ELSEVIER

European Journal of Pharmacology 276 (1995) 215–221

ejp

Studies on the protective effect of azepexole on ouabain-induced cardiac arrhythmias and lethality in guinea-pig

George P. Thomas *

Department of Pharmacology, IDPL Research Centre, Hyderabad-500 037, India

Received 11 August 1994; revised 29 December 1994; accepted 6 January 1995

Abstract

Azepexole, an α_2-adrenoceptor agonist (125, 250 and 500 μg/kg i.v.), was examined for its effect on ouabain-induced ventricular premature beats, ventricular tachyarrhythmias and lethality in guinea-pigs. The doses of ouabain required to cause ventricular arrhythmias and lethality were significantly higher in azepexole-treated animals. However, it did not offer any protection in reserpinised guinea-pigs. Idazoxan, the α_2-adrenoceptor antagonist (100 μg/kg i.v.) inhibited the protective action of azepexole while corynanthine, the α_1-adrenoceptor antagonist (1 mg/kg i.v.), potentiated the effect. Azepexole inhibited the rate of the ouabain-induced rise in mean arterial blood pressure and the peak pressor response. In isolated paced left atria of guinea-pig, azepexole (2.76×10^{-3} M) did not offer any protection against extrasystolic contractions induced by ouabain. Therefore the protective effect of azepexole may be mediated through the stimulation of α_2-adrenoceptors and the resultant suppression of the indirect neural components of ouabain toxicity.

Keywords: Arrhythmia; Digitalis; Ouabain; Azepexole; B-HT 933

1. Introduction

Centrally mediated alterations of the autonomic nervous system, mainly by the release of catecholamines and other neurotransmitters are considered to be a major cause of digitalis-induced ventricular arrhythmias (Saxena and Bhargava, 1975; Gillis and Quest, 1980). Many groups of drugs which can decrease sympathetic activity such as β-adrenoceptor antagonists (Sekiya and Vaughan Williams, 1963; Dohadwalla et al., 1969), ganglion blocking agents (Gillis et al., 1975) and drugs that interfere with catecholamine storage and release viz. reserpine (Dogget and Case, 1975), 6-hydroxydopamine (Saito et al., 1974), guanethidine (Raines et al., 1968), bretylium (Papp and Vaughan Williams, 1969) and α-methyl-*m*-tyrosine (Ciofalo and Treece, 1970) were found to induce significant protective effect against digitalis-induced arrhythmias. Clonidine (Lechat and Schmitt, 1982; Thomas and Tripathi, 1986) and some of its derivatives like flutonidine (Thomas and Varma, 1993) and St-93 (Thomas and Stephen, 1993) were shown to increase the doses of ouabain required to cause ventricular arrhythmias and lethality.

The azepine derivative azepexole (B-HT 933) has a chemical structure different from clonidine but exerts typical clonidine-like actions on the cardiovascular system (Kobinger and Pichler, 1977; Kobinger, 1978). It is chemically 2-amino-6-ethyl-5,6,7,8-tetrahydro-4*H*-oxazolo[4,5-*d*] azepine and is more selective to α_2-adrenoceptors than clonidine (Kobinger and Pichler, 1980a; Timmermans and Van Zwieten, 1980; Deniards et al., 1983). It exhibits a 300-fold selectivity for the α_2- over the α_1-adrenoceptors (Rhodes, 1986). Pharmacological studies indicate its selectivity for α_2-adrenoceptors both in the central nervous system and in the periphery (Kobinger and Pichler, 1980b; Andén et al., 1982). It was reported that azepexole has no protective effect against digoxin-induced arrhythmias in guinea-pigs (Plunkett and Tackett, 1983). Contrary to their results, azepexole exhibited significant antiarrhythmic effects against ouabain-induced arrhythmias and lethality in the present study. This study also elucidates its possible mode of action.

* Corresponding author. Present address: Department of Pharmacology, Sir Charles Tupper Medical Building, Dalhousie University, Halifax, N.S., Canada B3H 4H7.

