Br. J. Pharmac. (1981), **74,** 143–148

EFFECTS OF MIANSERIN, DESIPRAMINE AND MAPROTILINE ON BLOOD PRESSURE RESPONSES EVOKED BY ACETYLCHOLINE, HISTAMINE AND 5-HYDROXYTRYPTAMINE IN RATS

ICILIO CAVERO, FRANÇOISE LEFÈVRE-BORG & ALAN G. ROACH[1]

Biology Department SYNTHELABO (L.E.R.S.), Cardiovascular Group, 58 rue de la Glacière, 75013 Paris, France

1 In rats anaesthetized with pentobarbitone, intravenous administration of desipramine (0.1 mg/kg), maprotiline (0.5 mg/kg) or mianserin (0.3–3.0 mg/kg) did not modify the blood pressure lowering effects of acetylcholine (0.25–1.0 μg/kg, i.v.) which were significantly reduced by atropine (3.0 μg/kg, i.v.).

2 Maprotiline and mianserin, like promethazine (0.1 mg/kg, i.v.), inhibited the vasodepressor responses evoked by histamine (2.5–10.0 μg/kg, i.v.). However, desipramine was inactive against histamine.

3 In pithed rats, the pressor effects of intravenous 5-hydroxytryptamine (5-HT: 5.0–20.0 μg/kg) were antagonized by mianserin (0.01–0.3 mg/kg, i.v.) and cyproheptadine (0.01 mg/kg) but were unaffected by maprotiline and desipramine.

4 In syrosingopine pretreated rats given mianserin 0.1 mg/kg, intravenously, 5-HT (20.0 μg/kg, i.v.) produced a significant fall in blood pressure which could be reduced by a large dose of mianserin (10.0 mg/kg, i.v.).

5 In conclusion, desipramine, maptrotiline and mianserin, in doses previously found to inhibit noradrenaline neuronal reuptake in the rat cardiovascular system, lack muscarinic receptor antagonist properties. Whilst maprotiline and mianserin blocked vascular histamine receptors, only mianserin, like cyproheptadine, was a potent antagonist of the 5-HT receptors that mediate increases in blood pressure in rats. Finally, the vasodepressor effects of 5-HT in syrosingopine pretreated rats given a small dose of mianserin were antagonized by a large dose of mianserin, suggesting that 5-HT may activate two distinct types of receptors in the rat.

Introduction

Mianserin is considered to be an atypical antidepressant agent since in contrast to tri- and tetracyclic noradrenaline neuronal reuptake inhibitors it is inactive in most of the classical animal tests now used to identify possible antidepressant agents (Brogden, Heel, Speight & Avery, 1978; Peet & Behagel, 1978).

In the cardiovascular system of the rat, mianserin possesses postsynaptic α_1- and presynaptic α_2-adrenoceptor antagonist activity and like desipramine and maprotiline, inhibits the neuronal uptake mechanism for noradrenaline (Doxey, Everitt & Metcalf, 1978; Robson, Antonaccio, Saelens & Liebman, 1978; Cavero, Gomeni, Lefèvre-Borg & Roach, 1980a; Docherty & McGrath, 1980). Furthermore, mianserin is an antagonist of responses mediated by 5-hydroxytryptamine (5-HT) in various preparations (Vargaftig, Coignet, De Vos, Grijsen & Bonta, 1971; Robson *et al.*, 1978; Figge, Leonard & Richelson, 1979; Tang & Seeman, 1980).

This paper complements our previous study on the effects of desipramine, maprotiline and mianserin on the cardiovascular sympathetic nervous system (Cavero *et al.*, 1980a) and compares the effects of these antidepressant agents on the blood pressure responses to acetylcholine, histamine and 5-HT in rats.

Part of this work was the subject of a communication to the British Pharmacological Society (Cavero, Lefèvre-Borg & Roach, 1980b).

Methods

Male rats (Sprague Dawley, Charles River France) weighing 250–280 g were anaesthetized with pentobarbitone sodium (55 mg/kg, i.p.) and artificially ventilated (Harvard, Model 680) with room air (approximately 1 ml/100 g body weight delivered 40–50 times/min).

