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Effect of Lithium on Central Metabolism of 5-Hydroxytryptamine

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Abstract. The administration of lithium carbonate for 5 days to rats increased the synthesis rate of brain serotonin, without modifying the brain level of the amine. This increase was not due to a modification of the free tryptophan in the blood. The level of serotonin and 5 HIAA remained unchanged in seven areas of brain. These results are discussed in comparison with the results of the other authors on the same subject.

Key words: Lithium — 5-Hydroxytryptamine — Brain.

Introduction

The efficiency of lithium in the treatment of mania (Cade, 1949) and the prophylaxis of manic depressive disorders (Schou and Shaw, 1973) is now well known and accepted. Many authors have looked for a biochemical explanation for this action and have studied particularly the effect of lithium on the metabolism of 5-hydroxytryptamine (5-HT) (review by Poitou, 1974a).

After lithium treatment, Andreoli *et al.* (1968) observed a slight increase in the level of serotonin in the brain, while Sheard and Aghajanian (1970), Perez-Cruet *et al.* (1971) and Schubert (1973) have shown an increase in the rate of synthesis of 5-HT.

However, Genefke (1972) did not observe any modification in the level of 5-HT after treatment of 5 weeks or less and, after this time, only the hypothalamic level was changed. Bliss and Ailion (1970) did not observe any alteration in the metabolism of 5-HT after prolonged treatment. On the contrary, Corrodi *et al.* (1969) found that chronic lithium (3 weeks) decreased the serotonin depletion caused by inhibition of tryptophan-hydroxylase. Ho *et al.* (1970) have shown a decrease in the 5-HT turnover in the hypothalamus.

During our study, we have used the same treatment as Perez-Cruet *et al.* (1971) and studied different parameters of 5-HT metabolism: total

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and free plasma tryptophan; tryptophan, 5-HT and 5-HIAA in the whole brain and various parts of the brain, and the decrease of 5-HIAA after pargyline administration.

Materials and Methods

Male Sprague-Dawley rats, weighing 200—250 g, received intraperitoneally twice a day, for 5 days, 2 mEq/kg of lithium chloride (5 ml/kg of a 0.4 M solution). Control rats received 5 ml/kg of a 0.4 M solution of sodium chloride.

The animals were stunned and killed 5 h after the last injection. Their blood was collected and centrifuged: the serum was used for the determination of total and free tryptophan, and lithium. The brains, or the seven parts of brains, were weighed and stored at −22° C until studied.

Total and free tryptophan in serum were assayed according to Hess and Udenfriend (1969). Total tryptophan was determined on 0.2 ml of serum and free tryptophan was obtained after serum filtration: 1 ml of serum was placed in a Centriflo membrane ultrafilter (CF 50 Amicon) and centrifuged at 1000 g for 30 min. Tryptophan was determined on 0.5 ml of ultrafiltrate.

A single brain, or a pool of two brain areas, was homogenized with acidified n-butanol. Tryptophan, 5-HT and 5-HIAA were determined on the same extract by the method of Curzon *et al.* (1972). Tryptophan and 5-HT were estimated in the aqueous phase after extraction from the butanol phase with 0.4 ml of 0.1 N HCl containing 0.1% cysteine and heptane; in order to extract 5-HIAA, the organic phase is shaken with 0.6 ml of 0.5 M phosphate buffer, and 5-HIAA was assayed on an aliquot of the aqueous phase. Tryptophan was estimated in 0.2 ml of the aqueous phase according to Hess and Udenfriend (1969) and 5-HT and 5-HIAA were assayed according to Curzon *et al.* (1970).

The rate od synthesis of 5-HT was calculated using the decrease of the 5-HIAA concentration after pargyline administration (75 mg/kg) (Tozer *et al.*, 1966).

Serum lithium was estimated by atomic absorption spectroscopy.

Values were analyzed for statistical significant differences by the Student's *t* test.

Results

The mean value $\pm$ SEM of serum lithium, on the fifth day, was 1.47 ± 0.20 mEq/l.

The other results are summarized in the tables. The total (protein bound and free) tryptophan was significantly increased in the plasma, but the free form remained unchanged by the lithium treatment (Table 1). The brain 5-HIAA level was also increased under these experimental conditions but not the brain 5-HT and tryptophan levels (Table 2).

The synthesis rate of 5-HT, as calculated by multiplying the slope of the 5-HIAA decline after inhibition of monoamine oxidase by pargyline by the 5-HIAA concentration at zero time, was also increased (Fig. 1 and Table 3).

Nevertheless, in all the seven parts of the brain studied, no modification of 5-HT and 5-HIAA could be shown (Table 4).

Table 1. Effect of lithium on total and free tryptophan in the serum of the rat

	Controls	Lithium
Free tryptophan μg/ml	6.96 ± 1.04 (10)	5.90 ± 0.60 (11)
Total tryptophan μg/ml	49.7 ± 1.4 (10)	64.0 ± 2.5* (9)

Numbers of animals are shown in parentheses.
* $P < 0.001$.

Table 2. Effect of lithium on brain tryptophan, 5-HT and 5-HIAA in the rat

	Controls	Lithium
Tryptophan μg/g	3.44 ± 0.14 (15)	3.89 ± 0.19 (16)
5-HT μg/g	0.74 ± 0.03 (20)	0.73 ± 0.05 (20)
5-HIAA μg/g	0.49 ± 0.02 (25)	0.63 ± 0.03* (25)

Numbers of animals are shown in parentheses.
* $P < 0.001$.

