4. S. W. Holmes and E. W. Horton, *J. Physiol., Lond.* **195**, 731 (1968).
5. E. W. Horton, *Br. med. Bull.* **29**, 148 (1973).
6. P. W. Ramwell and J. E. Shaw, *Am. J. Physiol.* **211**, 125 (1966).
7. P. B. Bradley, G. M. R. Samuels and J. E. Shaw, *Br. J. Pharmac. Chemother.* **37**, 151 (1969).
8. F. Coceani and L. S. Wolfe, *Can. J. Physiol. Pharmac.* **43**, 445 (1965)
9. S. W. Holmes, *Br. J. Pharmac. Chemother.* **38**, 653 (1970).
10. W. Feldberg and K. P. Gupta, *J. Physiol. Lond.* **228**, 41 (1973).
11. D. B. Beleslin, B. Z. Radmanovic and M. M. Rakic, *Brain Res.* **35**, 625 (1971).
12. J. R. Vane, *Nature New Biol.* **231**, 232 (1971).
13. S. H. Ferreira, S. Moncada and J. R. Vane, *Nature New Biol.* **231**, 237 (1971).
14. B. V. Caldwell, L. Speroff, W. A. Brock, F. J. Auletta, J. W. Gordon, G. G. Anderson and J. C. Hobbins, in *The Prostaglandins, Clinical Applications in Human Reproduction* (Ed. E. M. Southern), p. 257. Futura, Mt. Kisco, New York (1972).
15. J. Nakano, A. V. Pracan and S. E. Moore, *Brain Res.* **39**, 545 (1972).
16. S. W. Holmes and E. W. Horton, *Br. J. Pharmac. Chemother.* **34**, 32 (1968).
17. P. Hedqvist, *Acta physiol. scand.*, **345**, *Suppl.* (1970).
18. W. B. Essman, *Neurochemistry of Cerebral Electrochock*, p. 95, Spectrum, New York (1973).
19. D. R. Haubrich, J. Perez-Cruet and W. D. Reid, *Br. J. Pharmac. Chemother.* **48**, 80 (1973).

Biochemical Pharmacology, Vol. 24, pp. 2103–2104. Pergamon Press, 1975. Printed in Great Britain.

Effect of acute and chronic iprindole on serotonin turnover in mouse brain*

(*Received* 9 *October* 1974; *accepted* 14 *March* 1975)

Iprindole, 5-(3-dimethylaminopropyl)-6, 7, 8, 9, 10, 11-hexahydro-5H-cyclooct [b] indole hydrochloride (WY-3263), a tricyclic compound, has been shown to be an effective clinical antidepressant agent [1–6]. It was further reported that it has a very weak inhibitory effect on the uptake of norepinephrine, dopamine and serotonin in rat brain tissue [7–10]. It also failed to alter the serotonin content of human platelets [11, 12]. Recently it has been reported that iprindole, after acute or chronic administration, had no effect on rat brain norepinephrine metabolism [13]. Clinically, it has been found to be a more potent antidepressant than imipramine [3–5]. All studies on its mode of action indicate that the mechanism of antidepressant effect of iprindole is not related to its ability to inhibit the neuronal uptake of biogenic amines.

It is well established that all clinically useful antidepressant agents require several days of administration before their beneficial effects are manifest. With this view in mind, we have investigated the effect of acute and chronic administration of iprindole on the turnover of serotonin (5-HT) in mouse brain.

Swiss–Webster male mice weighing 20–22 g were housed five in a cage at a controlled temperature of 22° and constant humidity. The light cycle was set at 12 hr of dark and 12 hr of bright light. The animals had free access to food and water. In acute experiments, one group of mice was given iprindole, 20 mg/kg intraperitoneally, and another group received an equivalent volume of 0·2 ml saline by the same route. In chronic experiments, iprindole (20 mg/kg i.p.) or saline was administered once daily for 3 weeks.

Turnover rate of 5-HT was determined by measuring the decline in 5-HIAA levels and the accumulation of 5-HT at various time intervals after pargyline (75 mg/kg, i.p.) according to the method of Tozer *et al.* [14]. Brain 5-HT and 5-HIAA were determined by the fluorometric procedures of Kuntzman *et al.* [15] and Curzon and Green [16] respectively. The values for brain 5-HIAA levels were logarithmically transformed for calculation of linearity of regression, standard error of the regression coefficients and significance of the difference between regression coefficients [17]. The values for 5-HT were statistically analyzed in the same manner without the logarithmic transformation.

