Br. J. Pharmac. (1979), **65**, 303–309

REVERSAL OF DOPA-INDUCED AROUSAL IN RESERPINE-TREATED RABBITS AND MICE BY HISTIDINE

Y.Z. ABOU[1] & I.B. FARJO

Department of Pharmacology, College of Medicine, Baghdad University, Iraq

1 The behavioural effects induced by histidine were studied in two species. In rabbits, sedation was assessed by the presence of blepharospasm, loss of righting reflex, and loss of response to painful stimuli. In mice, sedation and arousal were assessed by changes in the locomotor activity, exploratory activity, and minimal electroshock seizure threshold.

2 The administration of histidine to normal rabbits or mice, in doses of 800 mg/kg and 1000 mg/kg respectively, had no apparent effect on behaviour. Moreover, it did not affect the behavioural excitation induced by L-DOPA (100 mg/kg i.v. in rabbits and 750 mg/kg i.p. in mice) in these animals.

3 The administration of histidine with or after L-DOPA in reserpine-treated rabbits (2.5 mg/kg i.v.) or mice (5 mg/kg, i.p.) produced sedation. This sedative effect was dose-dependent.

4 The sedative effects induced by histidine after DOPA-induced arousal in reserpine-treated rabbits and mice were prevented by prior injection of the histamine H_1-receptor blockers, chlorpheniramine (2.5 mg/kg) or diphenhydramine (5 mg/kg).

5 Imipramine (7 to 10 mg/kg, i.v.)-induced arousal in reserpine-treated rabbits was also reversed by histidine infusion.

6 The infusion of 5-hydroxytryptophan (100 mg/kg, i.v.) with L-DOPA, or of arginine (450 mg/kg, i.v.) with or after L-DOPA, or of histamine (100 μg/kg), i.v.) after L-DOPA, did not affect the DOPA-induced arousal in reserpine-treated rabbits.

7 These findings indicate that histamine, formed centrally from exogenous histidine, and released in increased amounts at the synapses in reserpine-treated animals, possesses a central sedative effect. This effect may be sufficient to antagonize the behavioural excitation induced by high levels of catecholamines in the brain of these animals when aroused by L-DOPA administration.

8 It is concluded that in addition to the other monoamines, histamine may also be implicated in the regulation of brain excitability.

Introduction

The presence of histamine-containing neurones within the mammalian brain, together with neuropharmacological data, suggest that histamine is probably a central neurotransmitter (see Schwartz, 1977). However, the search continues to assign a behavioural as well as a physiological function to these histaminergic neurones.

The intravenous infusion of histidine in rabbits was shown to cause a rapid and dose-related rise in histamine concentration in various parts of the brain but this rise was not associated with any evident effect on behaviour (Abou, Adam & Stephen, 1973). However, recent data suggested that histidinaemia may carry an increased risk of neurological involvement and mental abnormality (Popkin, Clow, Scriver & Grove, 1974).

In an effort to demonstrate the central effects of histamine, L-3,4,dihydroxyphenylalanine (L-DOPA)-induced arousal in reserpine-treated rabbits was found to be quickly reversed following histidine infusion (Abou & Al-Katib, 1972). These workers suggested that histamine which was rendered 'free' by reserpine, might have a sedative effect.

The present work extends the above observations on L-DOPA-induced arousal in rabbits. In one experiment, the effect of histidine was also tested following the reversal of the sedative effect of reserpine by

[1] Present address: Department of Pharmacology, Al-Mustansiriyah Medical School, Baghdad, Iraq.

0007-1188/79/020303-07 $01.00

imipramine (Costa, Garattini & Valzelli, 1960; Sulser, Watts & Brodie, 1961; Maxwell & Palmer, 1961). Imipramine is known to block the active reuptake of monoamines by nerve terminals, thereby potentiating their action at postsynaptic receptor sites (Schildkraut, 1965). Moreover, the central effects induced by histidine were also assessed in mice in which the central effects of drugs could be more precisely quantified.

