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A COMPARISON OF THE ACUTE AND CHRONIC EFFECTS OF FOUR ANTIDEPRESSANT DRUGS ON THE TURNOVER OF SEROTONIN, DOPAMINE AND NORADRENALINE IN THE RAT BRAIN

B. E. LEONARD* and W. F. KAFOE†
Pharmacology Department, Organon International B. V., Oss, The Netherlands

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Abstract—The acute effects of imipramine, Org GB 94 and iprindol and the chronic effects of imipramine, desipramine, iprindol and Org GB 94 were studied on the turnover of dopamine, noradrenaline and serotonin in the whole rat brain. The effects of these drugs on amine turnover was assessed from the incorporation of tritiated tyrosine and tryptophan into the amines. Following acute administration, imipramine significantly reduced the turnover of serotonin whereas Org GB 94 increased that of noradrenaline. After chronic administration, imipramine and desipramine reduced the turnover of serotonin and dopamine, iprindol had no significant effect on brain amine turnover whereas Org GB 94 increased the turnover of noradrenaline. The effect of Org GB 94 on noradrenaline turnover was more marked after intraperitoneal than after oral administration.

It is widely assumed that the clinical efficacy of the dibenzazepine antidepressants of the imipramine type is largely due to their ability to reduce the reuptake of noradrenaline and/or serotonin into nerve endings thereby increasing the effective concentration of these amines at central receptor sites [1, 2]. Presumably such an effect helps to correct the relative deficiency of these transmitter substances in the brains of depressive patients [3, 4].

It is well established that a period of 7–14 days is normally required from the commencement of antidepressant therapy before the drug shows any beneficial effect [5]. As most experimental studies on the mechanism of action of these drugs appear to have been undertaken following the acute administration, it is possible that the results obtained from such studies may not reflect the neurochemical changes which occur following the chronic administration of the drugs. The present investigation was therefore undertaken to see what effect imipramine, desipramine, iprindol and the novel antidepressant drug Org GB 94 [6] have on the turnover of brain biogenic amines following their chronic administration. Some preliminary results of this study have been communicated elsewhere [7].

METHODS

In the first experiment, the acute effects of Org GB 94, iprindol and imipramine were compared. Groups of male Wistar rats (80–90 g) were treated with either Org GB 94 (10 mg/kg i.p.) or imipramine (120 mg/kg i.p.) or with iprindol 40 mg/kg i.p.) for 150 min. These doses were less than 1/5 of the acute LD_{50} [20–22]; the durations of treatment approximately coincided with the times for the peak drug effect on amine metabolism. The control group was injected with physiological saline. Exactly 40 min before the animals were killed by decapitation, all rats were injected i.p. with 50 μCi [^{3}H]tyrosine and 50 μCi of [^{3}H]tryptophan (sp. act 32 mCi/m-mole and 1.5 mCi/m-mole, respectively) in a total volume of 1.01 ml.

After decapitation, the brains were rapidly removed in a cold room (4°), the cerebellum, pineal gland and any adhering blood clots carefully discarded and the brains homogenized in 0.4 N perchloric acid. After centrifugation (20,000 *g* for 20 min) the concentrations and radioactivities of tyrosine, tryptophan, noradrenaline, dopamine and serotonin were determined by the method of Neff and coworkers [8].

In the second experiment, groups of male Wistar rats (initially weighing (70–80 g) were chronically treated with the four antidepressant drugs. The drugs were given in the drinking water in a concentration which corresponded to a daily intake of 7.2 mg of Org GB 94 or 2.3 mg of imipramine or 1.8 mg of desipramine or 6.0 mg of iprindol for 14 days. These doses correspond approximately to 1/32 of the acute LD_{50} of Org GB 94, imipramine and desipramine and 1/13 of the acute LD_{50} of iprindol [10]. At the end of this period, all animals were injected i.p. with [^{3}H]tyrosine and [^{3}H]tryptophan, killed by decapitation after exactly 40 min and concentrations and radioactivities of the amines and their precursors determined in perchloric acid extracts of the whole brain by the method of Neff *et al.*

In the third experiment, 10 rats were injected i.p. twice daily, at 0900 and 1800 hr, with Org GB 94 (30 mg/kg) or with physiological saline (controls) for 14 days. Twenty-four hours after the last injection of Org GB 94 or saline the rats were injected i.p. with the tritiated amino acids and killed 40 min later. Experimental details are the same as those given

* Present address: Pharmacology Department, University College, Galway, Republic of Ireland.
† To whom requests for reprints should be addressed.