SSDI 0014-2999(95)00025-9

2. Materials and methods

2.1. Ventricular arrhythmias and lethality induced by ouabain

Guinea-pigs of either sex (350–450 g) were used in this study. The method described by Thomas and Tripathi (1986) was utilized. The animals were anaesthetized by pentobarbitone sodium and positive pressure artificial respiration was maintained throughout the experiment. The mean arterial blood pressure was recorded on a Gemini Recorder (Ugo Basile, Model 7070) through a Bentley Trantec physiological pressure transducer and limb lead II electrocardiogram was recorded on a Grass Polygraph (Model 7D). Ouabain solution (80 μg/ml) was continuously infused at a rate of 100 μl/min. The amount of ouabain required per kg body weight to cause ventricular premature beats, ventricular tachyarrhythmias (denoted by ventricular tachycardia or ventricular fibrillation associated with a sudden fall in blood pressure) and lethality was determined in control and drug-treated animals. Azepexole was administered intravenously 10 min prior to ouabain infusion and the haemodynamic effects of the drug were also observed.

2.2. Ouabain-induced rise in mean arterial blood pressure

During the experiment (1) above, mean arterial blood pressure was recorded at 2 min intervals throughout the infusion period of ouabain until the onset of ventricular tachyarrhythmia. The rate of rise of mean arterial blood pressure induced by ouabain and the peak pressor effect reached were noted for control and azepexole (250 μg/kg)-treated guinea-pigs.

2.3. Effect in reserpinised guinea-pigs

Two groups of six guinea-pigs each were administered reserpine (5 mg/kg i.m.) 24 h before the experiment. The experimental procedure was the same as in 2.1. above. The first group was administered normal saline and the second group azepexole (250 μg/kg), 10 min prior to ouabain infusion.

2.4. Effect after α-adrenoceptor blockade

α_1-Adrenoceptor antagonist

Corynanthine hydrochloride (1 mg/kg i.v.) was administered to six animals, 10 min before azepexole (250 μg/kg), while another three groups of six animals each were administered azepexole, corynanthine and normal saline, respectively, before ouabain infusion.

α_2-Adrenoceptor antagonist

Idazoxan (100 μg/kg i.v.) was administered to six guinea-pigs and after 10 min, azepexole was administered at a dose of 250 μg/kg. Another three groups of six animals each were also studied, one as control and the others were treated with azepexole and idazoxan, respectively, before ouabain infusion.

2.5. Ouabain-induced arrhythmias in isolated paced left atrium of guinea-pig

The method described by Thomas and Varma (1991) was employed. Briefly, the left atrium from adult guinea-pigs were dissected free and hung vertically on a stimulating electrode and the free end was tied to an isometric transducer (Type DYO, Ugo Basile, Italy) in an organ bath containing Ringer Locke solution at 37 ± 1°C bubbled with oxygen. The atria were driven at a constant rate of 1 Hz of 2 ms duration at twice the threshold voltage delivered by a square wave pulse generator (Grass, Model S44, USA). Ouabain at a concentration of 10^{-6} M was added and the atrial contractions were recorded on a microdynamometer (Ugo Basile). The time taken for the development of premature beats was noted. After a stabilization period of 60 min, azepexole at concentrations of 2.76×10^{-5}, 2.76×10^{-4} and 2.76×10^{-3} M ($n = 5$ for each concentration in separate experiments) was added to the bath, challenged with ouabain and differences in the onset or duration, if any, were assessed. A set of six control experiments were also conducted.

The drugs used in this study were: azepexole (Boehringer Ingelheim), corynanthine hydrochloride (Sigma), idazoxan (Reckitt and Colman), ouabain octahydrate (Sigma) and reserpine (Boehringer Ingelheim). All the drugs except reserpine were dissolved in normal saline for intravenous infusion. Reserpine was dissolved in a minimal amount of glacial acetic acid, the pH of which was adjusted to 5.5 by 0.1 N NaOH and diluted with distilled water to give a concentration of 1 mg/ml.

2.6. Statistics

The results were statistically analyzed using Student's t-test. Probability was established when the value was less than 0.05.

