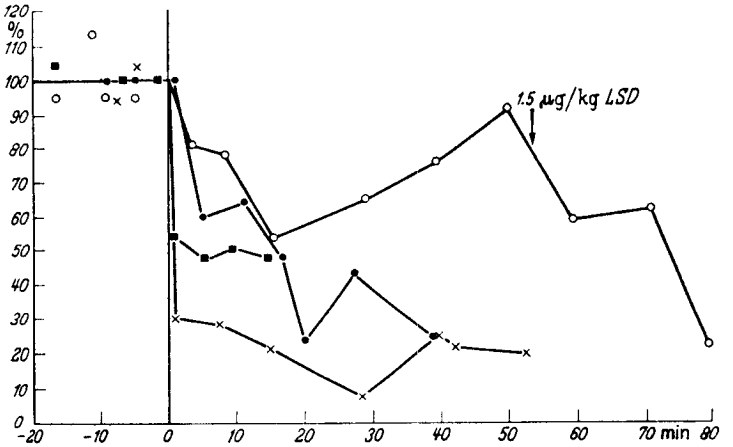

Fig. 2. Time course of drug effects on the average secondary response. The ordinate is the percentage of the pre-drug values, the average of the responses between —20 and 0 min having been set equal to 100%. The abscissa is the time in minutes with t = 0 the time of drug injection. LSD-25 1µg/kg. o 10µg/kg. • D-amphetamine 0.5 mg/kg. ■. Mescaline 10mg/kg × An additional 1.5µg/kg of LSD was injected at the point marked by the arrow

no change in the response occurred although the secondary was inhibited. In one of these, 20µg/kg LSD had no effect on the amplitude of the primary response.

Fig. 2 illustrates the time course of the drug effects on the secondary discharge. The average of all the control period secondary responses was set equal to 100% and the average amplitude of the responses during each stimulation period was calculated and expressed as a percentage of the control period average. Thus, each point on the graph represents an average of 10 to 18 responses. Time = 0 is the first administration of an active compound to a separate animal in each experiment. The immediate inhibition produced by amphetamine and mescaline is in contrast to a 10 to 20 min onset for LSD. Only four experiments are plotted here, but this contrast applied to all the other experiments where inhibition was obtained. There seems to be recovery from the inhibition after 50 min with the 1µg dose of LSD which was reversed by the addition of another 1.5µg/kg. This was seen in one other experiment, a single experiment with psilocybin in which 70µg/kg, which is equivalent to 1µg/kg of LSD in humans (Wolbach *et al.*, 1962b), produced a significant inhibition which

Table. *The mean ± S. D. of the secondary response in three time periods. The significance of A vs. B is indicated after the numbers in column B; that of A + B vs. C in column C. See text for further explanation*

	A −21 to −15 min	B −5 to 0 min	C +10 to +16 min	C/A
Normal Saline				
5 ml	25 ± 15	25 ± 14 NS	26 ± 17 NS	1.04
5 ml	46 ± 9	41 ± 26 NS	46 ± 23 NS	1.00
LSD				
1 μg	35 ± 18	35 ± 11 NS	20 ± 17[2]	0.57
2 μg		43 ± 13	35 ± 25 NS	0.81
2 μg	45 ± 33	42 ± 29 NS	28 ± 17 NS	0.62
5 μg	60 ± 23	58 ± 25 NS	10 ± 10[3]	0.17
10 μg		52 ± 17	32 ± 20[2]	0.63
20 μg	54 ± 12	53 ± 21 NS	18 ± 15[3]	0.34
Mescaline				
5 mg		52 ± 9	37 ± 15[2]	0.71
10 mg		84 ± 6	17 ± 19[3]	0.20
15 mg	33 ± 9	44 ± 14[1]	35 ± 9 NS	1.06
20 mg	33 ± 25	36 ± 18 NS	4 ± 3[3]	0.12
25 mg	26 ± 16	22 ± 14 NS	6 ± 7[3]	0.23
Amphetamine				
0.5 mg	28 ± 10	27 ± 16	13 ± 8[1]	0.46
3.0 mg		34 ± 12	7 ± 7[3]	0.49

NS = not significant, [1] $p < .05$, [2] $p < .01$, [3] $p < .001$.
For each time period the mean ± the standard deviation is given.

partially remitted after 50 min. In that same cat later doses of 70μg/kg and 280 μg/kg of psilocybin both appeared to cause transient deepening of inhibition followed by partial recovery.