Table 1. Effect of the acute administration of iprindol, imipramine and ORG GB 94 on the turnover of biogenic amines in the whole rat brain

Compound	Mean value	Deviation from control (%)	95 per cent Confidence limits	Mean value	Deviation from control (%)	95 per cent Confidence limits	Mean value	Deviation from control (%)	95 per cent Confidence limits
	Tyrosine concentration‡			Tyrosine sp. act§			Conversion index†		
Control	62.9			65.1					
Iprindol	43.2*	−31	(−46, −13)	68.6	+5	(−28,55)			
ORG GB 94	42.8*	−32	(−46, −14)	69.6	+7	(−27,57)			
Control	91.7			33.0					
Imipramine	62.9*	−31	(−44, −16)	39.7*	+20	(+1,43)			
	Dopamine concentration‡			Dopamine sp. act§			Dopamine conv. index†		
Control	5.1			24.0			0.73		
Iprindol	4.5	−14	(−32,9)	24.2	+1	(−15,19)	0.70	−5	(−33,35)
ORG GB 94	4.8	−7	(−27,17)	28.9*	+20	(+2,43)	0.83	+13	(−21,59)
Control	6.7			16.7			1.01		
Imipramine	6.6	−1	(−21,23)	18.3*	+10	(+2, +18)	0.92	−9	(−23,8)
	Noradrenaline concentration‡			Noradrenaline sp. act§			Noradrenaline conv. index†		
Control	2.2			34.7			1.06		
Iprindol	2.0	−9	(−21,5)	38.4	+11	(−21,56)	1.12	+5	(−23,43)
ORG GB 94	2.0*	−10	(−22, −3)	65.8*	+90	(35,168)	1.89*	+77	(30,142)
Control	2.9			18.7			1.13		
Imipramine	3.0	+4	(−16,29)	17.0	−9	(−41,39)	0.86	−25	(−48,9)
	Tryptophan concentration‡			Tryptophan sp. act§					
Control	8.7			116					
Iprindol	11.3	+35	(−10,104)	123	+6	(−27,56)			
ORG GB 94	9.0	+7	(−29,61)	116	0	(−32,46)			
Control	11.7			35.8					
Imipramine	13.7	+20	(−22,83)	53.8*	+50	(22,85)			
	Serotonin concentration†			Serotonin sp. act§			Serotonin conv. index†		
Control	2.5			104			1.03		
Iprindol	2.6	+3	(−12,22)	95.9	−7	(−34,31)	0.93	−10	(−42,41)
ORG GB 94	2.4	−7	(−21,9)	102.0	−2	(−30,39)	1.05	+2	(−34,59)
Control	1.9			42.9			1.43		
Imipramine	2.1	+11	(−6,30)	36.0	−16	(−34,7)	0.80*	−44	(−58,26)

Rats treated with imipramine or Org GB 94 for 120 min and iprindol for 150 min. Control animals injected with physiological saline. All rats were given an injection of tritiated tyrosine and tryptophan 40 min before decapitation. *Difference between control and experimental group significant at $P < 0.05$. Results given as mean of at least five rats: 95 per cent confidence limits shown in parentheses.

† Conversion Index = ratio of specific activity of the amine to its amino acid precursor.

‡ Mean values expressed an nmoles/g wet wt.

§ dpm./nmole/g wet wt.

above. The results are expressed in terms of the specific activities of the amines and their precursors; an indication of the effects of the drugs on amine turnover was obtained by calculating the Conversion Index (ratio of the specific activity of the amine in the brain to that of its precursor) as described elsewhere [8, 9]. All results are expressed as the mean values together with the 95 per cent confidence interval [11].

RESULTS

Effects after acute administration. Following the acute administration of all three antidepressants there was a decrease in the concentration of tyrosine but no significant changes in the concentrations of tryptophan or the three amines (Table 1). The specific activities of tyrosine, dopamine and tryptophan were increased following imipramine administration while the conversion index for serotonin was significantly decreased. Although the conversion indexes for noradrenaline and dopamine were slightly decreased after imipramine treatment, the change did not reach statistical significance. The rats which were acutely treated with Org GB 94 had slightly decreased concentrations of brain noradrenaline and tryptophan and the specific activity of brain tyrosine was also slightly reduced (Table 1). The most noticeable change was in the conversion index for noradrenaline, however; this was increased slightly, but significantly, following the administration of the drug. Apart from its effect on the concentration of tyrosine, iprindol did not affect the concentrations, specific activities or conversion indexes of any of the neurochemical parameters.