3. Results

3.1. Haemodynamic effects of azepexole in guinea-pig

Azepexole produced a biphasic response on mean arterial blood pressure. The initial rise in mean arterial blood pressure was followed by a gradual but consistent fall (Table 1). There was significant reduction in the heart rate immediately after intravenous administration of azepexole. Mean arterial blood pressure and

Table 1
Effect of azepexole on mean arterial blood pressure and heart rate in guinea-pigs

Dose (μg/kg)	n	Predrug	Postdrug		
			1 min	5 min	10 min
Mean arterial blood pressure (mm Hg)					
125	5	45± 3	50±3	29±2 [a]	30±2 [b]
250	6	44± 3	49±2	29±1 [b]	30±1 [b]
500	6	45± 3	53±3 [a]	24±3 [c]	27±3 [c]
Heart rate (bt / min)					
125	6	255± 7	229±4 [b]	214±4 [c]	212±5 [c]
250	6	240±10	226±8 [a]	223±6 [a]	211±8 [b]
500	6	250± 4	228±5 [a]	205±7 [b]	202±7 [b]

[a] $P < 0.05$, [b] $P < 0.01$ and [c] $P < 0.001$ compared to respective predrug control values (paired t-test).

heart rate values at the end of 5 and 10 min were significantly lower as compared to their respective predrug values at 125, 250 and 500 μg/kg doses. Azepexole did not induce any significant change in the electrocardiogram of guinea-pigs.

3.2. Effect of azepexole on ouabain-induced cardiac arrhythmias and lethality

At doses of 125, 250 and 500 μg/kg, azepexole increased the doses of ouabain required for the onset of ventricular premature beats and ventricular tachyarrhythmia compared to that of the control group (Table 2). However, at lower doses it was not effective. Similarly, azepexole at doses of 125, 250 and 500 μg/kg, caused significant increases in the doses of ouabain required to cause lethality (Fig. 1).

3.3. Effect of azepexole on the ouabain-induced rise in mean arterial blood pressure

Intravenous administration of ouabain produced a gradual increase in blood pressure in guinea-pigs. It attained a maximum peak before the onset of ventricular tachyarrhythmia and fell subsequently along with ventricular tachyarrhythmia. Azepexole induced significant inhibition in the rate of rise of blood pressure induced by ouabain (Fig. 2). Similarly, the peak pressor response caused by ouabain was also significantly lower ($P < 0.01$) in azepexole-treated guinea-pigs (30.8 ± 2.8 mm Hg) compared to that of control animals (49.0 ± 4.6 mm Hg).

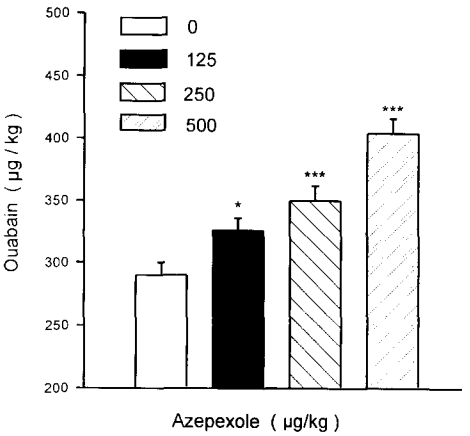

Fig. 1. Effect of i.v. administration of azepexole on ouabain-induced lethality in guinea-pig. * $P < 0.05$, *** $P < 0.001$ compared to control. Values are expressed as mean ± S.E.M. of the doses of ouabain (μg/kg body weight) required to cause lethality (n: Control – 11, 125 μg – 9, 250 μg – 8 and 500 μg – 8).

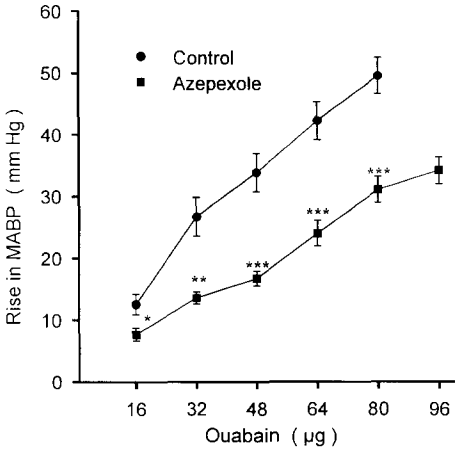

Fig. 2. Effect of azepexole (250 μg/kg i.v.) on the ouabain-induced rise of mean arterial blood pressure (MABP) in guinea-pig. Values are expressed as the means of five to six experiments. Vertical bars show S.E.M.

