# Psychopharmacology of Hallucinogens

LS

## AN ANIMAL BEHAVIOR MODEL FOR STUDYING THE ACTIONS OF LSD AND RELATED HALLUCINOGENS

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### Introduction

Lysergic acid diethylamide (LSD) and related compounds such as N, N dimethyltryptamine (DMT) and psilocybin are of great interest because of the profound psychological effects they produce in humans, and because of their widespread usage both in this country and cross-culturally. In spite of intense public and scientific interest in these drugs, however, many questions concerning the parameters and physiological bases of their action remain unanswered.

Experiments directed at these issues which employ human subjects are precluded either for moral or ethical reasons, or because they require examination of tissue derived from living organisms. Therefore, we must turn to animal experiments for the answers to most of these questions.

Animal models of human processes take two different forms, only one of which is appropriate for dealing with variables which are primarily psychological in nature, such as hallucinations. In Isomorphic Models, the change in the animal is assumed to be directly analogous, or even homologous, to the human condition. Such models are widely employed in medicine in the study of disease states such as diabetes or cancer. The validity of these models is difficult to establish in the case of psychopathology or changes in consciousness due to the inherent problem of knowing what a non-verbal organism is feeling, thinking, or perceiving. Using such models necessitates confronting what may be an unanswerable question of how one discerns whether an animal is experiencing an hallucination. By contrast, utilization of Parallel Models obviates the need to impute an underlying similarity of state in the animal and human. Such models are founded on the assumption that their validity can be established through demonstrating that changes in the animal directly parallel changes in the human. There is no need for the changes to be either isomorphic or even analogous, but merely that the two covary systematically. Examples of such models include: a) "wet

dog shakes" in rats as an indication of the abstinence syndrome which follows withdrawal from morphine (47), and b) the parallel between the behavior of cats depleted of brain serotonin and actively ill schizophrenic patients (23).

Among the criteria that would have to be met in order to qualify a behavior, or behaviors, as an animal model for the actions of hallucinogens, are the following:

1. Change specifically in response to this class of drugs, and no other.
2. Dose-dependent change in frequency or magnitude.
3. Responsive to dose levels approximately within the human range.
4. Closely parallel the major parameters of the action of these drugs in humans.
5. Observed in most, or all, animals tested (robust).
6. Reproducible across time (reliable).
7. Quantifiable.
8. Easy to score.

We reasoned that the establishment of an animal behavior model for the actions of LSD and structurally and/or functionally related hallucinogens could best be accomplished by close behavioral observation and analysis of animals administered these drugs. Although previous studies have examined the effects of LSD and related hallucinogens on behavior in a variety of species [e.g., mouse and rat (8-11,13,16,20-22,24, 29-31,46,50,56-58,62,63), cat (3,25,28,31,32,36,49,51,53,61), and monkey (26,42,43,54)], few of them have explored a complete dose range or closely examined the behavioral changes. More importantly, none of them have reported an effect which was demonstrated to be specific to these compounds [besides the studies cited above, see reviews by Holister (35) and Siegel (55)]. Accordingly, in our initial study, we administered LSD to cats in a dose range (2.5-200 ug/kg, i.p.) within an order of magnitude of that given to human subjects (0.5-16 ug/kg, p.o.) (2,44). The ensuing spontaneously occurring behavior was then scored in an attempt to compile a complete and detailed record of all major changes, with special attention toward those behaviors that might be evoked specifically by LSD. Four behaviors, which were either non-existent or occurred with a very low frequency in saline treated animals, showed a dose dependent increase in response to LSD, and emerged to the point where they often dominated the behavior of the animals (41). Subsequent experiments demonstrated that this effect was not due to a peripheral action of LSD, that these behaviors were also elicited by a related indole nucleus hallucinogen, and that they were <u>never</u> seen in response to a variety of other psychoactive drugs. Finally, the importance of the model is indicated by its ability to further elucidate some of the parameters of LSD's action such as time course, onset of tolerance, and duration of tolerance. This series of experiments will be reviewed in detail in the present chapter.