Effects after chronic administration. In the study of the chronic effects of the four antidepressants, following their oral administration, imipramine reduced the concentrations of tyrosine and noradrenaline; this drug also reduced the conversion index for dopamine and to a lesser extent noradrenaline and serotonin (Table 2). Desipramine reduced both the concentration and specific activity of noradrenaline and significantly reduced the conversion index for dopamine and serotonin the conversion index for noradrenaline was also reduced but this effect did not reach statistical significance. Iprindol, apart from causing a slight reduction in the specific activity of serotonin did not significantly affect any of the parameters studied. Org GB 94 had little effect on any of the parameters apart from causing a slight increase in the dopamine concentration and an increase in the incorporation index of noradrenaline.

In the third experiment, in which the effect of Org GB 94 was studied following its chronic i.p. adminis-

Table 2. Effect of the chronic oral administration of some antidepressant drugs on the turnover of biogenic amines in the whole rat brain

Compound	Mean value	Deviation (%)	95 per cent Confidence limits	Mean value	Deviation (%)	95 per cent Confidence limits	Mean value	Deviation (%)	95 per cent Confidence limits
	Tyrosine concentration†			Tyrosine sp. act§			Conversion index‡		
Control	115.7	—	—	57.2					
Imipramine	83.2*	−28	(−42, −2)	81.1	+42	(−4,110)			
Desimipramine	95.8	−17	(−39,13)	89.0*	+55	(+5,130)			
Iprindol	95.8	−17	(−33,15)	33.7	−42	(−77,50)			
ORG GB 94	88.9	−23	(−44,5)	57.7	+1	(−32,49)			
	Dopamine concentration†			Dopamine sp. act§			Dopamine conversion index‡		
Control	5.5	—	—	42.5	—	—	1.51	—	—
Imipramine	6.8	+24	(−1.57)	33.4	−21	(−42.6)	0.82*	−45	(−64, −18)
Desimipramine	6.9	+25	(−1,58)	41.0	−4	(−28,30)	0.93*	−38	(−59, −8)
Iprindol	5.0	−9	(−31,22)	30.4	−28	(−73,92)	1.82	+21	(−7,61)
ORG GB 94	7.3*	+33	(+6,68)	34.7	−18	(−39,10)	1.20	−20	(−47,20)
	Noradrenaline concentration†			Noradrenaline spec. act.§			Noradrenaline conv. index‡		
Control	2.9	—	—	34.9	—	—	1.22	—	—
Imipramine	2.4*	−18	(−29, −4)	41.7	+20	(−13,65)	1.03	−16	(−49,37)
Desimipramine	2.4*	−19	(−31, −6)	48.1*	+38	(0,90)	1.09	−11	(−46,45)
Iprindol	3.2	+10	(−19,44)	18.3	−48	(−83,56)	1.10	−11	(−36,27)
ORG GB 94	2.9	−3	(−16,13)	40.6	+16	(−15,10)	1.30	+14	(−3,86)
	Tryptophan concentration†			Tryptophan sp. act.§					
Control	9.4	—	—	90.7	—	—			
Imipramine	7.4	−22	(−51,25)	136.7	+51	(−2,131)			
Desimipramine	7.0	−25	(−53,20)	155.3*	+71	(+12,162)			
Iprindol	7.3	−23	(−48,16)	65.9	−27	(−60,71)			
ORG GB 94	9.2	−2	(−39,57)	96.4	+6	(−31,63)			
	Serotonin concentration†			Serotonin sp. act§			Serotonin conv. index‡		
Control	1.5	—	—	68.2	—	—	1.90	—	—
Imipramine	1.5	+4	(−23,42)	67.7	−1	(−28,36)	0.60	−68	(−60,7)
Desimipramine	1.6	+5	(−23,43)	72.1	+6	(−23,45)	0.56*	−71	(−62,0)
Iprindol	1.2	+20	(−33,2)	57.3	−16	(−76,32)	1.04	−45	(−23,31)
ORG GB 94	1.5	0	(−12,91)	76.3	+12	(−18,53)	0.95	+5	(−35,71)

Groups of rats treated orally with the antidepressants for 2 weeks. Control animals were untreated. 40 min before the animals were killed by decapitation, they were all injected with tritiated tryptophan and tyrosine. *Difference between control and experimental groups significant at $P < 0.05$. Results given as mean value; 95 per cent Confidence limits are shown in parentheses.

† Mean values expressed as nmole/g wet wt.

‡ Conversion Index = ratio of specific activity of the amines to its amino acid precursor.