3.4. Interaction studies

In reserpinised guinea-pigs, the doses of ouabain required to induce ventricular premature beats, ven-

Table 2
Effect of azepexole on ouabain-induced ventricular arrhythmias in guinea-pigs

Drug (dose)	n	Ventricular premature beats	Ventricular tachyarrhythmias
Control	11	172.8 ± 3.7	211.4 ± 8.5
Azepexole (μg / kg)			
125	9	203.5 ± 5.5 [c]	258.7 ± 12.3 [b]
250	8	220.6 ± 6.4 [c]	278.3 ± 13.9 [c]
500	8	260.4 ± 12.1 [c]	349.8 ± 13.2 [c]

[b] $P < 0.01$ and [c] $P < 0.001$ compared to control (unpaired t-test). Values are expressed as the means ± S.E.M. of the doses of ouabain (μg/kg body weight) required to cause the arrhythmic stages.

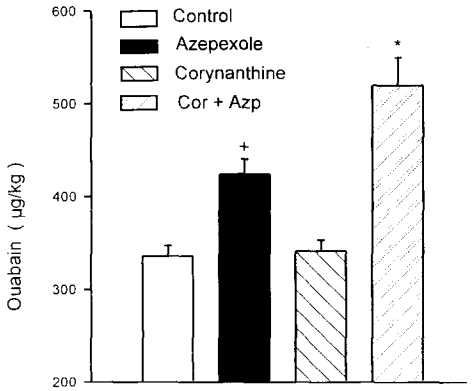

Fig. 3. Effect of corynanthine (Cor) (1 mg/kg i.v.) on the protective action of azepexole (Azp) (250 μg/kg i.v.) against ouabain-induced lethality in guinea-pig. Values are expressed as the means and S.E.M. of the doses of ouabain (μg/kg body weight) that caused lethality. $^{+}$ $P < 0.05$ compared to control ($n = 6$), and * $P < 0.05$ compared to azepexole ($n = 5$).

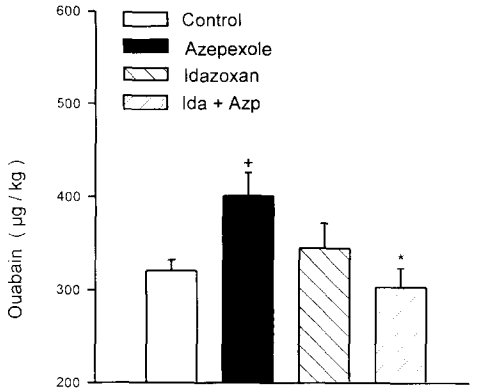

Fig. 4. Effect of idazoxan (Ida) (100 μg/kg i.v.) on the protective action of azepexole (Azp) (250 μg/kg i.v.) against ouabain-induced lethality in guinea-pig. Values are expressed as the means and S.E.M. of the doses of ouabain (μg/kg body weight) that caused lethality. $^{+}$ $P < 0.05$ compared to control ($n = 6$) and * $P < 0.05$ compared to azepexole ($n = 6$).

tricular tachyarrhythmia and lethality were higher (235.30 ± 8.60, 412.00 ± 22.70 and 467.60 ± 13.50, respectively) compared to controls. Azepexole did not alter the arrhythmogenic and lethal effects of ouabain in reserpinised animals (245.20 ± 11.70, 400.60 ± 18.20 and 481.60 ± 11.30 for ventricular premature beats, ventricular tachyarrhythmias and lethality, respectively). Corynanthine, an α_1-adrenoceptor blocking agent, not only failed to inhibit the protective effect of azepexole (Table 3), but it also induced significant potentiation of the protective effect of azepexole ($P < 0.05$ for ventricular premature beats and $P < 0.01$ for ventricular tachyarrhythmia) whereas idazoxan, the specific α_2-adrenoceptor antagonist, showed significant inhibition ($P < 0.05$ for ventricular premature beats and $P < 0.01$ for ventricular tachyarrhythmia) of the antiarrhythmic effect of azepexole (Table 3). Similarly, corynanthine potentiated the protective effect of azepexole against lethality induced by ouabain (Fig. 3) while idazoxan showed significant inhibition (Fig. 4).