[1] Present address: Reckitt & Coleman, Pharmaceutical Division, Dansom Lane, Kingston-upon-Hull, HU8 7DS

0007-1188/81/090143-06 $01.00

A carotid artery and femoral vein were cannulated for blood pressure monitoring and intravenous drug administration, respectively. Blood pressure was measured with a Statham 23Dd transducer connected to an appropriate preamplifier. Pulsatile and mean (electronically calculated) carotid blood pressure were displayed on a Grass 7D polygraph.

The blood pressure-lowering effects of intravenous bolus injections (given over 2–3 s in 1.0 ml/kg solution) of acetylcholine (0.25, 0.5 and 1.0 μg/kg) and histamine (2.5, 5.0 and 10.0 μg/kg) were studied in separate groups of rats before and after the intravenous administration of 0.9% w/v NaCl solution (saline, 0.4 ml/kg), desipramine (0.1 mg/kg), maprotiline (0.5 mg/kg) and mianserin (0.3–3.0 mg/kg). The mean blood pressure at the peak of the response was determined and expressed as the change (ΔMAP) from the pre-injection control level.

Atropine (3.0 μg/kg, i.v.) and promethazine (0.1 mg/kg, i.v.) were used as standard antagonists of the blood pressure-lowering effects of acetylcholine and histamine, respectively.

The vasopressor responses to 5-HT (5.0, 10.0 and 20.0 μg/kg, i.v.) were studied in atropine (0.5 mg/kg, i.v.) pretreated pithed rats under control conditions and after intravenous administration of saline, desip-

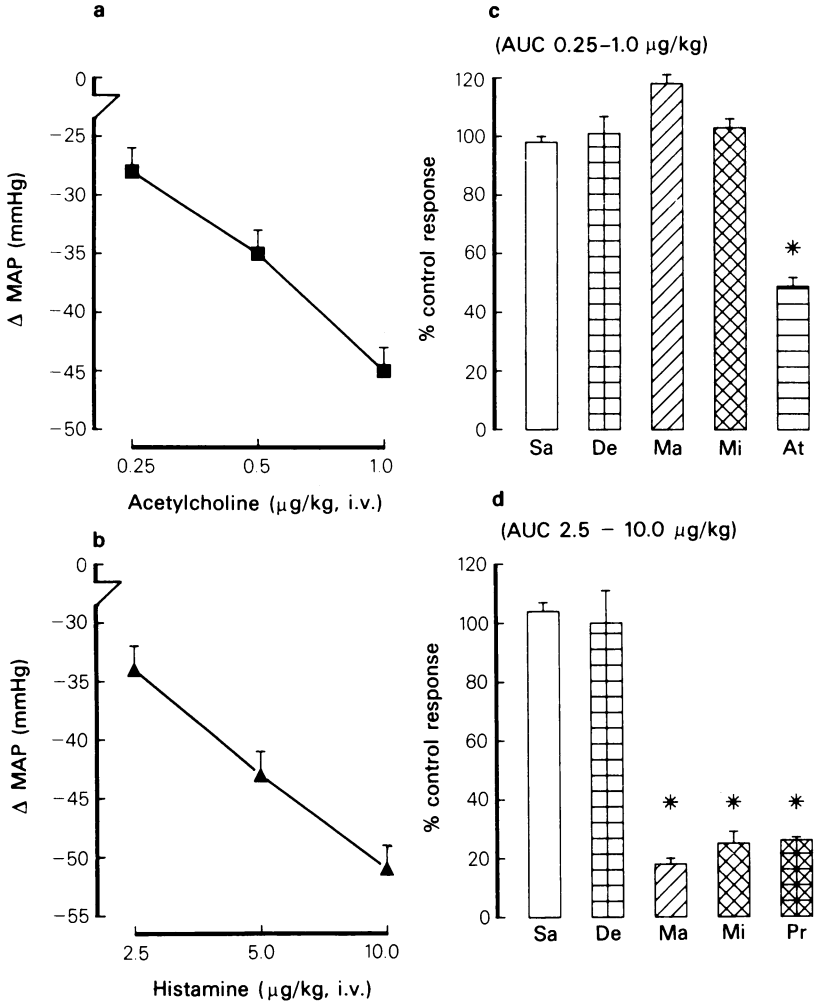

Figure 1 Change in mean carotid blood pressure (ΔMAP) evoked by acetylcholine and histamine (a and b) in intact rats ($n=21$). Effects of intravenous saline (Sa, 0.26 ml/kg), desipramine (De, 0.1 mg/kg), maprotiline (Ma, 0.5 mg/kg) and mianserin (Mi, 3.0 mg/kg) on the vasodepressor effects of acetylcholine (c) and histamine (d) in intact rats ($n=4–6$/group). Atropine (At, 0.003 mg/kg) and promethazine (Pr, 0.1 mg/kg) were used as standard antagonists of acetylcholine and histamine effects, respectively. For the expression of the results (% control response) see Methods. Response significantly decreased by the treatment: * $P<0.05$: Kruskall-Wallis test.

