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The effect of some hallucinogenic drugs on the amino acid precursors of brain monoamines.†

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Primary inborn errors of the metabolism or of the membrane transport of amino acids are known to be responsible for some mental disorders. Mental retardation is the most prominent symptom of the effects of the aminoacidopathies on the central nervous system, but disturbances resembling psychoses sometimes occur. The symptoms of Hartnup disease, for example, are sometimes so similar to those of schizophrenia that the two conditions have been confused, as Greer (1965) and Herzov and Rodnight (1960) have shown. It is therefore conceivable that some of the psychotomimetic effects of hallucinogenic drugs might be produced by disturbances of amino acid metabolism.

Four structurally dissimilar hallucinogenic drugs, phencyclidine, Ditran, lysergic acid diethylamide (LSD) and mescaline, have been shown to alter the concentrations of 5-hydroxytryptamine (5-HT) and noradrenaline (NA) in the brains of rats (Tonge and Leonard, 1969; Leonard and Tonge, 1969). However, the dose of each drug required to affect the levels of these monoamines was considerably greater than the minimum dose required for the production of a recognisable pattern of behavioural effects. This observation suggested the

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possibility that the changes in brain monoamine concentration might be secondary to effects on the precursors of these neurohumours. Changes in the levels of tryptophan and tyrosine in whole brain and in plasma from rats treated with small doses of hallucinogenic drugs were therefore measured over the period during which behavioural effects could be observed.

METHODS

All drugs were administered to rats of the Wistar strain (inbred over several generations) by the intraperitoneal route.

In the absence of a satisfactory specific test for hallucinogenic activity in rats, the gross behaviour pattern was used as an approximate guide to the course of action of the drugs. Animals were killed by decapitation at intervals after the administration of each drug to see whether any relationship between neurochemical changes and behaviour could be observed. Mixed venous and arterial blood was collected from the neck vessels immediately after the decapitation of the animals. It was centrifuged, and the plasma diluted 1:10 with 0.4N perchloric acid and centrifuged at 3,000g for 10 minutes. Whole brains were rapidly removed, homogenised (Potter-Elvehjem homogeniser) in 10-12 ml of 0.4N perchloric acid and centrifuged for 10 minutes at 3,000 g. Tryptophan was measured by the spectro photo fluorimetric method of Hess and Udenfriend (1959) and tyrosine by that of Waalkes and Udenfriend (1962).

RESULTS

The behavioural changes observed following the administration of these hallucinogens have been described elsewhere (Tonge and Leonard, 1969; and Leonard and Tonge, 1969 and 1970).

The effects of the hallucinogens on brain tryptophan levels are shown in Table 1.

TABLE 1

The effect of phencyclidine (3mg/kg), Ditran (2mg/kg), Mescaline (5mg/kg) and LSD (100 microg/kg) on brain tryptophan concentrations.

	TIME AFTER INJECTION (MINUTES)						
Drug	0	10	20	40	60	100	180
Phency-clidine	35.6±0.8	29.0±1.9	*** 27.6±0.6	*** 29.5±1.9	35.8±2.0	35.1±2.5	* 37.0±1.4
Ditran	41.8±3.4	44.1±10.4	39.4±6.5	38.0±5.6	*** 29.2±3.8	** 25.9±8.7	44.3±6.3
Mesca-line	32.9±3.1	33.1±2.9	** 27.8±2.1	* 27.7±3.5	** 25.1±1.9	** 27.4±1.1	** 27.0±1.1
LSD	31.1±2.1	33.0±1.3	* 37.2±4.4	** 35.9±2.5	*** 41.9±2.9	*** 37.3±1.8	** 35.8±2.4

TABLE 2

The effects of phencyclidine (3mg/kg), Ditran (2mg/kg), mescaline (5mg/kg) and LSD (100microg/kg) on plasma tryptophan concentrations.