Methods

Animals

Albino rabbits of either sex, weighing 1 to 2.5 kg, were used; 6 to 8 animals were assigned to each experiment.

Albino mice (Swiss strain) were housed, 4 mice to a cage, at a constant temperature of 22°C. Six to 10 mice were assigned for each experiment. All experiments were begun between 09 h 00 min and 10 h 00 minutes. Animals were deprived of water and food for about 30 min and during the experiments. Control experiments were conducted concurrently with their respective groups. Extreme care was taken to avoid noise and restraint during the experimental procedure.

Drugs and solutions

All solutions were withdrawn into sterile disposable syringes before being injected.

In rabbits, drugs were given by slow infusion through a cannula inserted into the marginal ear vein. Heparin (Evans) was given (800 iu/kg) routinely to avoid clotting within the cannula during the period of the experiment. In mice, all drugs were given intraperitoneally (i.p.).

All drug and amino acid solutions were freshly prepared in sterile saline (0.9% w/v NaCl solution) except reserpine which was dissolved in 10% ascorbic acid solution.

The following solutions were used: reserpine (Ciba) 1 mg/ml; L-DOPA (Koch-Light) 10 mg/ml; L-histidine (Koch-light) 80 mg/ml; 5-hydroxytryptophan (5-HTP, Koch-light) 5 mg/ml; arginine (Koch-light) 22.5 mg/ml; imipramine (Geigy) 1.5 mg/ml; chlorpheniramine hydrochloride (Schering Corp.) 1 mg/ml; and diphenhydramine hydrochloride (Schering Corp.) 2 mg/ml.

In rabbits, infusions were made slowly by hand at a rate of 0.5 ml/min for reserpine, 3 ml/min for amino acids, and 0.5 ml/min for imipramine and antihistamines, until the required dose of each drug was given.

In mice, the above solutions served to give the required amount of the drug in a minimal volume for injection into the small peritoneal cavity except L-DOPA , which was used in a concentration of 30 mg/ml, the solubility being increased by the addition of a small amount of 0.05 N HCl. The total volume of any solution injected intraperitoneally into any mouse did not exceed 0.1 ml/10 g. The doses of all drugs used are given in the results. Control animals received 10% ascorbic acid and/or saline as appropriate for each experiment.

Assessment of central effects

Rabbits Behavioural effects were assessed by the following criteria (Brodie, Comer, Costa & Dlabac, 1966): (a) delay of 30 s or more in the recovery of the righting reflex after placing the animal on its side, (b) lack of response to painful stimuli evoked by manual displacement of the tarsometatarsal joints, and (c) the presence of blepharospasm. Each sign was counted as (1) and the pharmacological effect of any drug was scored from (0) for full arousal to (3) for full sedation.

Testing was done at 15, 30, 60 and 120 min after reserpine, and at 5, 15, 30, 60 and 120 min following L-DOPA or imipramine infusion. Histidine was infused 10 min after L-DOPA. In some experiments, histidine was given 10 min after antihistamines which were given 10 min following L-DOPA infusion. Testing was also done at 5, 10, 15, 30, 60 and 120 min following histidine infusion.

Mice The central effects of drugs on behaviour were assessed by (a) the exploratory activity and (b) the locomotor activity which were measured simultaneously in the hole-board apparatus (File, 1972); (c) the minimal electroshock seizure threshold (MEST) (Azzaro, Wenger, Craig & Stitzel, 1972), and (d) the presence of belpharospasm (Rubin, Malone, Waugh & Burke, 1957).

The exploratory activity was assessed by recording the number of head-dips during a 3 min trial period; the number of different holes and the total number of holes were counted and entered on a score sheet for each animal. A head-dip was scored if both eyes of the animal disappeared into the hole and ended when the head was completely clear of the hole.

Locomotor activity was assessed by the method of Ahtee & Shillito (1970). The 3 min period was divided into 18 periods, each of 10 s, and the dominant type of behaviour (moving, resting or grooming) was recorded during each period.