ramine (0.1 mg/kg), maprotiline (0.5 mg/kg), mianserin (0.01–0.3 mg/kg) or cyproheptadine (0.01 mg/kg). Pithed rats were prepared as described previously (Cavero *et al.*, 1980a).

Rats were pretreated with syrosingopine (5.0 mg/kg, i.p. 18 h before the experimental procedure) and prepared for blood pressure measurement as described above. 5-HT (20.0 μg/kg, i.v.) was administered before and after mianserin (0.1 and 10.0 mg/kg, i.v.).

The maximal doses of the three antidepressant agents used in this study produce significant potentiations of the heart rate responses to exogenous and endogenously released noradrenaline, due to inhibition of the noradrenaline neuronal uptake mechanism (Cavero *et al.*, 1980a).

Statistical analysis

Results are given as mean values ± s.e.means. To facilitate the expression of the results of the three point dose-peak response curves (Figures 1 and 2), the areas under these curves (AUC) before (control) and after the treatment were calculated (Cavero *et al.*, 1980a; Figure 4) and the effects of treatment expressed as a percentage of the control AUC response which was taken as 100%.

The significance of the effects were assessed by using Student's *t* test or the Kruskall-Wallis test. *P* values of less than 0.05 were considered statistically significant.

Drugs

The drugs used were: acetylcholine hydrobromide (Sigma), atropine sulphate (Prolabo), 5-hydroxytryptamine creatinine sulphate (Sigma), cyproheptadine (Merck Sharp & Dome), desipramine hydrochloride (Ciba-Geigy), histamine dihydrochloride (Prolabo), maprotiline (Ciba-Geigy), mianserin hydrochloride (Organon), promethazine (Boyer), pentobarbitone sodium (Nembutal, Abbott), syrosingopine base (Ciba-Geigy).

All the compounds were dissolved in saline; doses in the text refer to the bases of the compounds.

Results

Effects of desipramine, maprotiline and mianserin on the vasodepressor activity of acetylcholine and histamine in intact rats

The mean carotid blood pressure of the intact rats (anaesthetized with pentobarbitone) used in this study averaged 123 ± 3 mmHg ($n = 40$). Intravenous administrations of saline, desipramine (0.1 mg/kg), maprotiline (0.5 mg/kg) or mianserin (0.3–3.0 mg/kg) produced at the most a 10–15% increase in this parameter.

Intravenous injections of acetylcholine elicited dose-related vasodepressor responses that were not significantly modified by saline, desipramine, maprotiline and mianserin but were reduced by approximately 50% after only 3.0 μg/kg atropine, given intravenously (Figure 1).

The blood pressure lowering activity of histamine was not affected by saline or desipramine but was inhibited to a similar extent by intravenous maprotiline (0.5 mg/kg), mianserin (0.3 mg/kg) and promethazine (0.1 mg/kg). Thus, on a dose-basis mianserin and maprotiline are approximately 3 and 5 times less potent, respectively, than promethazine as antagonists of the vasodepressor action of histamine (Figure 1).

Effects of desipramine, maprotiline and mianserin on the pressor effects of 5-hydroxytryptamine in pithed rats

In the pithed rat (baseline mean blood pressure: 56 ± 1 mmHg, $n = 30$), 5-HT produced dose-related pressor responses which were slightly enhanced by desipramine, unaffected by maprotiline and reduced in a dose-related manner by mianserin (Figure 2). On a dose-basis, mianserin was similar in potency to cyproheptadine, a classical antagonist of 5-HT receptors producing pressor effects in the pithed rat. After mianserin, particularly in a higher dose, the responses to 5-HT were converted predominantly to falls in blood pressure. For instance, after intravenous mianserin (0.3 mg/kg), 5.0, 10.0 and 20.0 μg/kg of 5-HT given intravenously produced hypotensive responses of −11 ± 3, −15 ± 3, −14 ± 3 mmHg ($n = 3$). However, even after 0.01 mg/kg mianserin (i.v.), 5-HT (20.0 μg/kg, i.v.) produced a vasodepressor effect (−13 ± 1 mmHg) preceded by an increase in blood pressure (31 ± 3 mmHg, $n = 3$).

