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# EFFECTS OF PRENATAL AND POSTNATAL EXPOSURE TO LSD ON BRAIN MATURATION

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**Abstract**—1. Mice treated by prenatal injection of pregnant females with LSD or postnatal injection of neonates with LSD or LSD and testosterone were analyzed at various stages of postnatal maturation for changes in brain or body weight or changes in indoleamine levels.

2. LSD caused some variable changes in weight of some brain regions, principally the brain stem and diencephalon of prenatally and postnatally treated mice. Monoamine oxidase levels were elevated in prenatally treated mice.

3. There does not appear to be any clear explanation for these effects of LSD.

## INTRODUCTION

THE POSSIBLE effects of lysergic acid diethylamide (LSD) in development appear to fall into three categories; interaction with various biogenic amine biochemical pathways, (2) teratology or gross morphological defect, and (3) genetic damage as chromosomal breakage. The best known and most studied of these effects are its interactions with various components of the indoleamine biochemical pathway. These have been recently summarized by Freedman & Boggan (1974). Evidence for teratologic or genetic damage effects have been reviewed by Dishotsky *et al.* (1971), Kalter (1972) and Long (1972), but these reviewers find this evidence often conflicting and unconvincing.

This laboratory has undertaken extensive investigations of the effects of LSD on the immature brain's indoleamine biochemistry and its possible effects on brain growth and behavior (Hoff *et al.*, 1972; Goodrich *et al.*, 1974; Baker & Hoff, 1975). Goodrich *et al.* (1974) found that LSD exposure on postnatal day 1 caused a significant reduction in weight of certain brain regions of male mice and suggested this effect may be related to residual levels of prenatal androgen. Baker & Hoff (1975) found that LSD exposure on postnatal day 7 altered 5-hydroxytryptamine (5-HT) and monoamine oxidase (MAO) levels in certain brain regions. This project was undertaken to further focus on what effects prenatal and postnatal exposure to LSD may have on brain weight, what effect prenatal exposure to LSD may have on indoleamine maturation, to determine if there is some interaction between LSD and residual prenatal androgens affecting brain weight, and to determine if there is a transient biochemical response to LSD.

## METHODS

Mice of the CFW strain (Carworth Farms) were used for these experiments. They were maintained on Purina Lab Chow with a lighting regime of 10 hr light and 14 hr darkness. For biochemical studies, mice were killed during the fourth hour of light. For prenatal exposure, pregnant females received a single dose of 5 μg/kg body weight i.p. of LSD in saline on postconception day 12 or 18. Offspring of both these were measured for weight changes at 8 weeks of age: offspring of the 12 day postconception treated mice were measured for biochemical changes as well at 1 day and 1, 2, 4 and 8 weeks postnatal. For postnatal exposure, all neonates received LSD at a dose of 50 μg/kg body weight i.p. of LSD in saline. To refine the time at which LSD might cause changed brain weight, neonates exposed to a single dose of LSD on postnatal day 1 were killed and weights taken on days 3, 8, 14, 28, 42 and 8 weeks. Neonates exposed to a single dose of LSD on day of birth and postnatal days 3 and 5 were killed and weights taken at 8 weeks of age. Neonates exposed to multiple doses of LSD on postnatal days 1, 2, 3 and 4 had body weights monitored at 1, 2, 4 and 8 weeks and brain weights taken at 8 weeks. To determine if there was an interaction between LSD and testosterone, neonates on postnatal day 1 received a single LSD dose of 50 μg/kg body weight along with 100 μg of testosterone proprionate in 0.02 ml peanut oil and brain weights taken at 8 weeks. To determine if there is a transient biochemical response to LSD, mice of 1 week and 2 weeks of age received a single LSD dose of 50 μg/kg body weight and were measured at 30 and 60 min after exposure. Brains were removed from the braincase according to the method previously described by Baker & Hoff (1972), and subdivided into five parts including the cerebellum, hemispheres, medulla–pons, mesencephalon and diencephalon.

5-Hydroxytryptamine (5-HT) and 5-hydroxyindole acetic acid (5-HIAA) were measured by the OPT method of Miller *et al.* (1970). Tryptophan-5-hydroxylase (T5-H) was measured by the Kuhar *et al.* (1971) modification of the *in vitro* $CO_2$ capture method of Ichyama *et al.* (1970) using $^{14}$C-tryptophan as substrate. 5-Hydroxytryptophan decarboxylase (5-HTPD) was measured using the Baker (1966a) modification of the method of Snyder & Axelrod (1964) using $^{14}$C-5-hydroxytryptophan as substrate. Monoamine oxidase (MAO) was measured by the Baker (1966b) modification of the method of Wurtman & Axelrod (1963) using $^{14}$C-5-hydroxytryptamine as substrate.

Table 1. Body and brain region weights in grams of 8 week old mice exposed to LSD at 12 or 18 days post-conception at a dose of 5 μg kg of maternal body weight

| | Body weight ♂ 12 day P.C.E. | Mesencephalon ♂ 12 day P.C.E. | Medulla pons ♀ 12 day P.C.E. | Medulla pons ♀ 18 day P.C.E. |
|---|---|---|---|---|
| Control | | | | |
| $N$ = | 29 | 29 | 21 | 28 |
| $X$ = | 29.23 | 0.0285 | 0.0512 | 0.0546 |
| S.E.M. = | 0.46 | 0.0004 | 0.0008 | 0.0007 |
| LSD treated | | | | |
| $N$ = | 29 | 29 | 26 | 21 |
| $X$ = | 30.98 | 0.0298 | 0.0531 | 0.0527 |
| S.E.M. = | 0.63 | 0.0004 | 0.0005 | 0.0006 |
| $P$ = | <0.05 | <0.05 | <0.05 | <0.05 |

P.C.E. = post-conception exposure; $N$ = number in sample; $X$ = mean value; S.E.M. = standard error of the mean; $P$ = probability of differences from controls derived from Student's $t$-test.

