Comparative simultaneous measurement of cerebrospinal fluid 5-hydroxyindoleacetic acid and blood serotonin levels in delirium tremens and clozapine-induced delirious reaction

CSABA M. BANKI AND M. VOJNIK

From the Department of Psychiatry, The County Neuropsychiatric Institute, Nagykálló, Hungary

SUMMARY Cerebrospinal fluid 5-hydroxyindoleacetic acid and total blood serotonin levels were measured simultaneously in 11 female patients with delirium tremens and nine schizophrenic women with clozapine-induced acute delirium. Both groups had significantly raised levels of 5HIAA in CSF and significantly reduced blood 5HT levels as compared with normal control subjects, symptom-free alcoholics, or clozapine-treated schizophrenics. The two delirious groups were not distinguishable from each other in respect of their CSF 5HIAA or blood 5HT values. After clinical recovery both values returned to normal levels.

There is strong evidence that acute and chronic ethanol consumption can disturb neurotransmitter, and in particular serotonin (5HT), metabolism (Eccleston *et al.*, 1967; Badawy and Evans, 1974). Acetaldehyde, rather than ethanol itself, has been suggested to divert the indoleamine catabolism from the normal predominating oxidative to a reductive pathway (Feldstein, 1969, 1973), and there are many indications that this metabolic shift may lead to formation of biologically highly active alkaloidlike condensation products (Dajani and Saheb, 1973; Majchrowitz, 1973; Walsh, 1973). During chronic ethanol ingestion, tolerance develops to its effects on the central nervous system. Its underlying biochemical mechanism is, however, still unclear; transmitter turnover studies and measurements of enzymatic activity have yielded controversial results (Redmond and Cohen, 1971; Raskin and Sokoloff, 1974). It seemed useful, therefore, to obtain data about serotonin metabolism in delirium tremens, especially in female patients as there is a very marked increase of alcoholism in women in

Hungary. While in the period 1960–69 only a single case of delirium tremens was admitted to hospital, there were two in 1970, and 26 in 1976. In the past seven months of 1977 19 women were hospitalised for delirium tremens.

Clozapine, a special type of neuroleptic drug, has been reported to affect 5HT metabolism (Ackenheil *et al.*, 1974; Bürki *et al.*, 1975); although it has negligible extrapyramidal side effects, a delirious reaction can occur in certain patients (Gabriel *et al.*, 1976). In an earlier study (Banki, 1977b) we demonstrated a transient elevation of 5-hydroxyindoleacetic acid (5HIAA) in the CSF and a progressive increase of blood 5HT content during clozapine administration, where delirious reactions were associated with pronounced indoleamine changes; this prompted us to include clozapine-induced delirium in this study.

Total blood 5HT content corresponds almost totally to platelet 5HT stores (Berstad, 1976) which, in turn, may serve as a peripheral model for central serotonergic neurones (Pletscher, 1968; Murphy and Costa, 1975). Measurement of 5HIAA in the CSF, on the other hand, serves as an index of CNS serotonin turnover. In this way, the simultaneous measurement of these two variables seemed likely to yield more information on 5HT metabolism than the separate determination of either of these metabolites.

Address for reprint requests: Dr C. M. Banki, Department of Psychiatry, The County Neuropsychiatric Institute, H-4321 Nagykálló 18, Hungary.

Accepted 20 December 1977

Subjects and methods

Eleven female patients with manifest delirium tremens and nine with clozapine delirium were studied; their ages were similar (average 39.5 and 42.0 years, respectively). The diagnosis of delirium tremens was established according to standard criteria (Reis, 1973) including the requirements for chronic alcoholism (Criteria Committee, 1972). The prominent symptoms were disturbed consciousness, disorientation, perception disorders, gross tremor, ataxia, sweating, anxiety, and agitation (less frequently euphoric-hyperthymic state). Clozapine-induced delirium was in many aspects similar to delirium tremens: hallucinations, confusion, automatic movements, and disorientation were identical, but tremor, vasodilatation, hypertension, and so on were absent.