Effects of large doses of mianserin on the vasodepressor effects of 5-hydroxytryptamine unmasked by a small dose of mianserin in syrosingopine pretreated rats

This preparation was chosen to study the effects of mianserin on the vasodepressor effects of 5-HT since it has a significantly higher mean carotid blood pressure (81 ± 3 mmHg, $n = 5$) than the pithed rat (56 ± 1 mmHg, $n = 30$) ($P < 0.05$: unpaired *t* test). The effectiveness of syrosingopine in depleting peripheral stores of neuronal catecholamines was indicated by the poor blood pressure response to tyramine (0.3 mg/kg, i.v.) which was only 3 ± 1 mmHg ($n = 5$) in syrosingopine-pretreated rats in contrast to 36 ± 2 mmHg ($n = 6$) in untreated animals.

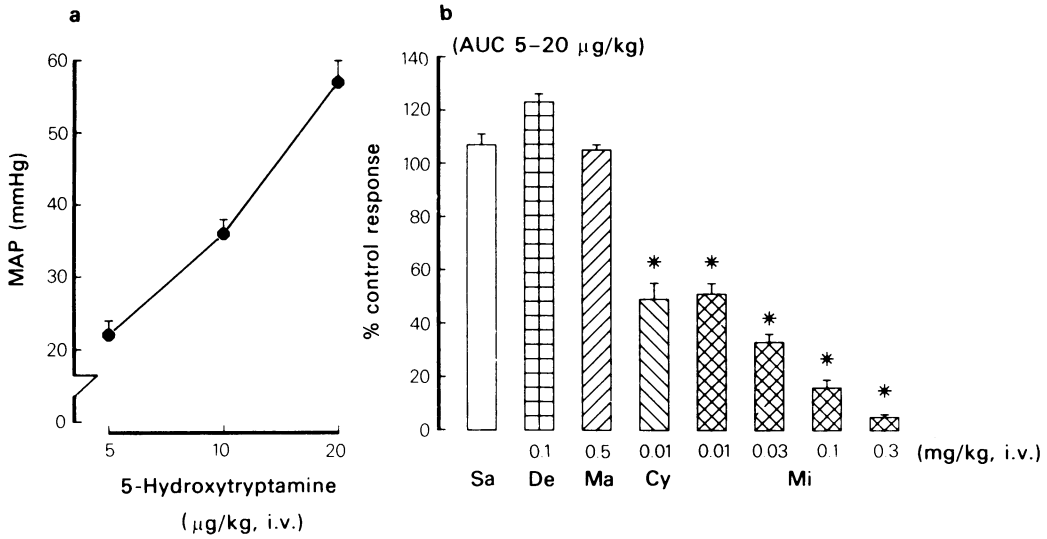

Figure 2 (a) Increases in mean carotid blood pressure (ΔMAP) produced by 5-hydroxytryptamine in pithed rats ($n=30$) and (b) effects of saline (Sa), desipramine (De), maprotiline (Ma) and mainserin (Mi) on vasopressor effects of 5-hydroxytryptamine in intact rats ($n=3-6$/group). Cyproheptadine (Cy) was used as a standard antagonist of 5-hydroxytryptamine. For the expression of results (% control response) see Methods. Response significantly decreased by the treatment: $^*P<0.05$: Kruskall-Wallis test.

Intravenous administration of 5-HT (20.0 μg/kg, i.v.) produced a biphasic blood pressure response in the syrosingopine pretreated rats that became exclusively vasodepressor after a small dose of mianserin (0.1 mg/kg, i.v.). The peak and the duration of the hypotensive effect of 5-HT were reduced by over 75% after mianserin 10.0 mg/kg, given intravenously (Figure 3). This inhibition cannot be due to an unfavourable change in baseline mean blood pressure since this parameter was significantly increased by mianserin from 89 ± 4 to 114 ± 12 mmHg ($n=5$) ($P<0.05$, paired t test).