The minimal electroshock seizure threshold (MEST) was employed for testing the effect of drugs on brain excitability. After moistening the ear canal with saline, clip electrodes with long leads were connected to each ear of the mouse. The test was carried out within 30 s after the application of the electrodes

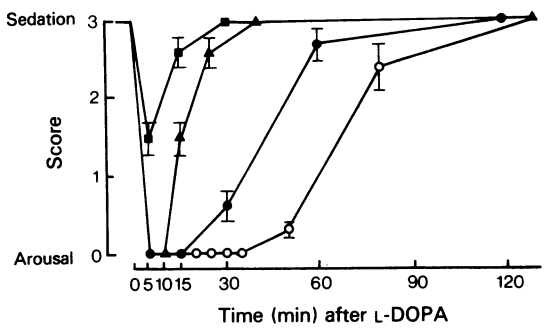

Figure 1 Effects of drugs on histidine-induced behaviour in reserpine-treated rabbits. The criteria for assessing central sedative effects were the presence of blepharospasm, loss of righting reflex and lack of response to pain; a mean score of (3) indicates full sedation and (0) indicates full arousal. Each value represents the mean result obtained from 8 rabbits; vertical lines show s.e. mean. (●) L-DOPA (100 mg/kg) infused 2 h after reserpine injection (2.5 mg/kg) when rabbits were heavily sedated; (▲) L-histidine (800 mg/kg) infused 10 min after DOPA-induced arousal in reserpine-treated rabbits; (○) chloropheniramine (2.5 mg/kg) injected 10 min after DOPA-induced arousal and histidine (800 mg/kg) infused 10 min after chlorpheniramine injection; (■) DOPA (100 mg/kg) and histidine (400 mg/kg) infused simultaneously 2 h after reserpine injection (2.5 mg/kg).

in unrestrained mice. The MEST was determined with 0.2 ms pulses at 6 Hz from a Grass Stimulator (Model S4 k). The intensity of the current required to evoke minimal seizure (stun responses or 3 to 5 s of continued minimal clonic activity) was measured.

During experiments, 3 min testing sessions were carried out as follows: Trial I: 3 h after reserpine; Trial II: 30 min after L-DOPA; Trial III: 60 min after L-DOPA; Trial IV: 90 min after L-DOPA and Trial V: 120 min after L-DOPA.

In other experiments histidine was injected 30 min after L-DOPA administration in reserpinized mice, i.e. after Trial II and therefore Trials III, IV and V also corresponded to 30, 60 and 90 min following histidine administration.

In some experiments, antihistamines were given 10 min after L-DOPA injection.

Results

Rabbits

Twenty-two groups of rabbits were employed; some of the results are shown in Figure 1. As expected from previous studies, histidine alone had no overt effect on behaviour in untreated animals but could be shown to oppose the stimulant effects of L-DOPA in reserpine-treated animals.

Reserpine (2.5 mg/kg) produced sedation which was evident at 1, 2 and 4 h. However, these sedative effects were completely reversed within a few min and full arousal was maintained for at least 30 min when L-DOPA (100 mg/kg) was injected 2 h after reserpine (Figure 1).

When histidine (800 mg/kg) was injected 10 min after L-DOPA in reserpine-treated rabbits, the animals became sedated within 5 min; sedation was marked at 15 and maximal at 30 min (Figure 1). The administration of another dose of L-DOPA at this stage reversed the sedative effects of histidine (not shown). A lower dose of histidine (600 mg/kg) also reversed DOPA-induced arousal, but the onset of sedation was slower; a dose of 550 mg/kg caused only mild sedation and a dose of 450 mg/kg had no demonstrable effect. This sedative effect of histidine was not seen in animals treated with vehicle instead of reserpine, and then given L-DOPA.