Iprindole, in a dose of 20 mg/kg i.p., administered acutely or chronically for 3 weeks had no significant effect on the rate of 5-HT synthesis or turnover time calculated from the rate of decline in 5-HIAA levels after pargyline (Fig. 1, Table 1), although it slightly increased the rate of 5-HT synthesis and caused a small reduction in its turnover time (Table 1). There was no significant effect of iprindole, whether given acutely or chronically, on the rate of 5-HT accumulation after pargyline (Fig. 2).

The methods used in this study to measure the turnover of 5-HT are based on certain assumptions such as complete inhibition of monoamine oxidase, inability of 5-HT to pass from brain to blood at an appreciable rate, the main catabolic pathway for 5-HT is oxidative deamination and lastly that MAO inhibitor does not affect 5-HT metabolism other than by blockade of MAO. Based on these assumptions, at steady state, the level of brain 5-HT is constant, as a result of equal rates of synthesis and efflux. Similarly the level of 5-HIAA is constant because of equal rates of formation and loss [14]. Data presented here sug-

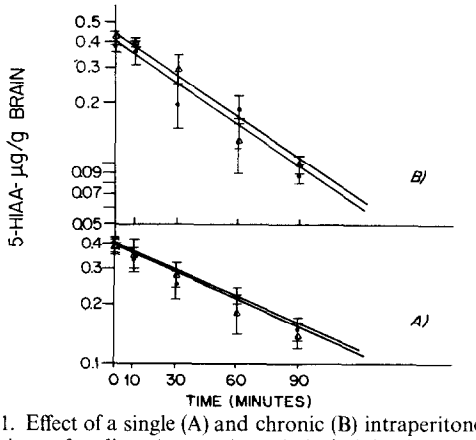

Fig. 1. Effect of a single (A) and chronic (B) intraperitoneal injection of saline (●----●) and iprindole 20 mg/kg (△----△) on 5-HIAA decline in mouse brain at various times after pargyline, 75 mg/kp i.p. Vertical bars represent the standard error of the mean. In Figs. 1 and 2, the method of least squares was used to calculate the best fit line.

* This study was supported by United States Public Health Service Grants MH 08618 and MH 04669.

Table 1. Effect of iprindole on the turnover rate and turnover time of serotonin measured from the 5-HIAA decline after monoamine oxidase inhibition (MAOI)

Treatment	Brain levels of 5-HIAA before MAO blockade* (μg/g ± S.E.)	Rate constant of 5-HIAA loss after MAOI [k(hr^{-1} ± S.E.]	5-HT turnover time (min)	Turnover rate of 5-HT† (μg/g/hr)
Acute:				
Saline + pargyline	0·40 ± 0·02 (15)	0·69 ± 0·02	87	0·25
Iprindole + pargyline	0·39 ± 0·04 (15)	0·69 ± 0·02	87	0·25
Chronic (3 weeks)				
Saline + pargyline	0·40 ± 0·04 (15)	0·69 ± 0·05	87	0·25
Iprindole + pargyline	0·44 ± 0·03 (15)	0·74 ± 0·05	80	0·30

* Figures in parentheses refer to number of animals in each group.

† Product of level and rate constant (k) of 5-HIAA decline. Correction has been made for the difference in molecular weights.

gest that, at steady state, iprindole does not appear to alter significantly the metabolism of mouse brain 5-HT. The biogenic hypothesis of depression suggests that tricyclic antidepressant agents alleviate depression by inhibition of the biogenic amine re-uptake pump in the neuronal membrane, thus making more amine available in the synaptic cleft to affect the post-synaptic receptor sites [18, 19]. Several studies have shown that, after acute administration, iprindole had no effect on the uptake mechanism of norepinephrine in heart and brain tissue of the rat [7–10]. Recently, it has been reported that iprindole, after acute or chronic administration, failed to effect significantly noreprinephrine metabolism of rat brain [13]. Furthermore, it did not inhibit appreciably the uptake of radioactive 5-HT or dopamine in rat brain [10, 13]. Present data would suggest indirectly that iprindole had no effect on the uptake mechanism of 5-HT (as measured by the level and turnover rate). It is conceivable that iprindole produces its effect by inhibition of monoamine oxidase. However, it had no significant effect on brain norepinephrine levels after acute injection or on brain or liver monoamine oxidase activity at a concentration of 1×10^{-3} M [7]. The effect of chronic administration of iprindole on monoamine oxidase remains to be elucidated. Partial indirect evidence showing its probable effect on monoamine oxidase was presented recently [13]. Our data also indicate that it may have some inhibitory effect on monoamine oxidase, especially after chronic administration (Fig. 1). Further experiments are needed for direct evidence to verify this possibility. From the data presented here, it can be concluded that iprindole does not affect brain 5-HT metabolism.