Discussion

The antidepressant drugs, desipramine, maprotiline and mianserin, in doses that significantly inhibit the neuronal uptake of noradrenaline in pithed rats (Cavero *et al.*, 1980a), exhibited different spectra of peripheral autonomic receptor blocking properties. Whilst desipramine did not modify the blood pressure responses to acetylcholine, histamine or 5-HT, maprotiline and mianserin were histamine receptor antagonists and only mianserin inhibited the effects of 5-HT.

The lack of muscarinic receptor antagonist effects of desipramine (Pendleton, Miller & Ridley, 1980) and mianserin (Vargaftig *et al.*, 1971) are in agreement with published results obtained in a variety of pharmacological tests. Recently, relatively high concentrations of mianserin were found to increase the magnitude of the maximal contractile responses to acetylcholine and carbamylcholine without modifying the effects of noradrenaline in anococcygeus muscle. This enhanced sensitivity to muscarinic receptor agonists was suggested to occur at the level of, or distal to, the acetylcholine receptor (Doggrell, 1979). However, if mianserin acts distal to the receptor, the sequence of intracellular events following activation of muscarinic receptors is not likely to be the same in the vascular and anococcygeus muscle preparations since in the latter, acetylcholine produces contractions whereas in the entire vascular bed we studied it causes myorelaxation.

Whilst clinical investigations have generally excluded the possibility that mianserin therapy is accompanied by major side effects attributable to blockade or activation of acetylcholine receptors, there are objective data indicating that this antidepressant agent produces sedation (Peet & Behagel, 1978). This property of mianserin may be related to its relatively potent histamine and 5-HT receptor antagonist activity (Peet & Behagel, 1978) which can be demonstrated both *in vivo* and *in vitro* (Vargaftig *et al.*, 1971; Figge *et al.*, 1979; Tang & Seeman, 1980).

Mianserin resulted from a chemical project initially aimed at finding a highly potent 5-HT receptor antagonist (Vargaftig *et al.*, 1971) and it is 5-HT antagonism which clearly distinguishes mianserin from desipramine and maprotiline. In the pithed rat

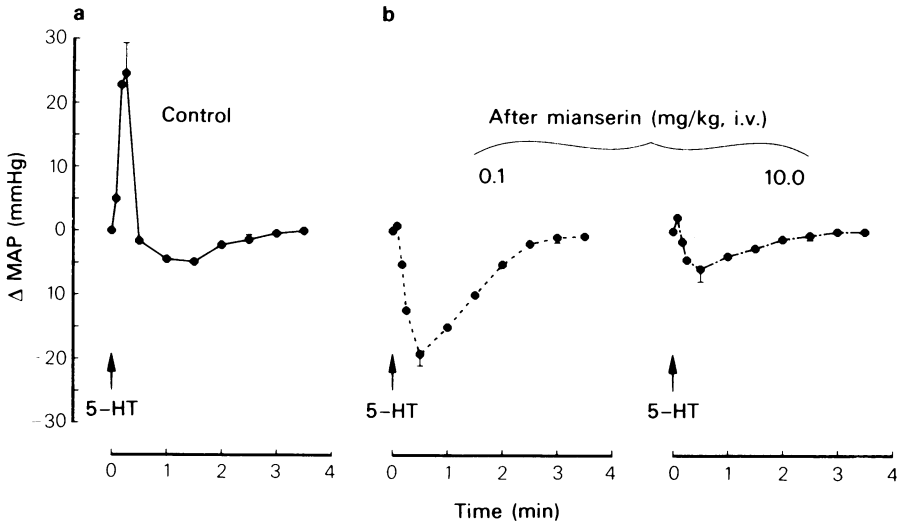

Figure 3 Time course of the mean carotid blood pressure effects (ΔMAP) of 5-hydroxytryptamine (5-HT) (20 μg/kg, i.v.) in syrosingopine pretreated rats ($n = 5$) before (a) and after (b) injection of a small followed by a large dose of mianserin (0.1 and 10.0 mg/kg, i.v.). The peak vasodepressor response produced by 5-HT after 0.1 mg/kg, mianserin (i.v.) was significantly inhibited by the higher dose of mianserin ($P < 0.05$: t test).

preparation, 5-HT produces pressor responses that are due to stimulation of receptors specific for this indolamine. On the basis of results obtained in a series of appropriate pharmacological tests, Vargaftig *et al.* (1971) proposed that mianserin inhibits only responses elicited by stimulation of 5-HT D-receptors although it failed to counteract the D-receptor-mediated effects in rat stomach fundus strips.