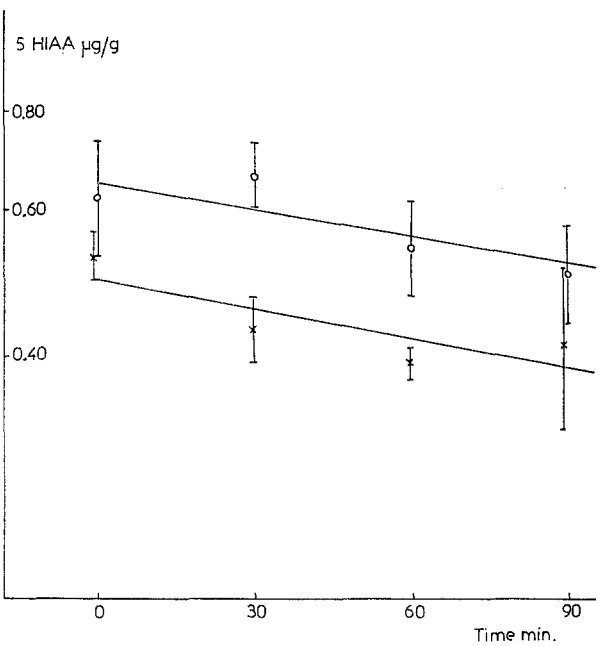

Fig. 1. Decrease of brain 5-HIAA after pargyline administration (75 mg/kg i.p.), 5 h after the last injection of LiCl. Each point is a mean + S.E. obtained from five animals. Lines were drawn by the method of least squares. o——o Lithium; x——x controls

Table 3. Effect of lithium on the synthesis rate of 5-HT in the rat brain

		Controls (19)[a]	Lithium (19)[a]
[5-HIAA] μg/g	0 min	0.52 ± 0.04	0.63 ± 0.09
	30 min	0.43 ± 0.04	0.66 ± 0.07
	60 min	0.39 ± 0.02	0.55 ± 0.08
	90 min	0.42 ± 0.09	0.50 ± 0.07
r[b]		−0.60	−0.54
Rate constant of 5-HIAA decline h^{-1}		0.17	0.16
$[5\text{-HIAA}]_0$ calculated		0.49	0.65
Synthesis rate of 5-HT μg/g/h		0.08	0.10

[a] Numbers in paretheses indicate numbers of animals.
[b] r is the correlation coefficient.

Table 4. Changes in 5-HT and 5-HIAA levels in regions of rat brain after lithium injection

Regions of rat brain	5-HT μg/g		5-HIAA μg/g	
	Controls	Lithium	Controls	Lithium
Hypothalamus (10)	2.48 ± 0.15	2.59 ± 0.10	2.38 ± 0.17	2.36 ± 0.17
Cerebellum (10)	0.34 ± 0.03	0.40 ± 0.04	0.41 ± 0.02	0.33 ± 0.03
Brain stem (10)	1.55 ± 0.07	1.47 ± 0.09	1.51 ± 0.08	1.78 ± 0.18
Hippocampus (10)	1.79 ± 0.15	1.46 ± 0.04	1.90 ± 0.17	2.08 ± 0.20
Caudate (10)	2.01 ± 0.15	1.81 ± 0.17	1.89 ± 0.16	1.94 ± 0.12
Olfactory bulb (10)	1.37 ± 0.12	1.22 ± 0.17	1.03 ± 0.08	1.04 ± 0.08
Cortex (10)	0.43 ± 0.02	0.38 ± 0.03	0.37 ± 0.05	0.42 ± 0.03

Numbers of animals are shown in parentheses.

Discussion

These data suggesting that lithium increases the synthesis rate and catabolism of serotonin are in agreement with those of Sheard and Aghajanian (1970), Perez-Cruet *et al.* (1971) and Schubert (1973). Tagliamonte *et al.* (1971 a, b) suggested that lithium increases the synthesis rate by increasing plasma tryptophan; but, like Schubert (1973), we found that free plasma tryptophan was not modified, and it is more probable that lithium acts by increasing the transport of tryptophan to the brain. Lithium action probably differs from the action of drugs which increase brain serotonin synthesis by displacing tryptophan from its binding with plasma proteins (Guerinot *et al.*, 1974). Moreover Knapp and Mandell (1973) have shown that lithium modified the synaptosomal uptake of tryptophan. Perez-Cruet *et al.* (1971) indicated that lithium may increase the serotonin synthesis by increasing the brain tryptophan. Considering our results, it may be thought that lithium increases the

transfer rate of tryptophan to the brain, without modification of brain tryptophan level, because of an increased utilisation. Schubert (1973) found that lithium had no effect on total plasma tryptophan, without altering the level of 5-HT and 5-HIAA. These differences may be due to the difference between the serum lithium levels which are twice as high in the present experiment, in which the stimulation of serotonin synthesis may be greater.

These biochemical data are, moreover, in agreement with other clinical or pharmacological results. Thus Wilson and Prange (1972) have shown that tryptophan is efficient, like lithium, in treatment of mania; Poitou *et al.* (1974b) found that lithium and tryptophan had a similar action on the amphetamine-chlordiazepoxide induced hyperactivity in mice. Mendels and Chernik (1973) have observed that lithium had an effect similar to tryptophan on the sleep of depressed patients.

However, these results are not in agreement with those of Corrodi *et al.* (1969). The length of administration may explain this difference, because Corrodi *et al.* have given lithium during 3 weeks. Indeed, Knapp and Mandell (1973) have shown that the lithium administration decreased the activity of tryptophan-hydroxylase from the fifteenth day.

In this study, no modification of the level of 5-HT and 5-HIAA in different parts of brain was found. However, the synthesis rate of serotonin might have been increased in some areas, without significant modification of the level of the mediator and its catabolite. Sensitive isotopic technics could be used to verify this hypothesis.

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