	TIME AFTER INJECTION (MINUTES)						
Drug	0	10	20	40	60	100	180
LSD	79.3±7.2	79.3±5.3	* 70.8±3.1	72.8±4.7	77.4±9.0	83.1±4.6	79.7±3.2
Phency-clidine	77.9±6.5	80.8±6.6	76.9±4.0	** 63.7±4.5	73.9±12.4	78.6±16.9	81.8±6.6
Ditran	85.7±9.0	86.2±14.9	* 74.7±9.7	** 66.1±10.2	77.4±7.4	** 66.0±6.7	* 70.0±11.1
Mescal-ine	66.9±4.6	68.0±2.2	** 59.3±2.0	60.7±10.6	65.1±3.1	* 60.6±1.9	65.5±3.7

It is apparent that, with the exception of LSD, the hallucinogens caused a depletion of tryptophan from the brains of rats. Changes in plasma levels of the amino acid at corresponding times after the injection of drugs are shown in Table 2.

Tables 3 and 4 show the effects of the hallucinogens on brain and plasma levels of tyrosine respectively.

Table 3

The effects of phencyclidine (3mg/kg), Ditran (2mg/kg), mescaline (5mg/kg) and LSD (100microg/kg) on brain tyrosine concentrations.

	TIME AFTER INJECTION (MINUTES)						
Drug	0	10	20	40	60	100	180
Phency-clidine	136.1±12.1	***167.8±7.7	**176.6±19.5	***199.8±21.8	*166.0±28.3	145.7±19.2	*157.0±11.7
Ditran	78.8±11.6	89.9±17.6	*95.2±10.3	72.6±13.9	83.6±16.8	**107.5±6.5	**102.9±6.7
Mescal-ine	102.2±10.3	91.2±12.0	110.6±5.2	105.9±0.6	*114.1±3.7	101.1±9.3	*97.9±4.5
LSD	94.8±10.6	93.0±8.4	*109.8±14.5	98.4±8.7	104.1±8.3	96.7±4.8	95.6±11.1

Table 4

The effects of phencyclidine (3mg/kg), Ditran (2mg/kg), mescaline (5mg/kg) and LSD (100microg/kg) on plasma tyrosine concentrations.

	TIME AFTER INJECTION (MINUTES)						
Drug b	0	10	20	40	60	100	180
Phency-clidine	65.8±11.3	*79.5±8.2	70.1±6.2	61.3±5.0	*52.2±4.5	62.6±4.1	60.2±2.9
Ditran	56.6±2.9	55.8±6.7	**47.1±5.9	***32.1±4.1	***37.2±6.9	***42.0±2.2	***51.4±1.6
Mescal-ine	58.9±2.2	***45.9±1.7	**54.1±0.9	**52.8±2.1	**48.1±5.1	***50.5±1.5	***44.8±1.8
LSD	56.3±1.6	***63.8±2.1	***40.9±1.4	***39.6±1.8	***30.3±1.0	***35.0±1.5	***40.8±1.7

Values in each table are the means of determinations from five animals, expressed as millimicromoles per gramme ± standard deviation.

P values are represented as $^{*}<0.05$, $^{**}<0.01$, $^{***}<0.001$.

The general pattern of changes is summarised in Table 5. In this table

+ = increased concentration

- = decreased concentration

Table 5

Summary table of changes in brain and plasma concentrations of tryptophan and tyrosine.

Maximum change in concentration of:	Hallucinogen			
	Phencyclidine (3mg/kg)	Ditran (2mg/kg)	Mescaline (5mg/kg)	LSD (100microg/kg)
Tryptophan				
Brain	-	-	-	+
Plasma	-	-	-	-
Tyrosine				
Brain	+	+	+	+
Plasma	-	-	-	-

Discussion

An adequate supply of amino acids to the CNS is essential if the biosynthesis of neurohumours is to proceed normally. There is general agreement that amino acids cross the blood-brain barrier by way of active transport mechanisms (Guroff, King and Udenfriend, 1961) specific for particular groups (Chirigos, Greengard and Udenfriend, 1960). Disturbances of amino acid metabolism or transport are known to be responsible for a number of mental disorders (Allan and Holt, 1965; Efron, 1965; Stanbury, Wyngaarden and Frederickson, 1966). The most characteristic feature of these aminoacidopathies in which cerebral dysfunctions (mental retardation, neurological defects and psychotic traits) occur is an elevation of the levels of particular amino acids in the plasma.