Traditionally, 5-HT is considered to produce its effects through activation of two distinct types of receptors, designated M-neuronal and D-muscular tryptamine receptors. The M-receptors situated in nervous tissue are blocked by morphine, cocaine and atropine, whereas the antagonists of D-receptors situated in smooth muscle are methysergide, dibenamine and 2-bromolysergic acid diethylamide (Gaddum & Picarelli, 1957; Day & Vane, 1963). However, this classification no longer appears to be adequate to account for all available experimental results. For instance, Feniuk, Humphrey & Watts (1979) showed that presynaptic 5-HT receptors, the stimulation of which decreases the release of noradrenaline from sympathetic nerves in the isolated saphenous vein preparation of the dog were resistant to blockade by classical 5-HT antagonists.

In the rat cardiovascular system, 5-HT produces pressor responses that are blocked by small doses of mianserin, methysergide or cyproheptadine suggesting that D-muscular tryptamine receptors are responsible for this action of 5-HT (Cavero *et al.*, 1980b; present results). It is proposed that this antagonism occurs on an almost 1:1 molar basis since the doses of the antagonist, mianserin, did not differ from those of the agonist, 5-HT.

In syrosingopine pretreated rats, 5-HT produced a biphasic blood pressure response, a rise followed by a fall, the latter phase becoming predominant after treating the animals with a small dose of either mianserin or methysergide. This hypotensive effect of 5-HT was inhibited by high doses of mianserin (present results) or methysergide and was not modified by blockade of vascular β_2-adrenoceptors, histamine or muscarinic receptors or inhibition of formation of endogenous prostaglandin-like substances with indomethacin (Cavero *et al.*, 1980b). These results exclude the possibility that M-receptors or classical receptors and mechanisms known to mediate falls in blood pressure are responsible for the vasodepressor activity of 5-HT in the rat.

Fozard & Leach (1968) found that 5-HT reduced blood pressure in the pithed rat. These authors attributed this effect to a pulmonary vasoconstrictor activity of 5-HT which would lead to a fall in cardiac output and, thereby, in systemic blood pressure. If this is the physiological mechanism of the hypotensive action of 5-HT observed after a small dose of mianserin or methysergide, then, the 5-HT receptors of the pulmonary vascular bed are unlikely to be of the same D-type family as those mediating the peripheral vasoconstrictor responses to 5-HT. In fact, the responses evoked by the postulated 5-HT receptors on pulmonary vessels would appear to be blocked by doses of mianserin or methysergide which

are at least 100 times higher than those required to impair 5-HT receptors mediating blood pressure increases at the level of peripheral resistance vessels (Cavero *et al.*, 1980b). Evidence for two distinct types of excitatory receptors for 5-HT was given by Apperley, Feniuk, Humphrey & Levy (1980) on the basis of results obtained in isolated vascular preparations of the dog. Alternatively, the fall in blood pressure produced by 5-HT after blockade of its vasoconstrictor effects could be mediated by a postsynaptic 5-HT receptor, the stimulation of which relaxes the vascular smooth muscle. This receptor would be located postsynaptically since no significant degree of sympathetic tone was present in these animals due to the pretreatment with syrosingopine.

There are experimental results suggesting that 5-HT mediates myorelaxation in certain vascular tissues (Vargaftig & Lefort, 1974; Chand, 1981). More studies appear to be required in order to clarify the physiological and pharmacological mechanisms responsible for the blood pressure-lowering effects of 5-HT in the pithed or syrosingopine pretreated rat given a small dose of a 5-HT D-receptor antagonist.

The authors express their gratitude to Mrs J. Lechaire and Miss C. Thiry for their technical assistance and to Mrs N. Sieller for her secretarial help.

Desipramine, maprotiline and phentolamine were generously provided by Ciba-Geigy (Basle, Switzerland), mianserin by Organon and cyproheptadine by Merck Sharp & Dohme (Paris, France).

References

APPERLEY, E., FENIUK, W., HUMPHREY, P.P.A. & LEVY, G.P. (1980). Evidence for two types of excitatory receptor for 5-hydroxytryptamine in dog isolated vasculature. *Br. J. Pharmac.*, **68,** 215–224.