## Method

### LSD, Other Hallucinogens, and Control Drugs

Adult female cats weighing 2.0-3.3 kg were used in all experiments. They were individually housed in standard stainless steel cat cages which also served as the experimental observation chamber. Using a counterbalanced design, each animal received all dose levels of LSD, with at least eight days intervening between consecutive sessions. Cats were given intraperitoneal injections of either saline or lysergic acid diethylamide tartrate (2.5, 10.0, 25.0, 50.0, 100.0 or 200.0 ug/kg, dose expressed as the salt). Behavioral observations, by raters who were "blind" to the treatment, were made during the hour immediately following drug administration. The frequency of occurrence of each behavior was tallied on a standard scoring sheet. Many of the behavioral descriptions are self-explanatory, however, a few require further comment. Abortive grooming is scored when the cat orients to the body surface as if to groom but does not emit the consummatory grooming response (bite, lick, or scratch), or emits the response in midair. Limb flicking is a behavior seen in normal cats almost exclusively in response to the presence of a foreign substance, such as water, on the hindpaw or forepaw. The paw is then lifted and rapidly flicked outward from the body. Investigatory or play behavior refers to pawing or sniffing at objects or in corners, chasing the tail, or batting at pieces of food, feces, etc. Hallucinatory-like behavior is scored when the cat looks around at the floor, ceiling, or walls of the cage and appears to be tracking objects visually, or when the cat either hisses at, bats at, or pounces at "objects" not seen by the observer.

In order to control for the peripheral serotonin antagonist effects of LSD, groups of animals were administered either its non-hallucinogenic congener, brom-LSD (25 and 100 ug/kg), which has the same peripheral potency, or methysergide maleate (25, 100, 500 and 2,500 ug/kg), a more potent blocker of serotonin's action in the periphery. The specificity of the behavioral changes observed in response to LSD was examined by administering groups of animals compounds which were representative of other major drug classes: d-amphetamine sulfate (0.25, 1.0 and 5.0 mg/kg), atropine sulfate (0.5 and 2.5 mg/kg), caffeine (1.0, 5.0 and 20.0 mg/kg), chlorpheniramine maleate (0.5, 2.5, and 5.0 mg/kg), and tryptamine (0.05, 0.1 and 5.0 mg/kg), a non-hallucinogenic indole nucleus drug. Finally, in order to examine the generality of the behavioral changes seen in response to LSD, animals were administered psilocybin (25, 100 and 500 ug/kg), an indole nucleus hallucinogen structurally related to LSD, or the structurally unrelated hallucinogen, $\Delta^9$-tetrahydrocannabinol (0.5, 1.0 and 5.0 mg/kg).

### Duration of Behavioral Effects of LSD

Two groups of female cats (N = 7/group) received a single intraperitoneal injection of LSD tartrate at a dose of either

10 or 50 ug/kg (dose expressed as the salt). Behavioral observations by "blind" raters were made during hours 1, 2, 3, 4, 6, 8, and 24 following drug administration. The behavior was scored as described above. Since limb flicking, abortive grooming, play or investigatory behavior, and hallucinatory-like behavior are proposed as the essential components of our model, only the data on these behaviors will be reported.

Duration and Onset of Tolerance to LSD

In order to examine the duration of tolerance, female cats were administered a single dose of LSD tartrate, either 10 or 50 ug/kg, i.p., and then given a test dose of 50 ug/kg of LSD at 1, 3, 5, 7 or 14 days later (separate groups for each time point). Behavioral observations and scoring during the hour following LSD administration were made as described in the previous experiment. In order to study the onset of tolerance to LSD, female cats were administered either a single dose of LSD tartrate (10 ug/kg, i.p.) or saline (1 ml, i.p.), and then given a test dose of 50 ug/kg of LSD at 2, 6 or 24 hours later (separate groups for each time point). Behavioral observations were taken during the 1 hour period immediately following the 50 ug/kg dose of LSD.

## Results

LSD

The behavioral effects of LSD can be divided into three categories. First, those that did not significantly change in frequency in response to LSD: rubbing, treading or kneading, and vocalization. The second group of behaviors manifested a relatively high frequency of occurrence following saline injection and then showed a significant dose dependent increase in response to LSD: staring (other than forward for more than five seconds), grooming, and head or body shakes. Finally, the third group of behaviors were either non-existent or occurred with a very low frequency in saline treated animals, but emerged to become the most prominent behaviors of LSD-treated animals. Since the latter group of behaviors will be proposed as an animal model for the actions of LSD, we will begin by discussing them in some detail. This will be followed by a discussion of the other two categories of behavior.

As shown in Table 1, limb flicking, abortive grooming, investigatory or play behavior, and hallucinatory-like behavior all have a curvilinear relationship to the dose of LSD. In general, these behaviors have a mean hourly frequency at or near zero in saline treated animals, increase in frequency in a dose dependent manner, reaching their peak at 25 or 50 ug/kg LSD, and then decline in response to the administration of 100 and 200 ug/kg doses.