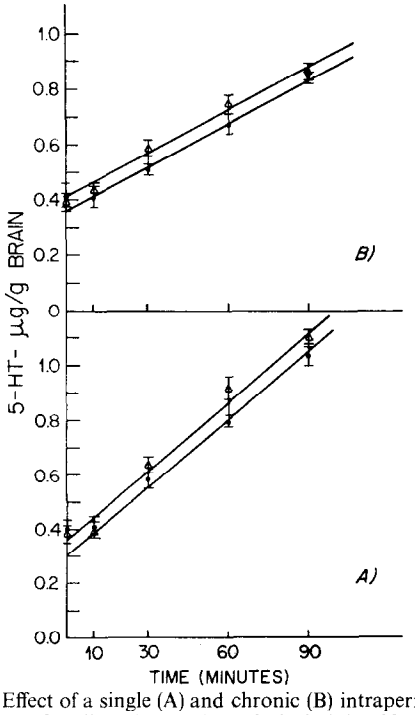

Fig. 2. Effect of a single (A) and chronic (B) intraperitoneal injection of saline (●----●) and iprindole 20 mg/kg (△----△) on serotonin (5-HT) accumulation in mouse brain at various times after pargyline, 75 mg/kg i.p. Vertical bars represent the standard error of the mean.

Neuropsychopharmacology Research Unit, Department of Psychiatry, New York University Medical Center, 550 First Avenue, New York, N.Y. 10016, U.S.A.

INDRAVADAN SANGHVI
SAMUEL GERSHON

REFERENCES

1. J. T. Hicks, *Illinois med. J.* **128**, 622 (1965).
2. E. A. Daneman, *Psychosomatics* **8**, 216 (1967).
3. W. T. McClatchey, J. Moffat and G. M. Irvine, *J. Ther. clin. Res.* **5**, 13 (1967).
4. J. Johnson and H. G. Maden, *J. clin. Trials* **4**, 787 (1967).
5. N. K. Eldeiry, A. D. Forrest and S. K. Littman, *Br. J. Psychiat.* **113**, 999 (1967).
6. A. D. Clift, *Practitioner* **205**, 89 (1970).
7. M. I. Gluckman and T. Baum, *Psychopharmacologia* **15**, 169 (1969).
8. L. Lemberger, E. Sernatinger and R. Kuntzman, *Biochem. Pharmac.* **19**, 321 (1970).
9. R. A. Lahti and R. P. Maickel, *Biochem. Pharmac.* **30**, 482 (1971).
10. S. B. Ross, A. L. Renyi and S. -O. Organ, *Life Sci.* **10**, 1267 (1971).
11. W. E. Fann, J. M. Davis, D. S. Janowsky, J. S. Kaufmann, J. D. Griffith and J. A. Oates, *Arch. gen. Psychiat.* **26**, 158 (1972).
12. I. C. Campbell and A. Todrick, *Br. J. Pharmac. Chemother.* **49**, 279 (1973).
13. B. N. Rosloff and J. M. Davis, *Psychopharmacologia* **40**, 53 (1974).
14. T. M. Tozer, N. H. Neff and B. B. Brodie, *J. Pharmac. exp. Ther.* **153**, 177 (1966).
15. R. Kuntzman, P. A. Shore, D. Bogodanski and B. B. Brodie, *J. Neurochem.* **6**, 226 (1961).
16. G. Curzon and A. R. Green, *Br. J. Pharmac. Chemother.* **39**, 653 (1970).
17. H. C. Batson, *An Introduction to Statistics in Medical Sciences*, p. 56. Burgess, Minneapolis (1965).
18. J. M. Davis, *Int. Rev. Neurobiol.* **12**, 145 (1970).
19. J. J. Schildkraut, *Neuropsychopharmacology and the Affective Disorders*, p. 24. Little Brown, Boston (1970).