Lysergic Acid Diethylamide (LSD-25) and Gamma Amino Butyric Acid (GABA) Interaction on Spontaneous Potentials and the Evoked Response in the Avian Tectum[1] (37648)

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Many compounds have been studied in attempts to antagonize the effects of LSD (1). These studies have been carried out in species ranging from the spider and guppy fish to monkeys and man, but the wide variety of test systems and methods of evaluating the LSD "syndrome" have made evaluation of the results difficult. The evaluation is further complicated by the fact that some of the compounds studied are inactive because of the permeability characteristics of the blood–brain barrier. These difficulties account for the lack of information on the effects of serotonin, GABA, epinephrine and norepinephrine on the LSD effect. Also hallucinogenic drugs have variable effects in subhuman species. Another problem is related to the variability of the effects of different hallucinogenic drugs in subhuman species.

The young avian preparation permits circumvention of the blood–brain permeability difficulties (2–4) and has the additional advantage of responding to many psychotomimetic drugs in the same way (5–7). This preparation could be a suitable tool for the study of antagonists to psychotoxic drugs.

This study is concerned with the action of one of the important neuroamines, gamma aminobutyric acid (GABA), on the effects of LSD in the avian tectum.

The selection of GABA as a potential LSD antagonist was not based on presumptive evidence but on the observation that physiologically and pharmacologically it is mainly depressant or inhibitory. Changes in the endogenous levels of GABA in the brain are inversely related to proportionate changes in brain excitability (8, 9). LSD action in the avian tectum is predominantly stimulatory in nature (see below).

Methods. Experimental animals were 19 acutely prepared New Hampshire chicks of indeterminate sex, 1 to 2 wk of age, weighing 70–90 g.

The chicks were prepared for recording bioelectric potentials from the optic tectum, the electrocardiogram, blood pressure, body temperature and expiratory CO_2 as described in detail elsewhere. Surgical procedures, techniques of artificial respiration, and drug administration have also been described (4, 6, 7).

When surgical procedures were completed, ether anesthesia was replaced by immobilization with *d*-tubocurarine and topical anesthesia at all operative sites.

GABA solution (100 mg/ml) was prepared immediately prior to use by dissolving the crystalline material in normal saline (0.9%). LSD-25 was prepared in a similar way (10 μg/ml). Both drugs were individually administered by slow infusion (0.77 cm^3/min) to the animal through the cannulated femoral vein.

Results. The results of a typical experiment are shown in Fig. 1. Following a control recording (top trace) of both spontaneous activity and the evoked response (arrows and column on the right), LSD was administered intravenously. At the first indication of an LSD response, specifically a change in the spontaneous activity (middle trace), LSD administration was discontinued and GABA infusion was started and continued until the evoked response was inhibited to where it was just discernible (bottom trace). The dose of LSD required to reach this cri-

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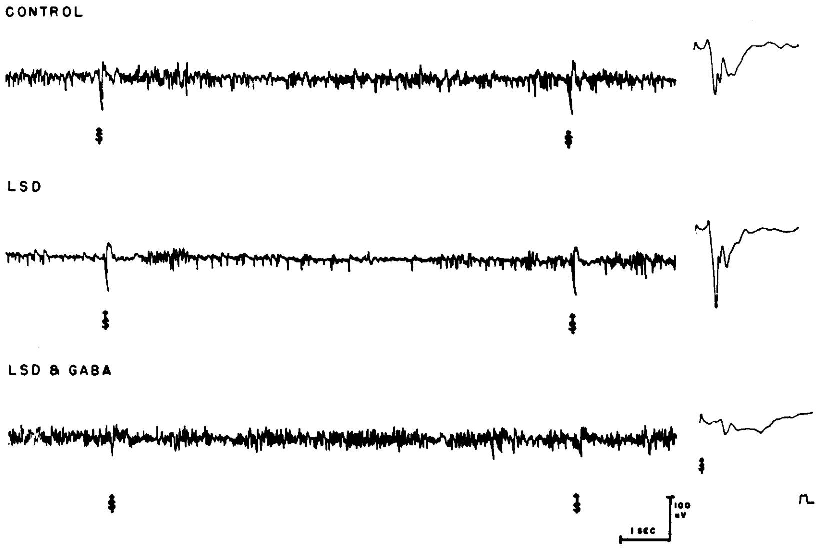

FIG. 1. Effect of GABA on LSD-induced changes in spontaneous and evoked potentials recorded from the avian optic tectum. Polygraph traces are as designated in the illustration. Arrows denote point of stimulation induced by photic activation of the intact retina. Trace to the right is the average of 25 evoked responses in each case. Negativity at the active electrode is denoted by a downward deflection. Calibration signal for the averaged responses (lower right hand corner) = 10 µV × 50 msec at the source.

terion was 170 µg/kg ± 95. The dose of GABA administered was recorded and is shown for each experiment in Table I.