When the lower dose of histidine (400 mg/kg) was given with L-DOPA (rather than 10 min after) 2 h after reserpine, the typical DOPA arousal was markedly attenuated (Figure 1). To exclude the possibility that the effects of histidine were merely due to hindrance of L-DOPA uptake or metabolism, two control experiments were used. The simultaneous administration of L-DOPA (100 mg/kg) and 5-HTP (100 mg/kg) 2 h after reserpine injection produced sudden arousal and excitation which was more intense and of longer duration than that induced by DOPA alone in reserpine-treated animals. Further, arginine (450 mg/kg) did not antagonize DOPA-induced arousal in reserpine-treated rabbits, whether it was given after or at the same time as L-DOPA.

The infusion of histamine (100 μg/kg) after DOPA-induced arousal did not produce detectable signs of sedation. Imipramine (7 to 10 mg/kg) had no demonstrable sedative effects in rabbits but imipramine-induced arousal in reserpine-treated animals was reversed by histidine (800 mg/kg). Sedation was marked after 15 min and maximal 30 min after histidine infusion in these aroused animals.

It seemed of interest to determine whether the histidine effects could be modified by antihistamines (H_1-receptor antagonists). Chlorpheniramine (2.5 mg/kg) did not produce significant sedation when used alone, but prevented the sedative effects of histidine on DOPA-induced arousal (Figure 1). Diphenhydramine (5 mg/kg) produced similar results.

Mice

As in rabbits, histidine alone in doses up to 1 g/kg had no effect on exploratory and locomotor activity or MEST, up to 3 h when compared with saline con-

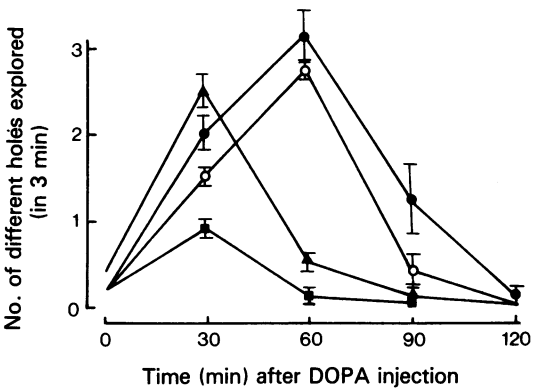

Figure 2 Effects of histidine after DOPA-induced arousal in reserpine-treated mice. Each value represents the mean (no. of different holes explored in 3 min) obtained from 10 animals; vertical lines show s.e. mean. (●) (Group a) L-DOPA (750 mg/kg) given 3 h after reserpine injection (5 mg/kg) when exploratory activity was markedly reduced; (▲) (Group b) L-histidine (1 g/kg) injected immediately after Trial II (i.e. 30 min after DOPA) when exploratory activity had partly recovered; (○) (Group c) chlorpheniramine (2.5 mg/kg) given 10 min after DOPA injection; then histidine (1 g/kg) injected immediately after Trial II (i.e. 30 min after DOPA); (■) (Group d) DOPA (750 mg/kg) and histidine (500 mg/kg) injected simultaneously 3 h after reserpine administration.

trols. Again, however, a sedative effect of histidine could be demonstrated against the background arousal produced by L-DOPA after reserpine. This is shown in Table 1 and Figure 2.

L-DOPA (750 mg/kg) was administered 3 h after reserpine injection (5 mg/kg), when mice were heavily sedated. DOPA-induced arousal was evident at 30 min and marked at 60 min; at 90 min the sedative effects of reserpine began to reappear, and at 120 min mice exhibited marked signs of sedation (Table 1, group a).

The injection of histidine (1 g/kg) 30 min after L-DOPA (Table 1, group b) produced sedation in all measures which was highly significant at 30 min (i.e. 60 min after DOPA). This sedation was more pronounced at 90 min (cf. reserpine and DOPA at corresponding times in Table 1 and Figure 2).

As in rabbits, a lower dose of histidine (500 mg/kg) given in conjunction with L-DOPA 3 h after reserpine prevented the arousal that usually took place when L-DOPA alone was given. Thirty min after the injection of the mixture, mice became slightly more active but sedation was marked at 60 min and exploratory and locomotor activities were nil at 90 and 120 min (Figure 2).