Perhaps the most impressive effect of LSD was the elicitation of the limb flicking response. This is a species-specific

TABLE 1

Behavioral Response to Varying Doses of LSD in the Cat

| | MEAN FREQUENCY OF OCCURRENCE/HR (±S.E.M.) | | | | | | |
|---|---|---|---|---|---|---|---|
| | LSD TARTRATE (UG/KG) | | | | | | |
| BEHAVIOR | 0.0 | 2.5 | 10.0 | 25.0 | 50.0 | 100.0 | 200.0 |
| Limb flick$^{xx}$ | 0.2±0.1 | 1.8±0.5$^{**}$ | 8.0±2.1$^{***}$ | 22.6±4.0$^{***}$ | 45.9±8.1$^{***}$ | 31.5±5.9$^{***}$ | 15.1±4.3$^{***}$ |
| Abortive grooming$^{xx}$ | 0.0±0.0 | 0.6±0.3 | 2.4±1.3$^{*}$ | 5.0±1.5$^{**}$ | 5.6±1.2$^{***}$ | 7.9±3.1$^{**}$ | 2.9±0.8$^{*}$ |
| Investigatory or play$^{x}$ | 0.3±0.1 | 3.4±1.7 | 2.7±1.1$^{*}$ | 8.8±3.9$^{**}$ | 10.8±3.8$^{**}$ | 6.8±2.5$^{**}$ | 1.2±0.6 |
| Hallucinatory-like$^{x}$ | 0.1±0.1 | 0.8±0.5 | 1.5±0.8$^{*}$ | 2.7±1.1$^{**}$ | 1.3±0.6$^{*}$ | 0.5±0.4 | 0.2±0.2 |
| Rubbing | 1.9±1.3 | 1.5±0.8 | 1.1±1.0 | 2.6±2.0 | 2.1±1.6 | 1.7±1.5 | 0.1±0.1 |
| Treading or kneading | 0.0±0.0 | 1.1±0.4 | 0.4±0.3 | 1.2±0.3 | 0.7±0.3 | 0.4±0.2 | 0.7±0.3 |
| Vocalization | 1.2±0.8 | 1.3±1.1 | 1.3±0.9 | 0.6±0.3 | 0.5±0.5 | 0.6±0.5 | 0.7±0.7 |
| Staring$^{xx}$ | 5.1±1.4 | 3.2±1.4 | 8.8±2.3$^{*}$ | 15.4±2.7$^{**}$ | 18.2±3.2$^{**}$ | 24.4±5.2$^{***}$ | 4.5±2.1 |
| Grooming$^{x}$ | 7.4±2.4 | 8.4±2.5 | 18.0±4.2$^{*}$ | 22.6±6.3$^{**}$ | 16.3±2.8$^{*}$ | 11.2±2.5 | 7.2±2.5 |
| Head or body shake$^{xx}$ | 6.4±1.1 | 9.3±2.2 | 10.8±2.7$^{*}$ | 13.2±3.1$^{*}$ | 30.3±6.9$^{***}$ | 13.5±3.4$^{*}$ | 6.7±2.2 |

Levels of significance for one-way analysis of variance: $^{x}p < 0.01$; $^{xx}p < 0.001$
Levels of significance for t-tests comparing each drug dose with the saline control:
$^{*}p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$

behavior normally used exclusively for removing foreign substances from the limbs. In saline treated animals, this response was seen only twice in the 1 hr. observation period on 12 cats ($\bar{X}$ = 0.2/hr). When the same animals were administered LSD, this response significantly increased in frequency in a dose-dependent manner to a peak of 45.9/hr at a dose of 50 ug/kg ($p < 0.001$, ANOVA). This represents a change of several orders of magnitude. After the 200 ug/kg dose, the frequency of flicks significantly declined from its peak of 45.9/hr to a mean of 15.1/hr ($p < 0.001$, t-test). It was not uncommon for a cat given a dose of 25-100 ug/kg of LSD to emit 20-30 flicks in a 15 minute period, and two or three sequential flicks within a few seconds. It is also important to note that every animal tested responded with at least 3 flicks to doses of 25, 50 and 100 ug/kg. This response may also be the most sensitive reflection of the effects of LSD in the cat since it was the only behavior to significantly increase in frequency in response to the lowest dose of 2.5 ug/kg ($p < 0.01$, t-test).

Along with limb flicking the other major emergent behavior which had not been described in previous studies was abortive grooming. This response increased in a significant dose-dependent manner up to 100 ug/kg ($p < 0.001$, ANOVA), and then significantly decreased at 200 ug/kg ($p < 0.01$, t-test). Although the mean frequency of 7.9 bouts of abortive grooming/hr. at the 100 ug/kg dose does not represent a large absolute value, this response was never observed in saline treated animals. As with limb flicks, every animal tested showed at least one instance of abortive grooming in response to high doses of LSD. Emergence of abortive grooming may be reflective of the fragmentary or disjunctive nature of all behaviors in LSD treated animals. These animals would rarely sustain any active behavior for more than several seconds. Their attention seemed to be constantly shifting. As a result, their ongoing behavior was frequently changed, interrupted, or even aborted prior to consummation, as in the case of grooming.