On the average, the dose of GABA required to inhibit the evoked response was greater following LSD than when given alone. The difference between the mean dosage in the two cases is significant at $p < 0.01$ level as determined by Student's t test. While GABA alone has little observable effect upon spontaneous activity (4, 6) it appears capable of reversing or interfering with the LSD effect upon this parameter. The right column of Fig. 1 shows the algebraic average of 25 evoked responses. Note the enhanced response following LSD, while the opposite occurs with GABA. LSD activity, in the absence of GABA, persists for well over 1 hr (7). Since the time interval between onset of LSD action (7) and the administration of GABA is less than 15 min, it is unlikely that normal deterioration of LSD action contributes to the GABA reversal.

If GABA is administered first in a dose which inhibits the evoked response (Table I, Column 2), LSD will change the spontaneous potentials, but it will not reverse the depressed evoked response. This is true even if the amount of LSD given is two to three times the usual ED_{50}.

Discussion and Conclusion. In the chicken, parenteral administration of GABA causes an inhibition of the photically evoked response recorded from the tectum. It does not have any marked effect on spontaneous potentials

TABLE I.

Expt no.	GABA alone (g/kg)	Expt no.	GABA after LSD (g/kg)
1	2.18	12	1.83
2	1.79	13	1.98
3	1.82	14	1.90
4	1.52	15	1.98
5	1.43	16	2.52
6	2.06	17	3.28
7	1.58	18	1.89
8	1.07	19	2.42
9	2.10		
10	1.21		
11	1.23		
Mean[a]	$1.64 = \bar{x}_1$		$2.33 = \bar{x}_2$
S^2	0.157		0.248

[a] Difference in means significant at $p < 0.01$.

recorded from the same structure (4, 6). These results, considered with other data, lead to the conclusion that GABA acts primarily as an inhibitor upon axosomatic synapses (6, 10). LSD decreases the amplitude of spontaneous activity, enhances the evoked response and uncovers a prolonged series of potentials (afterpotentials) following the evoked response (7, 11). At the synaptic level, these results suggest an inhibitory effect upon axodendritic (depression of spontaneous activity) and an excitatory effect upon axosomatic connections (evoked response plus afterpotentials) (12, 13).

The interaction of LSD and GABA is not surprising. It now appears possible to state more precisely the site and mechanism of LSD action in the avian brain.

It has long been known that lowered brain GABA levels are associated with increased CNS excitability. The motor component of this excitation, however, is not easily correlated with GABA levels. Convulsions induced by the thiosemicarbazides and pyridoxine antagonists are accompanied by a decrease in brain GABA levels (14). But other experiments have shown that under the appropriate conditions thiosemicarbazide-induced motor convulsions can be terminated even though brain GABA levels remain low (9). Moreover, in the opposite situation, seizures can be induced with thiosemicarbazides even in the presence of raised GABA levels (8).

Since there have been no previous studies concerned with brain GABA levels and sensory excitability, we feel that the current results support a view that GABA and CNS excitability studies could best be restricted to sensory mechanisms.

The action of LSD on the electrophysiology of the tectum is predominantly excitatory, but there appears to be no concomitant motor effect as observed in the intact animal (personal observation) that cannot be attributed to sensory stimulation.

It is interesting to speculate that the primary action of LSD is to lower synaptic thresholds, primarily in sensory areas (*i.e.*, act as a specific sensory convulsant). Such an action would account for the hallucinatory effects (or increased sensory responsivity) produced by LSD without the concomitant effects upon motor areas. If this were true, it must be explained why the sensory areas are sensitive to LSD while motor areas are not. GABA must be considered, for, although not proven, studies indicate that GABA in the CNS is modulatory, principally in the complex coordinating centers of the brain.

Studies done with adrenochrome, which is also hallucinogenic in man, have shown this substance interferes with GABA metabolism *in vitro* through an inhibitory action on glutamic acid decarboxylase (15). Such data, in addition to our results, support the suggestion that the central mechanism of action of all hallucinogens may indeed involve the GABA system in the sensory areas of the CNS.

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