Nine patients with delirium tremens were hospitalised for the first time, and one had her fourth delirium. Symptoms usually started 18 to 48 hours before admission. The last alcohol intake took place between six and 72 hours (average 36 hours) before arriving at the hospital. The patients with clozapine-induced delirium were all schizophrenic, admitted two to five days earlier, and receiving only clozapine in an average daily dose of 200 mg. The examination took place six to 18 hours after delirium had begun.

From all patients 5 ml heparinised blood and 5 ml CSF were taken (the latter by lumbar puncture) simultaneously, after careful medical examination. Delirium tremens patients were investigated immediately after admission, clozapine-delirium patients at the time when symptoms developed fully. At this time the interval since the last clozapine intake was between six and 12 hours. The delirium tremens patients were drug-free at this first sample taking.

Both kinds of delirium were treated with intravenous chlormethiazol (Heminevrin Astra), followed when necessary by oral medication, and supplemented with antibiotics, fluid therapy, vitamins, magnesium, and potassium (Hudolin *et al.*, 1973; Sattes, 1973). All patients with delirium tremens recovered substantially within three to six days. Clozapine-induced delirium never lasted for longer than 24 hours; these schizophrenic patients were then treated with other neuroleptics or ECT or both.

Blood and CSF samples were mixed with 0.05 ml 0.3% ascorbic acid and immediately frozen to -20°C . Laboratory assay took place within six days by the O-phthalaldehyde fluorescent method (Curzon and Green, 1970; Geeraerts *et al.*, 1974).

After clinical recovery (usually 18–22 days)

further simultaneous blood and CSF samples were taken from all patients. Results were compared with the blood 5HT and CSF 5HIAA data obtained from groups of symptom-free alcoholic women, schizophrenic women under clozapine-treatment, and female patients without major psychiatric illness who were hospitalised for hysteria, neurotic, or other conflict behaviour, character disturbance, or other personality problems. Average age was similar in all the groups (44 years for alcoholics, 37 years for schizophrenics, and 41.4 years for control subjects).

Statistical analysis was done using analysis of variance and Student's *t* test (two-tailed).

Results

Table 1 shows 5HIAA levels in CSF in the patient groups. Alcoholics had the same average levels as control subjects, but the delirium tremens and clozapine-delirium groups both had significantly higher values though they did not differ statistically from each other. Clozapine-treated schizophrenic patients who had no delirium also showed slightly but significantly raised average 5HIAA levels.

Table 1 5HIAA levels in CSF of patients with delirium tremens, clozapine-induced delirium, and alcoholism, and in control subjects

	Number	5HIAA ng/ml $\bar{x} \pm \text{SEM}$	Range	Analysis of variance
1 Delirium tremens	11	67.0 ± 3.6	50–87	F = 28.63 P = < 0.001
2 Clozapine-delirium	9	70.7 ± 2.8	59–85	
3 Clozapine-treated*	8	40.0 ± 4.4	20–64	
4 Alcoholic	16	28.9 ± 2.7	12–47	
5 Control	12	26.5 ± 1.9	16–39	

*Schizophrenic patients without delirious reaction.

Groups 1 and 2 both differ significantly from either control subjects or alcoholics; group 3 levels are significantly higher ($P < 0.05$) than control values. Groups 1, 2 and 4, 5 do not differ statistically from each other.

Blood serotonin content showed a different pattern (Table 2). Here both delirious groups had marked *lower* average values as compared with either control subjects, alcoholics, or schizophrenics under clozapine; in addition, this last group had *higher* blood 5HT levels as compared with controls—that is, changes were in the opposite direction. However, this increase in blood 5HT was not statistically significant. Delirium tremens and clozapine-delirium patients had practically identical average levels; symptom-free alcoholics also tended to have a lower blood 5HT content, but this was not statistically significantly different from control subjects.