## RESULTS

Exposure of pregnant mice to LSD at 12 days post-conception resulted in an increase in body weight and in the mesencephalon weight of male offspring at 8 weeks and in medulla–pons weight of female offspring at 8 weeks (Table 1). Exposure at 18 days postconception resulted in a decrease in medulla–pons weight of female offspring at 8 weeks (Table 1). Exposure of neonates at 1 day postpartum resulted in an increase in diencephalon weight of female mice at 8 days and 14 days, but was not detectable after this, and an increase in mesencephalon weight of male mice at 6 weeks of age (Table 2). No other significant brain weight or body weight changes were detected for any of the LSD or testosterone treated mice.

The only biochemical change detected was an elevation of MAO levels in the mesencephalon of 12 day postconception LSD treated animals at 1, 4 and 8 weeks of age (Table 3). There was no transient response to LSD exposure.

## CONCLUSIONS

The results indicate that some significant changes in brain and body weights may be caused by exposure to LSD at various stages of development. There is no indication that this effect may be related to residual androgens present at birth as suggested by Goodrich *et al.* (1974). The results tend to indicate that, if anything, LSD may cause an increase in weight of certain brain regions, principally the brain stem or diencephalon. The decrease in medulla–pons of 8 week old female mice exposed at 18 days post-conception is the one exception to this. The increase in MAO activity of 12 day postconception treated mice is in accord with the increased MAO activity in 1 week postnatally treated mice reported by Baker & Hoff (1975).

There does not appear to be any explanation for these weight changes. It does not appear to be a nutritional factor since mice receiving multiple doses of LSD on postnatal days 1, 2, 3 and 4 had no significant weight deviations. Nair (1974), however, reported that LSD may cause early death and some reduction of growth, but no brain weight changes, but a higher dose was used in these experiments. One possible mechanism is that LSD may be affecting brain cell lysosomal systems and resulting in abnormal cytology (Hendelman, 1972). The effect of LSD on brain weight may be related to its effect on the brain's serotonergic

Table 2. Brain region weights in grams of maturing mice exposed to LSD on post-natal day 1 at a dose of 50 μg/kg of body weight

| | Diencephalon ♀ 8 days old | Diencephalon ♀ 2 weeks old | Mesencephalon ♂ 6 weeks old |
|---|---|---|---|
| Control | | | |
| $N$ = | 20 | 19 | 13 |
| $X$ = | 0.0409 | 0.0493 | 0.0284 |
| S.E.M. = | 0.0006 | 0.0006 | 0.0005 |
| LSD treated | | | |
| $N$ = | 28 | 23 | 15 |
| $X$ = | 0.0424 | 0.0511 | 0.0299 |
| S.E.M. = | 0.0005 | 0.0005 | 0.0004 |
| $P$ = | <0.05 | <0.05 | <0.05 |

$N$ = number in sample; $X$ = mean value; S.E.M. = standard error of the mean; $P$ = probability of differences from controls derived from Student's $t$-test.

Table 3. Monoamine oxidase levels in mesencephalon of maturing mice exposed to LSD on post-conception day 12 as a dose of 5 μg/kg of maternal body weight. Results expressed as a percentage of control values

| | 1 week old | 4 weeks old | 8 weeks old |
|---|---|---|---|
| Control | | | |
| $N$ = | 8 | 15 | 10 |
| $X$ = | 100 | 100 | 100 |
| S.E.M. = | 2.61 | 2.62 | 4.33 |
| LSD treated | | | |
| $N$ = | 8 | 15 | 13 |
| $X$ = | 117 | 109 | 113 |
| S.E.M. = | 6.41 | 2.93 | 2.86 |
| $P$ = | <0.05 | <0.05 | <0.05 |

$N$ = number in sample; $X$ = mean value; S.E.M. = standard error of the mean; $P$ = probability of differences from controls derived from Student's $t$-test.

system since some other drugs that interact with this system also affect brain weight (Hole, 1972a; 1972b).

The most studied of LSD's biochemical effects are its interaction with the serotonergic system. Its effect here may be related to the stage of development this system is in at the time of exposure to LSD. Changes in MAO levels in mice treated with LSD at 12 days postconception correlate well with the time at which serotonergic neurons are first demonstrable (Olson & Seiger, 1972). LSD induced changes in 5-HT and MAO in mice treated with LSD at 1 week postpartum reported by Baker & Hoff (1975) correspond to the time when final serotonergic outgrowth and terminalization is accomplished (Loizou, 1972).

How LSD can effect such apparently permanent, or at least long-term, disruption of the serotonergic system is unclear. It is tempting to relate it to a possible trophic function for indoleamines in early development (Seiger & Olson, 1973; Lauder & Bloom, 1974), but since such a function is unproven, this would be hazardous. Also puzzling is the fact that LSD consistently elevates MAO activity (Baker & Hoff, 1975), whereas most drug manipulations usually result in enzymatic inhibition. The fact that it does affect MAO more consistently then other enzymes of the serotonergic system may indicate that this enzyme is more important in serotonergic maturation than previously suspected.

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