3.5. Effect of azepexole on ouabain-induced extrasystolic contractions in isolated paced left atrium of guinea-pig

Control preparations developed extrasystolic contractions on exposure to ouabain for 6.78 ± 1.21 min. Azepexole at concentrations of 2.76×10^{-5}, 2.76×10^{-4} and 2.76×10^{-3} M, did not accord any protection against ouabain-induced extrasystolic contractions in isolated paced left atrium of guinea-pigs. At the highest concentration of azepexole, the incidence of extrasystolic contractions was at 6.94 ± 0.70 min. Similarly, the duration of the extrasystolic contractions also remained unaltered with all the three concentrations of azepexole.

4. Discussion

In the present study, azepexole showed significant protection against the arrhythmogenic and lethal ef-

Table 3
Effects of corynanthine and idazoxan on the antiarrhythmic effect of azepexole

Drug (dose/kg)	n	Ventricular premature beats	Ventricular tachyarrhytmia
Control	6	197.1 ± 9.2	260.3 ± 16.0
Azepexole (250 μg)	6	261.9 ± 9.0	349.4 ± 16.4
Corynanthine (1 mg)	6	176.5 ± 6.9	264.6 ± 16.0
Corynanthine (1 mg) + azepexole (250 μg)	5	297.6 ± 11.0 [a]	454.7 ± 24.8 [b]
Control	6	188.6 ± 6.7	234.3 ± 6.6
Azepexole (250 μg)	6	259.4 ± 20.4	337.0 ± 28.7
Idazoxan (100 μg)	5	208.6 ± 12.9	252.2 ± 12.7
Idazoxan (100 μg) + azepexole (250 μg)	6	203.0 ± 6.1 [a]	238.5 ± 7.2 [b]

[a] $P < 0.05$ and [b] $P < 0.01$ compared to azepexole. Values are expressed as the means ± S.E.M. of the doses of ouabain (μg/kg body weight) required to cause the arrhythmic stages.

fects of ouabain. This is in contrast to the previous finding of Plunkett and Tackett (1983) where azepexole at a dose of 5 μg/kg (i.v.) failed to alter the toxic and lethal effects of digoxin in their study. Even though azepexole is highly selective for α_2-adrenoceptors (Rhodes, 1986), its relative potency is 0.006 compared to clonidine (Kobinger and Pichler, 1977). Kobinger and Pichler (1977) used 300 μg/kg (i.v.) of azepexole in cats and dogs and 500 μg/kg (i.v.) in rats, for inducing significant centrally mediated cardiovascular effects. Van Zwieten and Timmermans (1980) observed that 30 μg/kg administered through cat's vertebral artery produced a significant hypotensive effect, whereas the same dose administered intravenously did not induce any significant effect. Thus a dose of 5 μg/kg used intravenously by Plunkett and Tackett (1983) in their study is insufficient to produce any significant centrally mediated cardiovascular effects. In preliminary studies it was observed that in guinea-pig 62.5 μg/kg failed to induce any significant hypotensive or antiarrhythmic effects. So in the present study, azepexole was administered intravenously at doses of 125, 250 and 500 μg/kg.

In all the above doses, azepexole produced significant protection against the cardiotoxic effects of ouabain. It was observed that the hypotensive effect of azepexole was not dose-dependent in these doses. The blood pressure values in anaesthetized guinea-pigs are relatively low (40–55 mm Hg), giving little scope for a dose-dependent lowering of blood pressure in the dose range used in this study. Moreover, the mean arterial blood pressure readings, taken 10 min after intravenous administration of azepexole, may not represent the peak hypotensive effect of the drug. The species specific factors such as the negligible vagal tone (Tripathi et al., 1984) and the role endothelin plays in the maintenance of blood pressure (Veniant et al., 1994) also can modify the blood pressure response in guinea-pig depending on the mode of action of the test drug. Thus the blood pressure values in this study serve only as an indicator of the centrally mediated effect. Higher doses of azepexole were not used, in order to avoid possible non-specific effects.