Three weeks later, after clinical recovery, all

Table 2 Total blood serotonin content in the patient subgroups

	Number	5HT ng/ml $\bar{x} \pm \text{SEM}$	Range	Analysis of variance
1 Delirium tremens	11	80 $\pm$ 6.9	49–130	F = 10.13 P = < 0.01
2 Clozapine-delirium	9	81 $\pm$ 7.5	50–113	
3 Clozapine-treated*	8	207 $\pm$ 17.0	114–284	
4 Alcoholic	16	143 $\pm$ 16.2	66–302	
5 Control	12	178 $\pm$ 12.5	128–266	

*As in Table 1.

Groups 1 and 2 do not differ statistically from one another, but both are significantly lower as compared with groups 3, 4, or 5. None of the latter differ statistically.

values showed a clear tendency toward the normal: clozapine-delirium patients had both CSF 5HIAA and blood 5HT values within the normal range, delirium tremens patients approached the levels found in alcoholics—that is, a lower blood 5HT content and a tendency toward elevated CSF 5HIAA (Table 3).

Discussion

Although every biochemical measurement carried out in either blood or CSF can give only indirect information about changes in the CNS and must, therefore, be interpreted with caution, the above data seem to indicate that indoleamine metabolism is altered profoundly in both types of delirious states. The concurrent decrease of the stored 5HT in the blood platelets together with the 2.5 fold increase in its metabolite 5HIAA in the CSF would suggest an increased release and turnover of the transmitter amine. However, some of the following points may also be relevant. Firstly, hepatic insufficiency, a common finding in delirium tremens which has also been speculated on as a copathogenetic factor (Wilson, 1973), may be associated with raised 5HIAA levels in CSF (Young *et al.*, 1974) or in brain (Knott and Curzon, 1975). This, however, would not account for the high values in clozapine-induced delirium. Secondly, delirious patients usually have excessive

motor activity which may influence their CSF amine metabolite levels (Post *et al.*, 1973). We also found a positive correlation between HVA (but not 5HIAA) levels in CSF and psychomotor activity (Banki, 1977a). This possibility has still to be elucidated. Thirdly, ethanol may impair the removal process of 5HIAA from the CNS, probably by inhibiting Na⁺-K⁺-activated ATPase in the choroid plexus membrane (Tabakoff and Boggan, 1974; Tabakoff *et al.*, 1975). This possibility can be tested by the probenecid technique, and deserves special attention. Finally, blood serotonin content reflects peripheral changes which are not necessarily parallel to central processes; in addition, a decreased 5HT storage may also be due to nonspecific stress-response.

The most interesting result of this study is that, in clozapine-induced delirium, indoleamine changes were virtually identical to those observed in delirium tremens. Clozapine causes elevation of CSF 5HIAA in patients, and of brain 5HT and 5HIAA in animals (Ackenheil *et al.*, 1975; Stralendorff *et al.*, 1976), either by increasing 5HT synthesis or by indirect raphe depression (Gallagher and Aghajanian, 1976a, b). In psychiatric patients Banki (1977b) showed that elevation of 5HIAA in the CSF is only transient, while increased 5HT storage—as reflected by the total blood 5HT content—develops gradually but is prolonged. At present we cannot explain why in certain patients indoleamine metabolism responds in a different way, and whether this difference may be related to the clinical picture of delirium. However, the clinical observation that alcoholics are particularly sensitive to the delirium-inducing potency of clozapine (Banki, 1976; Gabriel *et al.*, 1976) suggests a link between delirium tremens and clozapine-induced delirium. Both states are characterised by excessive activation, while withdrawal from chronic ethanol intoxication leads to greatly enhanced release of noradrenaline (Carlsson and Haeggendal, 1967) and altered noradrenaline-receptor sensitivity; at the same

Table 3 5HIAA levels in CSF and blood serotonin levels in delirium tremens and clozapine-delirium before and after clinical recovery