BROGDEN, R.N., HEEL, R.C., SPEIGHT, T.M. & AVERY, G.S. (1978). Mianserin: A review of its pharmacological properties and therapeutic efficacy in depressive states. *Drugs*, **16,** 273–301.

CAVERO, I., GOMENI, R., LEFÈVRE-BORG, F. & ROACH, A.G. (1980a). Comparison of mianserin with desipramine, maprotiline and phentolamine on cardiac presynaptic and vascular postsynaptic α-adrenoceptors and noradrenaline reuptake in pithed normotensive rats. *Br. J. Pharmac.*, **68,** 321–332.

CAVERO, I., LEFÈVRE-BORG, F. & ROACH, A.G. (1980b). Comparison of mianserin with desipramine and maprotiline on blood pressure responses to acetylcholine, histamine and 5-hydroxytryptamine in normotensive rats. *Br. J. Pharmac.*, **69,** 342–343P.

CHAND, N. (1981). 5-Hydroxytryptamine induces relaxation of goat pulmonary veins: evidence for the non-involvement of M and D-tryptamine receptors. *Br. J. Pharmac.*, **72,** 233–237.

DAY, M. & VANE, J.R. (1963). An analysis of the direct and indirect actions of drugs on the isolated guinea-pig ileum. *Br. J. Pharmac. Chemother.*, **20,** 150–170.

DOCHERTY, J.R. & McGRATH, J.C. (1980). An examination of factors influencing adrenergic transmission in the pithed rat, with special reference to noradrenaline uptake mechanisms and post-junctional α-adrenoceptors. *Naunyn-Schmiedebergs Arch. Pharmac.*, **313,** 101–111.

DOGGRELL, S.A. (1979). Mianserin potentiates responses to acetylcholine in the rat anococcygeus muscle. *Eur. J. Pharmac.*, **55,** 311–314.

DOXEY, J.C., EVERITT, J. & METCALF, G. (1978). Mianserin—an analysis of its peripheral autonomic actions. *Eur. J. Pharmac.*, **51,** 1–10.

FENIUK, W., HUMPHREY, P.P.A. & WATTS, A.D. (1979). Presynaptic inhibitory action of 5-hydroxytryptamine in dog isolated saphenous vein. *Br. J. Pharmac.*, **67,** 247–254.

FIGGE, J., LEONARD, P. & RICHELSON, E. (1979). Tricyclic antidepressants: potent blockade of histamine H_1 receptors of guinea pig ileum. *Eur. J. Pharmac.*, **58,** 479–483.

FOZARD, J.R. & LEACH, G.D.H. (1968). The hypotensive action of 5-hydroxytryptamine (5-HT) in anaesthetized and pithed rats. *Eur. J. Pharmac.*, **2,** 239–249.

GADDUM, J.H. & PICARELLI, Z.P. (1957). Two kinds of tryptamine receptors. *Br. J. Pharmac. Chemother.*, **12,** 323–328.

PENDLETON, R.G., MILLER, D.A. & RIDLEY, P.T. (1980). Studies on the putative anticholinergic effects of desmethylimipramine. *Archs int. Pharmacodyn.*, **243,** 37–47.

PEET, M. & BEHAGEL, H. (1978). Mianserin: a decade of scientific development. *Br. J. clin. Pharmac.*, **5,** 5–9S.

ROBSON, R.D., ANTONACCIO, M.J., SAELENS, J.K. & LIEBMAN, J. (1978). Antagonism by mianserin and classical α-adrenoceptor blocking drugs of some cardiovascular and behavioral effects of clonidine. *Eur. J. Pharmac.*, **47,** 431–442.

TANG, S.W. & SEEMAN, P. (1980). Effect of antidepressant drugs on serotonergic and adrenergic receptors. *Naunyn Schmiedebergs Arch. Pharmac.*, **311,** 255–261.

VARGAFTIG, B.B., COIGNET, J.L., DE VOS, G.J., GRIJSEN, H. & BONTA, I.L. (1971). Mianserin hydrochloride: peripheral and central effects in relation to antagonism against 5-hydroxytryptamine and tryptamine. *Eur. J. Pharmac.*, **6,** 336–346.

VARGAFTIG, B.B. & LEFORT, J. (1974). Pharmacological evidence for a vasodilator receptor to serotonin in the nasal vessels of the dog. *Eur. J. Pharmac.*, **27,** 216–225.

(Received March 27, 1981.
Revised May 6, 1981.)