During a one hour observation period, very few cats housed for at least 2 or 3 weeks in a standard cat cage spontaneously emit behavior that would be classified as play or investigatory. However, this dramatically changes when they are administered LSD. As shown in Table 1, the frequency of these responses significantly increased in a dose-dependent manner, reaching their peak of 10.8/hr at 50 ug/kg ($p < 0.01$, ANOVA). This response markedly decreased in frequency ($\bar{X}$ = 1.2/hr), when the dose was increased from 50 to 200 ug/kg ($p < 0.01$, t-test). Unlike limb flicking and abortive grooming, investigatory or play behavior was not observed in every animal tested.

The final emergent response, hallucinatory-like behavior, proved to be the least objective and the most difficult to score. In general, this response was scored when the animal pounced on, batted at, attacked or seemed to be visually tracking something not apparent to the experimenter. Like play, this behavior is virtually non-existent in well habituated,

Levels of significance for one-way analysis of variance: x $p < 0.01$; xx $p < 0.001$
Levels of significance for t-tests comparing each drug dose with the saline control:
*$p < 0.05$; **$p < 0.01$ ***$p < 0.001$

untreated animals. The mean frequency of occurrence of hallucinatory-like behavior significantly increased as a function of dose ($p < 0.01$, ANOVA), reached its peak at 25 ug/kg, and then declined in response to higher doses, ultimately approaching zero at 200 ug/kg. The mean frequency of occurrence was significantly less at 200 than at 50 ug/kg ($p < 0.05$, t-test). This behavior was also emitted with a high frequency in some animals and not at all in others.

Rubbing, treading or kneading, and vocalization do not require discussion since they did not significantly change frequency in response to increasing doses of LSD (Table 1).

Staring, grooming, and head or body shakes showed significant dose-dependent increases in frequency which peaked at 25-100 ug/kg ($p < 0.01$ or 0.001, ANOVA). The frequency of all of them declined significantly at the highest dose ($p < 0.01$ or 0.001, t-test) (Table 1). Saline treated animals spent most of their time either with their eyes closed or staring out the front of their cages, whereas LSD treated animals frequently stared (a 5 sec fixation) at the top, bottom and sides of their cages. LSD treated animals engaged in an inordinately large number of grooming bouts, but these bouts were of very short duration. Limb flicking and, of course, abortive grooming were frequently associated with these occurrences. Head or whole body shakes increased in a dose-dependent manner up to 50 ug/kg where its frequency was approximately five times that of the saline injected controls. Head and body shakes might be important behaviors in a model of the actions of LSD were it not for the fact that they are spontaneously emitted at a relatively high rate ($\bar{X}$ = 6.4/hr) in saline treated cats.

Peripheral Serotonin Blockers

Since LSD is known to be a potent serotonin antagonist in the periphery, we examined the effect of its non-hallucinogenic congener, brom-LSD, which has the same potency peripherally, and methysergide, which is more potent than LSD in blocking the action of serotonin in the periphery. Brom-LSD in doses of 25 and 100 ug/kg produced very little behavioral change except for a mean rate of 1.6 flicks/hr at the higher dose. In general, there was very little gross change in behavior following either drug dose. Methysergide maleate in doses of 100 ug/kg and higher produced effects similar to a low dose of LSD, e.g., the 100 ug/kg dose elicited 3.8 flicks/hr ($p < 0.025$, t-test) and 1.8 episodes of abortive grooming/hr ($p < 0.025$, t-test).

Drugs from Other Classes

In an attempt to explore the specificity of the behavioral response to LSD, we examined the effects of psychoactive drugs from various drug classes. In general, none of these compounds produced any effects similar to those produced by LSD. D-amphetamine sulfate in low doses (0.25 mg/kg) produced very

little behavioral change. In higher doses (1.0 and 5.0 mg/kg) it produced complete immobility, except for continuous stereotyped head and eye movements. Atropine sulfate had virtually no effect at 0.5 mg/kg, and produced ataxia, restlessness and plaintive meowing at 2.5 mg/kg. Caffeine was almost without effect except at the highest dose (20 mg/kg) where it seemed to activate or arouse the animals. Chlorpheniramine appeared to have no effect other than mild sedation in any dose.