	Subgroup	Before recovery	After recovery	Difference
CSF 5HIAA (ng/ml)	Delirium tremens	67 $\pm$ 3.6	36 $\pm$ 3.4*	31 $\pm$ 5.6‡
	Clozapine-induced delirium	71 $\pm$ 2.8	30 $\pm$ 3.0†	41 $\pm$ 5.4‡
Blood 5HT (ng/ml)	Delirium tremens	80 $\pm$ 6.9	133 $\pm$ 10.2*	53 $\pm$ 17.6§
	Clozapine-induced delirium	81 $\pm$ 7.5	156 $\pm$ 14.4†	75 $\pm$ 18.8

Values expressed as $\bar{x} \pm \text{SEM}$.*Not significant versus alcoholics, significant versus normal control subjects ($p < 0.02$ and < 0.01 for CSF 5HIAA and blood 5HT, respectively).

†Not significant versus normal controls.

‡($p < 0.001$).§($p < 0.02$).||($p < 0.01$).

time clozapine has been found to act primarily at these receptors (Gallagher and Aghajanian, 1976). Our further studies will be aimed at investigating these possibilities.

References

- Ackenheil, M., Beckmann, H., Hoffmann, G., and Markianos, E. (1974). Einfluss von Clozapin auf die MHPG- NHS- und 5HIES-Ausscheidung in Urin und Liquor. *Arzneimittel-Forschung*, **24**, 984–987.
- Ackenheil, M., Blatt, B., and Lampart, L. (1975). Effect of clozapine on 5HIAA excretion in urine and CSF of psychotic patients and on serotonin metabolism in rats. *Acta Vitaminologica et Enzymologica (Milano)*, **29**, 79–84.
- Badawy, A. A., and Evans, M. (1974). Alcohol and tryptophan and metabolism. *Journal of Alcoholism*, **9**, 97–116.
- Banki, C. M. (1976). 5HIAA level in the CSF of manic and depressed patients. *Ideggyógyászati Szemle*, **29**, 467–479. In Hungarian.
- Banki, C. M. (1977a). Correlation between CSF amine metabolites and psychomotor activity in affective disorders. *Journal of Neurochemistry*, **28**, 255–257.
- Banki, C. M. (1977b). Influence of clozapine on CSF-5HIAA and blood 5HT content in psychiatric patients. *Psychopharmacologia*. In press.
- Berstad, J. (1976). Total 5-hydroxyindoles in blood related to migraine attacks. *Acta Neurologica Scandinavica*, **54**, 293–300.
- Bürki, H. R., Ruch, W., and Asper, H. (1975). Effect of clozapine, thioridazine, perlapine and haloperidol on the metabolism of biogenic amines in the brain of rat. *Psychopharmacologia*, **41**, 27–32.
- Carlsson, D., and Haeggendal, J. (1967). Arterial norepinephrine level after ethanol withdrawal. *Lancet*, **2**, 889.
- Criteria Committee of the National Council on Alcoholism (1972). Criteria for the diagnosis of alcoholism. *American Journal of Psychiatry*, **129**, 127–135.
- Curzon, G., and Green, A. R. (1970). Rapid method for the determination of 5HT and 5HIAA in small regions of rat brain. *British Journal of Pharmacology*, **39**, 653–655.
- Dajani, R. M., and Saheb, S. E. (1973). A further insight into the metabolism of certain beta-carbolines. *Annals of the New York Academy of Sciences*, **215**, 120–123.
- Eccleston, D., Reading, W. H., and Ritchie, I. M. (1967). Serotonin metabolism in brain and liver slices and the effect of ethanol. *Journal of Neurochemistry*, **16**, 274–276.
- Feldstein, A. (1969). The effect of ethanol on in vivo conversion of 5HTP to 5HT, 5HTOL and 5HIAA. *International Journal of Neuropsychopharmacology*, **8**, 347–351.
- Feldstein, A. (1973). Ethanol-induced sleep in relation to 5HT turnover. *Annals of the New York Academy of Sciences*, **215**, 71–76.