The injection of chlorpheniramine (2.5 mg/kg) or diphenhydramine (5 mg/kg) in otherwise untreated animals did not produce significant alterations in the parameters tested at 1, 2 and 3 h in comparison with saline-treated controls. However, when chlorpheniramine was given 10 min after L-DOPA it reduced but did not prevent DOPA-induced reversal of reserpine sedation (see comparisons of Trial II in Table 1). However, when histidine was administered 20 min after chlorpheniramine (i.e. following Trial II), and mice tested 30 min later (i.e. Trial III), the typical histidine sedative effect on exploratory and locomotor activity and on MEST was blocked substantially, the values approaching those seen with L-DOPA only in reserpine-treated mice.

The results with diphenhydramine in blocking the sedative effects of histidine were essentially similar to those obtained with chlorpheniramine.

Discussion

The onset and duration of L-DOPA-induced reversal of reserpine sedation has been attributed to an increase in the concentration of central catecholamines synthesized following the decarboxylation of exogenous L-DOPA (Carlsson, Lindqvist & Magnusson, 1957; Bartholini & Pletscher, 1969). About 90% of administered L-DOPA is decarboxylated by the peripheral tissue and thus a small part is available for penetration of the blood-brain barrier into the brain tissue. Furthermore, peripherally formed catecholamines do not pass the blood-brain barrier (Weil-Malherbe, Axelrod & Tomchick, 1959).

The return to the reserpinized state at 90 to 120 min following L-DOPA infusion reflects the catabolic elimination of centrally formed catecholamines. Moreover, reserpine does not inhibit the decarboxylation of L-DOPA (Carlsson *et al.*, 1957) or the catabolizing enzymes monoamine oxidase and catechol-*O*-methyl transferase (Glowinski & Axelrod, 1965). Furthermore, reductions of brain noradrenaline levels by fusaric acid (FLA-63), a dopamine-β-hydroxylase inhibitor, leads to attenuation of locomotor activity after L-DOPA-induced arousal in reserpine-treated animals, despite the high levels of brain dopamine (Dolphin, Jenner & Marsden, 1975). Thus, it seems that normal or high levels of noradrenaline are more essential for locomotor stimulation following L-DOPA administration.

The administration of large doses of histidine to normal rabbits or mice failed to produce any detectable effect on behaviour. The major pathways of histidine utilization are incorporated into proteins, degradation by histidase, and excretion in the urine; decarboxylation into histamine is a minor pathway

Table 1 Effect of histidine after DOPA-induced arousal in reserpine-treated mice

Trial no.	*I 3 h after reserpine*			*II 30 min after DOPA*¶			*III 60 min after DOPA*			*V 120 min after DOPA*		
Group no.‡	a	b	c	a	b	c	a	b* (i.e. 30 min after histidine)	c	a	b (i.e. 90 min after histidine)	c
Exploratory† activity	0.2 ± 0.1	0.4 ± 0.2	0.2 ± 0.1	2.0 ± 0.2	2.5 ± 0.2	1.5 ± 0.1	3.1 ± 0.3	0.5 ± 0.1	2.7 ± 0.1	0.1 ± 0.1	0	0
Locomotor activity	0.3 ± 0.1	0.8 ± 0.2	0.1 ± 0.1	5.8 ± 0.2	6.8 ± 0.7	4.0 ± 0.4	9.1 ± 0.6	1.0 ± 0.3	7.6 ± 0.4	0.5 ± 0.1	0	0.3 ± 0.1
MEST	24 ± 2.3	23 ± 0.7	24 ± 2.0	73 ± 6.0	62 ± 9.0	53 ± 2.5	78 ± 5.3	37 ± 3.7	52 ± 2.0	36 ± 3.7	31 ± 3.3	26 ± 2.0

† Exploratory activity: no. of different holes explored in 3 min; locomotor activity: no. of 10 s periods during which the animal was moving, taken over 3 min (a total of 18 periods); MEST: minimal electro-shock seizure threshold in volts. Each value represents the mean ± s.e. (10 mice).