Other Hallucinogens

The generality and importance of the behavioral changes observed in response to LSD could best be demonstrated by testing a structurally related indole nucleus hallucinogen, for example, psilocybin. Psilocybin closely mimicked the effects of low doses of LSD. A dose of 100 ug/kg elicited larger behavioral effects than either a lower dose (25 ug/kg) or a higher dose (500 ug/kg), which is similar to the curvilinear relation seen in response to LSD. The 100 ug/kg dose produced 6.3 flicks/hr ($p < 0.025$, t-test) and 2.7 episodes of abortive grooming/hr ($p < 0.05$, t-test). For comparison, we examined the effect of a non-indole nucleus hallucinogen, $\Delta^9$-tetrahydrocannabinol (THC). THC in doses ranging from 0.5 to 5.0 mg/kg produced little more than emesis and ataxia at the high doses. There was no similarity to any aspect of the LSD effect. As a final control, we examined the effect of a non-hallucinogenic indole nucleus compound, tryptamine. In doses ranging from 50 ug/kg to 5 mg/kg, no behavioral change was observed, except emesis at the highest dose.

Duration of Behavioral Effects

The 50 ug/kg dose of LSD produced behavioral changes lasting for at least 8 hours, while the 10 ug/kg dose produced changes lasting up to 4 hours. Analysis of variance revealed significant time-related changes in limb flicking, abortive grooming and investigatory or play behavior following both doses of LSD. Hallucinatory-like behavior, however, showed no significant time-related changes at either dose of LSD (Table 2).

Limb flicking was the most persistent behavior, following both doses of LSD. It still occurred at a frequency significantly above baseline 8 hours after 50 ug/kg and 4 hours after 10 ug/kg doses of LSD ($p < 0.05$, Dunnett's Test). By contrast, the frequency of abortive grooming, investigatory and hallucinatory-like behavior returned to baseline levels within 3-4 hours following either 10 or 50 ug/kg doses of LSD.

Following the administration of LSD many of the cats became very attentive and responsive. They often continually bounded about their cages, and occasionally fell off their perches. This general arousal effect typically lasted for 2-3 hours following drug administration. From 4-8 hours after LSD administration, the cats became relatively inactive and rested

TABLE 2

Duration of Behavioral Effects of LSD*

| BEHAVIOR | LSD (UG/KG) | MEAN % OF 1ST HR RESPONSE FREQUENCY HOURS POST-INJECTION 1 | 2 | 3 | 4 | 6 | 8 | 24 |
|---|---|---|---|---|---|---|---|---|
| Limb flick | 10 | 100% (11.3±5.0) | 89.4 | 102.7 | 70.8 | 30.1 | 16.8 | 0.0 |
| | 50 | 100% (40.0±6.6) | 79.0 | 81.0 | 79.8 | 30.0 | 21.8 | 0.0 |
| Abortive grooming | 10 | 100% (3.0±0.9) | 46.7 | 13.3 | 20.0 | 0.0 | 0.0 | 0.0 |
| | 50 | 100% (3.3±1.6) | 93.9 | 42.4 | 30.3 | 0.0 | 0.0 | 0.0 |
| Investigatory or play | 10 | 100% (6.6±4.0) | 86.4 | 59.1 | 28.8 | 0.0 | 0.0 | 0.0 |
| | 50 | 100% (16.9±4.5) | 56.8 | 37.3 | 23.7 | 0.0 | 0.0 | 0.0 |
| Hallucinatory-like | 10 | 100% (0.7±0.5) | 0.0 | 42.9 | 0.0 | 0.0 | 0.0 | 0.0 |
| | 50 | 100% (1.3±0.4) | 23.1 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |

*Data are presented as mean percent of the first hour responses, which are indicated in parentheses. N=7 per group.

quietly in their cages, only occasionally moving about. Even though they displayed virtually no spontaneous behavior during this period, limb flicks were still being emitted at a rate significantly above baseline levels. The behavior of all cats appeared completely normal 24 hours after a single dose of LSD.

Duration of Tolerance

A single 50 ug/kg dose of LSD produced a long-lasting tolerance which was maximal at 1 day and persisted for approximately 5 days (Table 3). In the case of limb flicking, tolerance to a test dose of 50 ug/kg of LSD one day after a single dose of 50 ug/kg of LSD was virtually complete, and a substantial tolerance (80.5% below baseline) was present at 3 days post-injection ($p < 0.05$, Dunnett's Test). Five days post injection, the mean number of limb flicks was still 29.1% below baseline. By comparison, the tolerance to a test dose of 50 ug/kg of LSD following a single dose of 10 ug/kg of LSD was not as pronounced nor long-lasting. Tolerance under these conditions was quite marked at one day post-injection (91.0% below baseline, $p < 0.05$, Dunnett's Test), but was only 41.0% below baseline 3 days post-injection. In general, abortive grooming and the other behavioral measures showed a similar pattern of tolerance (Table 3).