- Gabriel, E., Küfferle, G., and Lenz, D. (1976). Über individuellen Bedingungen pharmakogener Verwirrtheiten. *Psychiatria Clinica*, **9**, 5–13.
- Gallagher, D. W., and Aghajanian, G. K. (1976a). Effect of antipsychotic drugs on the firing of dorsal raphe cells. *European Journal of Pharmacology*, **39**, 341–355.
- Gallagher, D. W., and Aghajanian, G. K. (1976b). Effect of antipsychotic drugs on the firing of dorsal raphe cells II. *European Journal of Pharmacology*, **39**, 357–364.
- Geeraerts, F., Schimpffessel, L., and Crokaert, R. (1974). A simple routine method to preserve and determine blood 5HT. *Experientia*, **30**, 837–839.
- Hudolin, V., Kalousek, M., and Thaller, V. (1973). Chlormethiazol in the treatment of alcoholism. *Alcoholism*, **9**, 71–81.
- Knott, P. J., and Curzon, G. (1975). Tryptophan and tyrosine disposition and brain tryptophan metabolism in acute carbontetrahydrochloride poisoning. *Biochemical Pharmacology*, **24**, 963–966.
- Majchrowitz, E. (1973). Alcohols, aldehydes and biogenic amines. *Annals of the New York Academy of Sciences*, **215**, 84–88.
- Murphy, D. L., and Costa, E. (1975). Utilisation of cellular studies of neurotransmitter related enzymes. In *Psychobiology of Depression*. Edited by J. Mendels. Spectrum Publications: New York.
- Pletscher, A. (1968). Metabolism, transfer and storage of 5HT in blood platelets. *British Journal of Pharmacology*, **32**, 1–16.
- Post, R. M., Kotin, J., and Goodwin, F. K. (1973). Psychomotor activity and CSF amine metabolites in affective illness. *American Journal of Psychiatry*, **130**, 67–72.
- Raskin, N. H., and Sokoloff, L. (1974). Changes in brain ADH activity during chronic ethanol ingestion and withdrawal. *Journal of Neurochemistry*, **22**, 427–434.
- Redmond, G., and Cohen, A. (1971). Induction of liver aldehyde dehydrogenase and possible role in ethanol tolerance. *Science*, **171**, 387–389.
- Reis, D. J. (1973). A possible role of central noradrenergic neurons in withdrawal states from alcohol. *Annals of the New York Academy of Sciences*, **215**, 249–252.
- Sattes, H. (1973). Chlormethiazol treatment of delirium tremens. *Alcoholism*, **9**, 111–115.
- Stralendorff, B., Ackenheil, M., and Zimmermann, J. (1976). Akute und chronische Wirkung von Caripramin, Clozapine und Haloperidol auf den Stoffwechsel biogener Amine im Rattenhirn. *Arzneimittel-Forschung*, **26**, 1096–1098.
- Tabakoff, B., and Boggan, W. O. (1974). Effect of ethanol on serotonin metabolism in brain. *Journal of Neurochemistry*, **22**, 759–764.
- Tabakoff, B., Ritzmann, R. F., and Boggan, W. O. (1975). Inhibition of the transport of 5HIAA from brain by ethanol. *Journal of Neurochemistry*, **24**, 1043–1051.
- Walsh, J. M. (1973). Biogenesis of biologically active alkaloids from amines by alcohol and acetaldehyde.

- Annals of the New York Academy of Sciences*, **215**, 98–110.
- Wilson, G. C. (1973). Management of postoperative and posttraumatic delirium due to alcoholism. *Alcoholism*, **9**, 116–119.
- Young, S. N., Gorelis, E., and Lal, S. (1974). Tryptophan and 5HIAA in human CSF. *Journal of Neurochemistry*, **22**, 772–776.