‡ Group (a) (control) received reserpine (5 mg/kg) followed 3 h later by L-DOPA (750 mg/kg); DOPA was given immediately after Trial I. Group (b) received reserpine followed by DOPA (as above); histidine (1 g/kg) was then given immediately after Trial II (i.e. 30 min after DOPA). Group (c) received reserpine followed by DOPA (as above); chlorpheniramine (2.5 mg/kg) was then given 10 min after DOPA and histidine was injected immediately after Trial II.

¶ The differences for the 3 groups are very highly significant ($P < 0.001$) in comparison with reserpine-treated state (Trial I).

* The values in group (b) are significantly reduced ($P < 0.001$) in comparison with groups (a) and (c) and in comparison with values at Trial II.

of histidine catabolism in mammals (Tabor, 1954). It has been suggested that the newly-formed brain histamine from exogenous histidine is probably stored in granules and thereby inactive. This conclusion was based on the finding that newly-formed histamine disappeared only slowly from the rabbit brain (Abou *et al.*, 1973). However, it has been shown that storage of histamine in the rabbit brain was impaired by reserpine pretreatment (Abou, 1968). Thus, in reserpine-treated animals, one can anticipate the presence of larger quanta of newly-formed histamine from infused histidine to be released into the central synapses for action at receptor sites. The administration of histidine to reserpine-treated animals did not aggravate sedation, probably because the animals were already heavily sedated. However, the infusion of histidine in conjunction with or after L-DOPA, inhibited L-DOPA-induced arousal in reserpine-treated mice and rabbits.

The observation that, in both species, the dose of histidine required to prevent arousal was lower than that needed to reverse it, might be due to larger quantities of central catecholamines already formed when histidine infusion followed that of L-DOPA.

It has been shown that the L-system is the predominant transport mechanisms for L-DOPA and other neutral amino acids at the level of the blood-brain barrier (Wade & Katzman, 1975). Moreover, the transport system for neutral amino acids differs from that for basic amino acids like arginine, histidine and lysine, across the blood-brain barrier (Shah, Kamano, Glisson & Callison, 1958; Oldendorf, 1971). The intraperitoneal administration of histidine (0.5 mg/g) raises the cerebral histidine concentration by seven fold, while there is a simultaneous reduction in the levels of branched chain and aromatic amino acids in the same rat brain (Tyfield & Holton, 1976). This could be explained by inhibition of transport of neutral amino acids at sites shared with histidine (McKean, Boggs & Peterson, 1968).

In the present work, however, the effect of histidine was unlikely to be mediated through competition with the L-DOPA transport mechanism with consequent diminished penetration of the latter into the brain for three reasons. First, the sedative effect induced by infused histidine was very rapid in onset occurring within a few min of its administration. Moreover, this sedation was dose-dependent and could be reversed by re-infusion of L-DOPA. Secondly, the sedative effect following histidine infusion was blocked by pretreatment with antihistamines. This effect adds further support for the assumption that the sedative effects of histidine are mediated by centrally-formed histamine acting on H_1-receptor sites, rather than by the parent amino acids itself. Thirdly, it has been shown that cerebral histidine levels are reduced by arginine (which is a basic amino acid, like histidine) and *vice versa* (Richter & Wainer, 1971). However, infusions of arginine or 5-HTP after or with L-DOPA did not prevent DOPA-induced arousal in reserpine-treated rabbits. This is in contrast to the sedative effect induced by histidine under these conditions. Thus, one can conclude that the hindrance of L-DOPA transport at the level of the blood-brain barrier does not seem to play an essential role in histidine-induced sedation. Moreover, the sedative effects following histidine infusion were also seen in reserpine-treated rabbits aroused by slow infusion of imipramine.