The general behavioral state of these cats was dramatically different under the tolerance condition as compared to their behavior following LSD alone. All of the behaviors which were quantified were greatly decreased one day after the initial dose of LSD, especially at the higher initial dose (50 ug/kg) (Table 3). Furthermore, many of the tolerant cats did not show the typical arousal effect elicited by LSD administration. In addition, the general disjunctive nature of behavior produced by LSD was greatly diminished in tolerant animals.

Onset of Tolerance

A significant tolerance was observed as early as 2 hours after administration of a single dose of LSD (Table 3). Limb flicking showed tolerance to a test dose of 50 ug/kg of LSD 2 hours after a single dose of 10 ug/kg of LSD (46.8% below baseline, $p < 0.05$, Dunnett's Test). Furthermore, cats receiving 50 ug/kg of LSD 2 hours after 10 ug/kg did not show the typical arousal response seen following 50 ug/kg of LSD alone. The tolerance is much more marked at 6 hours after the initial dose of LSD (75.6% below baseline, $p < 0.001$), and reaches a maximum at 24 hours (91.0% below baseline, $p < 0.001$). In general, abortive grooming and the other behavioral measures manifested a similar rapid onset of tolerance, reaching significance either by 2 or 6 hours following the initial LSD injection.

TABLE 3

Onsert and Duration of Tolerance to LSD*

| BEHAVIOR | BASELINE HOURLY RATE | LSD DOSE (UG/KG) | MEAN % OF LSD BASELINE RATE INTER-LSD INJECTION INTERVAL (HRS) 2 | 6 | 24 | 72 | 120 | 168 | 336 |
|---|---|---|---|---|---|---|---|---|---|
| Limb flick | 34.4±8.7 | 10 | 53.2% | 24.4 | 9.0 | 59.0 | 92.2 | 105.5 | ---- |
| | | 50 | ---- | ---- | 3.8 | 19.5 | 70.9 | 110.2 | 116.3 |
| Abortive grooming | 4.6±2.2 | 10 | 52.2% | 10.9 | 23.9 | 67.4 | 56.5 | 130.4 | ---- |
| | | 50 | ---- | ---- | 2.2 | 8.7 | 78.3 | 110.9 | 71.7 |
| Investigatory or play | 10.8±4.0 | 10 | 36.1% | 19.4 | 28.7 | 95.4 | 133.3 | 120.4 | ---- |
| | | 50 | ---- | ---- | 3.7 | 61.1 | 105.6 | 110.2 | 156.5 |
| Hallucinatory-like | 1.0±0.5 | 10 | 50.0% | 20.0 | 40.0 | 100.0 | 170.0 | 140.0 | ---- |
| | | 50 | ---- | ---- | 10.0 | 40.0 | 60.0 | 160.0 | 80.0 |

*A single dose of either 10 or 50 ug/kg of LSD tartrate was administered, and test doses of 50 ug/kg of LSD were administered to separate groups of cats (N=7/group) at 2, 6, 24, 72, 120, 168 or 336 hours later. Data presented are mean percent of BASELINE HOURLY RATE (2nd column), which are the absolute values recorded in response to a 50 ug/kg dose of LSD in non-tolerant animals.

## Discussion

### Validity of the Model

Because limb flicking, abortive grooming, investigatory or play behavior, and hallucinatory-like behavior are emitted in response to LSD and psilocybin, and because they are never seen in response to a variety of other psychoactive compounds, we propose that they may be used as an animal behavior model for the actions of these hallucinogens and structurally and/or functionally related compounds (see below). Furthermore, observation and measurement of the first two of these behaviors, limb flicking and abortive grooming, may be sufficient for utilization as an animal model because: they are the easiest behaviors to score; they are the most reliable measures; they occur at a high frequency; and they are observed in all animals tested.

Previous studies of the effects of LSD and related hallucinogens in cats failed to report either limb flicking or abortive grooming, and failed to delineate any behavioral changes that might be specific to these compounds (3,25,28,31, 32,36,49,51,53,61). Studies of hallucinogens in rats and mice typically have utilized behavioral measures such as startle or the disruption of a conditioned response, effects which are obviously not specific to hallucinogenic drugs (11,13,21,22, 29-31,46,50,56,58,62,63). By contrast, the present behavioral measures appear to be specific to hallucinogenic drugs and are essentially nonexistent in control animals. These measures also have face validity in the sense that the constituent behaviors can be described as bizarre or hallucinatory-like in the context which they occur.