§ dpm/nmole/g wet wt.

tration for 2 weeks, both the specific activity and the conversion index for noradrenaline were significantly increased (Table 3). Apart from a reduction in the concentration of tryptophan, no other change reached statistical significance.

DISCUSSION

The results of this investigation show that the chronic effects of imipramine on the turnover of brain amines differ from the acute affects. Thus after the acute administration of this drug there was a significant reduction in the turnover of serotonin and to a lesser extent of noradrenaline, whereas following chronic administration the turnover of dopamine was significantly reduced; there was also a reduction in the turnover of noradrenaline and serotonin after chronic administration but this did not reach statistical significance. Other investigators have shown that the chronic administration of imipramine causes a decrease in the concentration of 5-hydroxyindole acetic acid and this was interpreted as indicating a reduced turnover of serotonin [12]. The concentration of noradrenaline was unchanged, whereas that of dopamine increased in the cerebellum and pons medulla region. In contrast, Schildkraut and co-workers [13] found evidence to suggest that whereas imipramine decrease in the concentration of 5-hydrozyindole acetic administration, it increases noradrenaline turnover after chronic administration. Clearly, these results are at variance with those found in the present study; the reason for this is not apparent. Desipramine had a qualitatively similar effect to imipramine following its chronic administration; it significantly reduced the turnover of dopamine and serotonin and slightly reduced that of noradrenaline. This effect of desipramine was surprising in view of the findings of other investigators who reported that following the administration of desipramine and imipramine, the former drug primarily reduced the turnover of noradrenaline while the latter preferentially reduced that of serotonin [14]. One possible explanation for the difference could be that both of these drugs were given chronically in the present study but acutely in the study of Schubert *et al.* [14]

Iprindol is a tricyclic antidepressant with a unique biochemical profile in that it does not apparently reduce amine reuptake [15], nor does it affect amine turnover. Thus in the present study, iprindol did not significantly affect the steady-state concentrations of any of the amines or their precursors, neither did it affect the incorporation index for the three amines. Although the mechanism of action of iprindole is unclear, it is of particular interest that it acts by a

Table 3. Effect of chronic intraperitoneal administration of ORG GB 94 for 14 days on the turnover of biogenic amines in the whole rat brain

Compound	Mean value	Deviation from control (%)	95 per cent Confidence limits	Mean value	Deviation from control (%)	95 per cent Confidence limits	Mean value	Deviation from control (%)	95 per cent Confidence limits
	Tyrosine concentration			Tyrosine sp. act			Conversion index		
Control	98.0			92.3					
ORG GB 94	60.7	−38	(−81,103)	64.7	−30	(−81,152)			
	Dopamine concentration			Dopamine sp. act			Dopamine conv. index		
Control	4.4			45.8			0.87		
ORG GB 94	5.2	+18	(−2,43)	35.1	−23	(−85,294)	1.16	+33	(−84, +9)
	Noradrenaline concentration			Noradrenaline sp. act			Noradrenaline conv. index		
Control	0.7			170			3.20		
ORG GB 94	0.8	+14	(−19,60)	326*	+92	(+46,252)	6.65*	+108	(+12,287)
	Tryptophan concentration			Tryptophan sp. act					
Control	12.4			99.6					
ORG GB 94	8.3*	−33	(−52, −6)	117	+18	(−85,340)			
	Serotonin concentration			Serotonin spec. activity			Serotonin conv. Index		
Control	1.8			103			1.05		
ORG GB 94	2.1	+17	(−35,112)	77.9	−25	(−76, 134)	1.45	+38	(−19,135)

A pulse injection of tritiated tyrosine and tryptophan was administered 24 hr after the last injection of Org GB 94 and killed 40 min later. Org GB 94 was injected twice daily with 30 mg/kg i.p. Details otherwise as given in legend to Table 2.

mechanism which does not apparently involve the turnover of brain biogenic amines.

Org GB 94 differs from the other antidepressants studied in that it increases the turnover of noradrenaline after both acute and chronic administration without appreciably affecting the steady-state concentrations or specific activities of the amines or their precursors.

This effect on noradrenaline metabolism was greater after i.p. than after oral administration. The reason for this effect is unclear at present; one possibility could be that the metabolism of Org GB 94 varies according to the route of administration. Previous studies have shown that *in vivo*, Org GB 94 does not affect the reuptake of noradrenaline or serotonin [7, 17, 18]. Thus it differs from the tricyclic antidepressants of the dibenzazepine type and from iprindol in that it increases brain noradrenaline turnover. The mechanism whereby this occurs is uncertain, it seems unlikely that it acts like lithium in increasing noradrenaline reuptake [19].

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