Thus, it seems that centrally-formed histamine from exogenous histidine in reserpinized animals is capable of counteracting the central stimulant effects of catecholamines, whether these are formed from the decarboxylation of exogenous L-DOPA or endogenously accumulated at receptor sites. Moreover, although DOPA or 5-HTP do not interfere with histidine-induced rise in brain histamine (Abou *et al.*, 1973), histidine in large doses may have reduced the formation of catecholamines from L-DOPA by encroaching upon the more abundant aromatic amino acid decarboxylase during its increased catabolism into histamine.

Histamine does not penetrate the blood-brain barrier (Halpern, Neveu & Wilson, 1959). Since the infusion of a relatively large dose of histamine did not alter DOPA-induced arousal in reserpine-treated rabbits, it seemed unlikely for histidine to have induced a state of hypotensive shock resulting from increased peripheral levels of histamine which would induce or aggravate any sedative effect. Moreover, it has been reported that histamine does not produce hypotension in reserpinized rabbits (Al-Katib & Baba, 1969) and that the concentration of histamine in the blood of these animals does not rise detectably after the infusion of large doses of histidine (Abou *et al.*, 1973).

The significant lowering of seizure threshold by histidine after L-DOPA-induced arousal in reserpine-treated mice suggests that brain histamine may also be implicated in seizure susceptibility. It is possible that reserpine-induced reduction in seizure threshold, which is mainly mediated by depletion of brain monoamines (Azzaro *et al.*, 1972), may also involve histaminergic mechanisms.

Finally, it is also possible that histaminergic mechanisms are implicated in the mental subnormality, speech and possibly neurological dysfunction in patients with histidinaemia.

The authors are deeply grateful to Professor D. W. Straughan of London University for valuable criticism in the preparation of this manuscript. This work was done by Dr I. B. Farjo, Dept. of Pharmacology, Baghdad Medical School, in partial fulfilment of the degree of M.Sc., from Baghdad University, Iraq.

References

ABOU, Y.Z. (1968). A study of the concentration of histamine in the hypophysis and brain of the rabbit and of the changes produced by treatment with amino acids and drugs. *Ph.D. Thesis, Faculty of Medicine, Edinburgh University.*

ABOU, Y.Z., ADAM, H.M. & STEPHEN, W.R.G. (1973). Concentration of histamine in different parts of the brain and hypophysis of the rabbit: effect of treatment with histidine, certain other amino acids and histamine. *Br. J. Pharmac.* **48,** 577–589.

ABOU, Y.Z. & AL-KATIB, H.A. (1972). Central effects of 'free' histamine on DOPA-induced arousal in reserpinized rabbits. *Proceedings of the First Scientific Meeting of the Research Establishment, Baghdad, Iraq,* pp. 135–139. Baghdad: University & Research Press.

AHTEE, L. & SHILLITO, E. (1970). The effect of benzodiazepines and atropine on exploratory behaviour and motor activity in mice. *Br. J. Pharmac.*, **40,** 361–371.

AL-KATIB, H.A. & BABA, W.I. (1969). The action of certain haloalylamines on some biological activities of bradykinin. *Br. J. Pharmac.*, **36,** 635–641.

AZZARO, A.J., WENGER, G.R., CRAIG, C.R. & STITZEL, R.E. (1972). Reserpine-induced alterations in brain amines and their relationships to changes in the incidence of minimal electro-shock seizures in mice. *J. Pharmac. exp. Ther.*, 180, 558–568.

BARTHOLINI, G. & PLETSCHER, A. (1969). Effect of various decarboxylase inhibitors on cerebral metabolism of L-DOPA. *J. Pharm. Pharmac.*, **21,** 323–324.

BRODIE, B.B., COMER, M.S., COSTA, E. & DLABAC, A. (1966). The role of brain serotonin in the mechanism of the central action of reserpine. *J. Pharmac. exp. Ther.*, **152,** 340–349.

CARLSSON, A., LINDQVIST, M. & MAGNUSSON, T. (1957). 3,4-Dihydroxyphenylalanine and 5-hydroxytryptophan as reserpine antagonists. *Nature, Lond.*, **180,** 1200.