The specificity of these emergent behaviors is indicated by the fact that they were never seen in response to other classes of major psychoactive drugs, such as those affecting the catecholamines (amphetamine), acetylcholine (atropine), or histamine (chlorpheniramine). Caffeine, a phosphodiesterase inhibitor, was also ineffective. These behavioral effects of LSD are not attributable to the inactivation of serotonin in the periphery (18,64), since methysergide, a more potent blocker of serotonin's action in the periphery (27), did not elicit any of these emergent behaviors at 25 ug/kg, a dose of LSD that produced potent behavioral changes. However, when the dose was increased, several flicks and episodes of abortive grooming were observed. This correlates well with the fact that high doses of methysergide have been reported to be moderately hallucinogenic in humans (15,34). Further support that the behavioral effects are not due to a peripheral action of LSD comes from the fact that its non-hallucinogenic congener, brom-LSD (18), which has the same peripheral action as LSD (17), was ineffective in eliciting the emergent behaviors in a dose of 25 ug/kg. However, a dose of 100 ug/kg produced 1.6 flicks/hr which might be related to the observation that high doses of brom-LSD are mildly hallucinogenic (38).

response to a 50 ug/kg dose of LSD in non-tolerant animals.

Mechanism of Action of LSD

Systemic administration of low doses of LSD selectively depresses the activity of serotonin-containing raphe neurons (5), and because of serotonin's exclusively inhibitory synaptic action this results in an increase in the activity of neurons postsynaptic to raphe cells (33). On the other hand, while low doses of LSD have an effect on raphe neurons, which acts to decrease the serotonergic input to their postsynaptic neurons, high doses of LSD have the opposite functional effect on these same postsynaptic neurons: a direct serotonin agonist effect (60). These opposing effects of high and low doses of LSD may account for the significant decreases in virtually all behaviors seen in response to the highest doses of LSD in the present study. Thus, while low doses of LSD may disinhibit the activity of postsynaptic neurons, high doses may directly inhibit their activity. In many cases the 200 ug/kg dose of LSD was only as effective behaviorally as the 2.5 ug/kg dose.

If the emergent behaviors seen in response to LSD are a manifestation of the depression or inactivation of the brain serotonin system, then other functionally similar compounds should also elicit these behaviors. Inactivation could be accomplished by a blockade of serotonin receptors, a depression of raphe unit activity, or by a decrease in brain serotonin. Consistent with this hypothesis, the presumptive central serotonin blocker methysergide produced moderate LSD-like effects in high doses, and psilocybin, which depresses the activity of serotonin-containing raphe neurons in the rat (unpublished observations), also elicited the emergent behaviors. We have also previously reported that cats administered the serotonin depleting drug, p-chlorophenylalanine, manifest limb flicking and abortive grooming (59). Finally, the model's applicability might extend beyond the indole nucleus hallucinogens to include amphetamine derivatives such as mescaline and DOM (STP), both of which are known to depress the activity of select groups of raphe neurons (6). Preliminary evidence from our laboratory supports the validity of extending the model to include these two compounds. Thus, the model's utility as a heuristic device encompasses a structurally heterogeneous group of drugs, which share the functionally homogeneous property of inactivating the brain serotonin system.

Duration of Behavioral Effects of LSD

Several studies have reported that the behavioral and perceptual effects of 1-2 ug/kg of LSD in humans peaks in 1-1/2 to 2 hours and lasts for approximately 8 hours (2,37, 45,52). This is consistent with the relatively long half-life of LSD in human plasma of 3 hours (4). Since the half-life of LSD in cat plasma is approximately 2 hours (14), we predicted that, if the behavioral syndrome in the cat were a good model of the action of LSD in humans, high doses of LSD should induce behavioral changes lasting for at least 8 hours. The data revealed that a dose of 50 ug/kg of LSD produced a

significant increase in limb flicking lasting until at least 8 hours post-injection. None of the other components of the behavioral syndrome showed significant changes 8 hours post-LSD injection. At the lower dose of LSD (i.e., 10 ug/kg), limb flicking was significantly increased for 4 hours post-injection, while none of the other behaviors were significantly increased beyond 2 hours post-injection at this dose of LSD. Thus, limb flicking seems to be the most sensitive measure of the action of LSD in the cat, and thus most closely parallels the action of LSD in humans. The limb flick is also a highly reliable index, as indicated by the fact that the mean number of flicks emitted by a group of 7 cats on three different occasions separated by 3 weeks were 34.4, 37.9 and 40.0 per hour (see Table 3).

Onset and Duration of Tolerance to LSD

Several early investigators found that a dramatic, long-lasting tolerance develops to the psychological and perceptual effects of LSD following its repeated administration to humans (1,19,37). This effect has been confirmed in a large number of studies on animals as well as humans (2,3,12,29,30,42,62). There is considerable disagreement, however, as to how long the tolerance persists and how rapidly it develops.