The Table presents a comparison of the mean secondary responses during the three stimulation periods in each experiment which fall in the period 15 to 21 min before the drug was given (A), in the five minute period before administration (B), and from 10 to 16 min after administration. Each mean is an average of about 10 responses, so that even though only three out of the six to twenty stimulation periods in each experiment are being compared, a statistical analysis is possible. Each row of the Table refers to the first administration of the active drug to a separate animal. All experiments with these drugs are listed.

Using the F test, the significance of A vs. B was calculated to give an indication of the stability of the responses in the control period, and is indicated after the means in column B. The significance of A + B vs C is a test of the drug effect and is listed in column C. In some experiments there was no stimulation period during A, and so the means in B and C are compared with the t test.

The secondary response is quite stable in spite of the large standard deviation of each sample. Only the experiment with 15mg mescaline

showed a significant difference between A and B. Normal saline has no effect on the base line. In general the three active drugs all produced significant inhibition of the secondary discharge. Twice 2 μg/kg of LSD and once 15 mg of mescaline did not produce significant change.

The ratio of C to A gives an estimate of the degree of inhibition, though it is liable to error in the case of LSD, for which the period C mean is too early for full inhibition to have taken place in some instances. No definite trend in this ratio is present, precluding establishment of dose-response curves from these data. This is probably largely a reflection of the small number of animals and individual differences in response.

The role of blood pressure was examined by comparing the change in blood pressure from the control period to 15 min after drug injection. Amphetamine and mescaline produce rises of 30—50 mm Hg systolic with the doses used in these experiments. On the other hand, LSD caused inhibition of the secondary discharge in one experiment where the blood pressure remained constant (1 μg/kg LSD), and in one experiment where the blood pressure fell (10 μg/kg with a 15 mm fall).

Respiratory rate and depth remained constant with the exception of two of the experiments with mescaline and the two with amphetamine, in which respiration became more shallow for a one to two minute period after drug injection.

Discussion

Because of the variability of the secondary discharges, averages of a number of responses should be used. This allows the detection of effects from a dose of LSD one-tenth the amount reported in PURPURA's experiment (PURPURA, 1956).

FORBES et al. (1949) have shown that changes in the depth of anesthesia can alter the evoked secondary response. At most levels, a decrease in anesthesia will decrease the evoked response average. In some reported experiments, the effects of LSD and barbiturates are antagonistic, so in several LSD trials, after the inhibition was at a maximum, more anesthetic was added. No reversal of inhibition was ever observed, however.

Possibly a rise in blood pressure alone would have an effect similar to lightening the anesthesia, since blood pressure rises in themselves will produce cortical arousal (BAUST *et al.*, 1963). The actions of mescaline and amphetamine might be explained by their vasopressor effects, but the action of LSD could not, because inhibition occurred using this drug without a rise in blood pressure.

If the inhibition of the secondary discharge were related to the hallucinogenic properties of LSD and mescaline, amphetamine would not be expected to produce inhibition. The psychological effects of the hallucinogens are quite distinct from those of amphetamine. Also LSD is cross tolerant with psilocybin and mescaline in human experiments (ISBELL *et al.*, 1961; and WOLBACH *et al.*, 1962a), but not with amphetamine

(ROSENBERG *et al.*, 1963), probably reflecting different mechanisms of action for the two types of drugs. The dose of D-amphetamine used by ROSENBERG et al. was 0.6 mg/kg which is comparable to the 0.5 mg/kg used here, but the dose of 3.0 mg/kg used in the second cat is in the range that will sometimes produce psychotic ideation in humans. More data with low doses of amphetamine are necessary to conclude definitely that its inhibition of the evoked secondary discharge is a non-specific effect. Recovery from the 1 μg/kg dose of LSD after 50 min is quite different from the clinical time course of LSD, and may be regarded as another bit of evidence for the non-specificity of this system.

Summary

The effects of various doses of LSD, mescaline, and amphetamine on the secondary discharge of Forbes were investigated in cats anesthetized with a long-acting barbiturate. Peripheral stimulation of the sciatic nerve evoked a secondary discharge, which was recorded using a cortical electrode on the contralateral anterior lateral gyrus. Doses of LSD as low as 1 μg/kg produced a decrease in the average evoked secondary discharge. Mescaline and amphetamine also produced a decrease. An increase in blood pressure could not explain the results with LSD, since inhibition occurred without a rise in blood pressure, but blood pressure rises accompanied the inhibition from the other drugs. These experiments indicate that this physiological system is very sensitive to LSD, but that a non-hallucinogen such as amphetamine can affect it, too.

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