COSTA, E., GARATTINI, S. & VALZELLI, L. (1960). Interactions between reserpine, chlorpromazine and imipramine. *Experientia,* **16,** 461–463.

DOLPHIN, A., JENNER, P. & MARSDEN, C.D. (1975). The mechanism of effect of dopamine-β-hydroxylase inhibitor FLA-63 on the L-DOPA reversal of reserpine akinesia. *Br. J. Pharmac.*, **54,** 246–247P.

FILE, S.E. (1973). Effects of chlorpromazine on exploration and habituation in the rat. *Br. J. Pharmac.*, **49,** 303–310.

GLOWINSKI, J. & AXELROD, J. (1965). Effect of drugs on the uptake, release and metabolism of ^{3}H-norepinephrine in the rat brain. *J. Pharmac. exp. Ther.*, **149,** 43–49.

HALPERN, B.N., NEVEU, T. & WILSON, C.W.M. (1959). The distribution and fate of radioactive histamine in the rat. *J. Physiol.*, **147,** 437–449.

MAXWELL, D.R. & PALMER, H.T. (1961). Demonstration of antidepressant or stimulant properties of imipramine in experimental animals. *Nature, Lond.*, **199,** 84–85.

MCKEAN, C.M., BOGGS, C.E., & PETERSON, N.A. (1968). The influence of high phenylalanine and tyrosine levels on the concentrations of essential amino acids in the brain. *J. Neurochem.*, **15,** 235–241.

OLDENDORF, W.H. (1971). Brain uptake of radiolabelled amino acids, amines, and hexoses after arterial injection. *Am. J. Physiol.*, **221,** 1629–1639.

POPKIN, J.S., CLOW, C.L., SCRIVER, C.R. & GROVE, J. (1974). Is hereditary histidinaemia harmful? *Lancet,* **i,** 729–732.

RICHTER, J.J. & WAINER, A. (1971). Evidence for separate systems for the transport of neutral and basic amino acids across the blood-brain barrier. *J. Neurochem.*, **18,** 613–620.

RUBIN, B., MALONE, M.H., WAUGH, M.H., & BURKE, J.C. (1957). Bioassay of Rawoulfia roots and alkaloids. *J. Pharmac. exp. Ther.*, **120,** 125–132.

SCHILDKRAUT, J.J. (1965). The catecholamine hypothesis of affective disorders. A review of supporting evidence. *Am. J. Psychiat.*, **122,** 509–522.

SCHWARTZ, J.C., (1977). Histaminergic mechanisms in brain. *A. Rev. Pharmac. Tox.*, **17,** 325–339.

SHAH, N.S., KAMANO, A., GLISSON, S. & CALLISON, D. (1968). Studies on the uptake of radiolabelled DOPA, 5-HTP and tryptophan in rat tissue *in vivo*: Effect of methionine, tryptophan and some keto acids. *Int. J. Neuropharmac.*, **7,** 75–86.

SULSER, F., WATTS, J. & BRODIE, B.B. (1961). Blocking of reserpine action by imipramine, a drug devoid of stimulatory effects in normal animals. *Fedn. Proc.*, **20,** 321.

TABOR, H. (1954). Metabolic studies on histidine-histamine and related imidazoles. *Pharmac. Rev.*, **6,** 299–343.

TYFIELD, L.A., & HOLTON, J.B. (1976). Effect of high concentrations of histidine on the level of other amino acids in plasma and brain of the mature rat. *J. Neurochem.*, **26,** 101–105.

WADE, L.A. & KATZMAN, R. (1975). Synthetic amino acids and the nature of L-DOPA transport at the blood-brain barrier. *J. Neurochem.*, **25,** 837–842.

WEIL-MALHERBE, H., AXELROD, J. & TOMCHICK, R. (1959). Blood-brain barrier for adrenaline. *Science, N.Y.*, **129,** 1226–1227.

(Received July 13, 1978.)