Much of the variability in the data on the development of tolerance to LSD undoubtedly arises from the variety of behavioral indices used, as well as the different species and pretreatment regimens employed. More importantly, however, is the fact that none of the behavioral measures employed are a specific reflection of the action of hallucinogens. Since the behavioral syndrome in the cat is elicited only by a functionally homogeneous class of hallucinogens, these behavioral changes provide the opportunity to study the parameters of tolerance to those drug effects which are specific to these compounds. We have demonstrated in the present study that tolerance to the behavioral effects of LSD at a dose of 50 ug/kg is virtually complete within one day after a single dose of 50 ug/kg of LSD, and is very marked after a single dose of 10 ug/kg. Not only is the tolerance after the higher dose of LSD more pronounced, but longer lasting as well: the 50 ug/kg dose of LSD induced a significant tolerance lasting for 3-5 days, while the 10 ug/kg dose produced a significant tolerance for 1-3 days. We also demonstrated that partial tolerance to a 50 ug/kg test dose of LSD following a single injection of 10 ug/kg of LSD develops very rapidly. For example, using the limb flick as a behavioral index, tolerance is significant within 2 hours (46.8% below baseline) and is quite dramatic after 6 hours (75.6% below baseline).

Usefulness of the Model

Because the model specifically reflects the functional activity of a distinct class of hallucinogens, it would be useful in the study of a wide variety of neurochemical,

pharmacological, electrophysiological and behavioral questions related to this class of drugs.

In addition to examining the onset and duration of tolerance, as we have done in the present studies, cross-tolerance could also be investigated with this model. The development of cross-tolerance between two compounds may be a probe into the similarity of the neuronal substrates of their actions. A strong hypothesis would state that only those compounds sharing the same mechanisms of action will show cross-tolerance. In the present context this would be transformed into the following specific hypothesis: only those compounds which elicit the emergent behaviors in cats should show cross-tolerance to LSD. Because some of the behaviors, such as limb flicks, give reliably quantifiable data, the degree of cross-tolerance, if less than total, could be assessed.

The relative potency of hallucinogenic compounds could be examined by comparing both the dose necessary to produce the emergent behaviors and the magnitude of the response elicited. In the same vein, the model could be used to predict whether as yet untested compounds would have hallucinogenic potency in humans.

Blocking the action of hallucinogens could be explored through the use of the model. These studies would potentially be of interest both clinically and for basic research. If the emergent behaviors are a manifestation of the depression of the activity of the brain serotonin system, as suggested above, then compounds which act in a reciprocal fashion should block the syndrome. Compounds which selectively increase the functional activity in serotonin mediated synapses of the cat should be effective blockers, whereas those compounds which increase the activity of other neurotransmitters should be less effective (or ineffective) blockers. These studies would complement our previous finding that prior serotonin depletion potentiated the behavioral effects of LSD (59). The model could also be used to study the blocking action of compounds such as the phenothiazines, which may act by blocking the output of those neuronal systems disinhibited by the hallucinogens, rather than by increasing the functional activity in serotonin mediated synapses.

Finally, the model could be used to determine whether a correlation exists between the depression of raphe unit activity and the behavioral changes induced by LSD, by recording raphe unit activity in freely moving cats. A dose of LSD which is just at the threshold of producing behavioral effects significantly different from saline could be examined to see if it significantly depresses raphe unit activity. Also, a range of doses of LSD could be examined to see whether the quantifiable behavioral changes positively correlate with the degree of depression of raphe unit activity. This experiment would also provide a major test of the hypothesis that inactivation of the brain serotonin system is a necessary precondition for

the hallucinogenic effects of LSD. Other hallucinogens known to depress raphe unit activity could also be tested (6,7,40, 48).

## Conclusion

We propose that the emergent behaviors in the cat, especially limb flicking and abortive grooming, represent a viable animal behavior model for the actions of hallucinogens in humans. The model fulfills all the criteria for a valid model that were discussed in the introduction.

1. Specificity - the behaviors were seen exclusively in response to compounds which inactivate the brain serotonin system.
2. Dose dependent - the behaviors significantly increased in frequency in a dose-dependent manner.
3. Human range - limb flicking showed a significant increase in frequency in response to a dose of 2.5 ug/kg LSD, while the other three emergent behaviors significantly increased in response to a 10 ug/kg dose.
4. Parallel major parameters - the behavioral changes observed in the cats were similar to those previously reported in humans with regard to the duration of action of LSD, and the onset and duration of tolerance to LSD.
5. Robust - the changes in limb flicking and abortive grooming were observed in every animal tested.
6. Reliable - the mean frequency of limb flicks in response to a 50 ug/kg dose of LSD was stable when tested over a period of several weeks.
7. Quantifiable.
8. Easy to score.

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