Imbalances in the concentrations of amino acids in the plasma may result in changes not only in the amino acids, but also in the amine levels of the brain. Changes may be brought about as a result of competition between the amino acid present in excess in the plasma and the precursors of the monoamines for the same transport system, producing a depletion of the monoamines. This situation is thought to occur in phenylketonuria, where the elevation of plasma phenylalanine is assumed to be at least partly responsible for the changes in cerebral monoamine levels (Zellweger, 1961). It has not yet been established whether any of the disorders of the CNS that occur in aminoacidopathies such as phenylketonuria, histidinemia, "maple sugar urine" disease and branched chain aminoaciduria can be attributed to effects on the monoamines of brain, but it is clear that amino acid and amine metabolism are closely interrelated.

Phencyclidine, Ditran and mescaline all caused depletion of endogenous tryptophan from brain tissue and from plasma in rats. The doses required to produce significant changes in the levels of this amino acid were lower than those required to produce an elevation of 5-HT or a depletion of NA (Tonge and Leonard, 1969; Leonard and Tonge, 1969). The period for which depletion persisted after the injection of the hallucinogen in each case corresponded well with the period during which behavioural effects were most intense. The fact that doses of hallucinogens that do not alter monoamine concentrations do affect behaviour and tryptophan levels suggests some interesting possibilities. The decreased concentrations of tryptophan found in both the cerebral tissues and in the blood plasma might reflect an increased conversion of the amino acid to 5-HT, with a consequent increase in the amount of the monoamine available for storage in the brain. Jequier, Lovenberg and Sjoerdsma (1967) have demonstrated that tryptophan hydroxylase may not be fully saturated with substrate under normal conditions and that the rate of 5-HT synthesis may be partially dependent on the availability of the substrate. Thus, the hallucinogens might produce elevations of 5-HT concentrations in the brain by making

tryptophan more accessible to the action of tryptophan hydroxylase. Experiments carried out by other investigators have shown that it is necessary to use large doses of tryptophan to produce a detectable change in cerebral 5-HT concentrations (McKean, Schanberg and Giarman, 1967; Weber, 1966). Therefore, if the effects of the hallucinogens on 5-HT levels have their origin in effects on the availability of tryptophan it is not surprising that relatively large doses of the drugs are required to produce detectable elevations in 5-HT concentrations.

An alternative explanation of the effects of phencyclidine, Ditran and mescaline on the concentrations of tryptophan and 5-HT might be that decreased levels of tryptophan inhibit the release of 5-HT. A decreased availability of tryptophan might lead to increased retention of 5-HT in the bound form. This situation might arise if the hallucinogens increased the metabolism of tryptophan through some other metabolic pathway, e.g. via the tryptophan pyrrollase system of the liver.

The finding that LSD produced an elevation of brain tryptophan levels whilst its other effects (elevation of brain 5-HT, depletion of brain NA and depletion of plasma tryptophan) were the same as those of the other three hallucinogens is difficult to explain. It can only be assumed that LSD either has a slightly different mode of action to the other hallucinogens tested, that it possesses some additional "tryptophan-releasing" activity, or that it decreases the turnover of tryptophan.

All four hallucinogens produced elevations of brain tyrosine concentrations, accompanied by decreased plasma levels of the amino acid. The elevated cerebral concentrations of tyrosine might arise from a feed-back inhibition of tyrosine hydroxylase by free NA, as the depletion of NA produced by hallucinogens appears to result from an increased release of the monoamine within the neurone (Leonard and Tonge, 1969). Stjarne, Lishajko and Roth (1967) have shown that very low concentrations of NA can cause an 85% inhibition of NA synthesis in a fraction from a homogenate of bovine splenic nerves.

Inhibition occurs at both tyrosine hydroxylation and the dopamine-β-hydroxylation stages. Thus, if the hallucinogens release NA in an active form within the neurones, the free amine might inhibit its own synthesis with a consequent build-up of the precursor tyrosine. Alternatively, both the increased cerebral levels and the decreased plasma levels of tyrosine might be interpreted as reflecting an increased turnover of NA in response to the hallucinogens, with increased uptake of tyrosine from the blood into the brain.

Further experiments are clearly required before it can be suggested which, if any, of these possibilities is correct. The most immediately relevant result of this investigation is the demonstration that four different hallucinogenic drugs produce both behavioural and biochemical effects in doses which do not affect the monoamine levels.

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