Digitalis-induced arrhythmias are the net result of an interplay of its effects on the myocardium (Ferrier, 1977; Smith et al., 1984) the central nervous system (Gillis and Quest, 1980) and the autonomic nervous system (Saxena and Bhargava, 1975). Although it has been suggested that an alteration in the function of the sympathetic nervous system is the primary mechanism by which ventricular arrhythmias are generated by digitalis (Saxena and Bhargava, 1975), it can act directly on myocardium to induce changes in automaticity, conduction and/or the effective refractory period and thereby elicit arrhythmias. Premature beats induced by ouabain in left atrium (Thomas and Varma, 1991) are the result of a direct effect of ouabain as they exclude the indirectly mediated effects of the central nervous system and the autonomic nervous system. In the present study, azepexole, up to a concentration of 2.76×10^{-3} M, failed to offer any protective effect against ouabain-induced arrhythmias in left atrium, indicating that azepexole may be acting upon the indirect components of the toxic effects of ouabain. These results also indicate that the protective effect of azepexole is not due to any direct effect on the myocardium.

In reserpinised guinea-pigs, azepexole was unable to alter the arrhythmogenic or lethal doses of ouabain. The absence of any protective effect in reserpinised animals supports the assumption that it is the indirect components of ouabain toxicity that are affected by azepexole, as the effect produced by ouabain in reserpinised animals would be comparable to a situation where indirect components of its toxic effects are absent or inhibited.

The involvement of α_1-adrenoceptors in some of the pharmacological actions of clonidine-like drugs have been demonstrated (Drew, 1976; Hamilton and Longman, 1982). At a dose of 1 mg/kg, corynanthine, an α_1-adrenoceptor antagonist (Weitzell et al., 1979), did not have any effect on ouabain-induced arrhythmic stages and lethality, whereas it potentiated the protective effect of azepexole. Prazosin is reported to cause a significant antiarrhythmic effect against ouabain-induced arrhythmias (Lechat and Schmitt, 1982; Thomas and Tripathi, 1986). α_2-Adrenoceptor agonists can inhibit the release of catecholamines and α_1- adrenoceptor antagonists can inhibit the action of any released catecholamines at effector sites. The potentiation of the protective effect of azepexole by corynanthine, observed in this study may be due to its antagonism at α_1-adrenoceptors, preventing activation by any released catecholamines.

Idazoxan, the selective α_2-antagonist (Al-Damluji et al., 1988), completely abolished the protective effect of azepexole against arrhythmias and lethality, indicating the involvement of α_2-adrenoceptors in its protective action.

In the present study, azepexole significantly inhibited the pressor effect induced by ouabain. The ouabain-induced rise in blood pressure in guinea-pigs is mainly due to the activation of the sympathetic nervous system (Trzeciakowski, 1985). Thus the simultaneous inhibition of the pressor effects and reduction in the arrhythmogenic and lethal effects of ouabain indicate that both may be the consequence of the suppression of ouabain-induced sympathetic stimulation by azepexole.

Most of the evidence derived from studies of digitalis overdosage in neurally intact animals firmly suggest a distinct contributory role of the adrenergic nervous system in the genesis of cardiac arrhythmias (Cagin

et al., 1974; Gillis and Quest, 1980; Sivam et al., 1980). All the clonidine-like centrally acting antihypertensive drugs are known to act on the central α_2-adrenoceptors in the brainstem to reduce sympathetic tone and thereby lowering blood pressure (Kobinger, 1978; Isaac, 1980; Van Zwieten and Timmermans, 1983). Azepexole decreases sympathetic tone by stimulation of α_2-adrenoceptors (Pichler et al., 1980). Thus, it is possible that the reduction in the sympathetic tone is the cause of its protective effect against the arrhythmogenic and lethal effects of ouabain.

Higher concentrations of digitalis often induce ventricular arrhythmias in patients. Withdrawal of digitalis and the use of lignocaine or phenytoin are indicated in their treatment. This study shows the effectiveness of azepexole against ouabain-induced ventricular arrhythmias and points to its potential as an antiarrhythmic in such clinical conditions. Clonidine which has been shown to have a significant antiarrhythmic effect against digitalis-induced experimental arrhythmias, can produce abnormalities like sinus bradycardia and sinus arrest in patients treated for cardiovascular disorders. Since the pharmacological profile of azepexole is very similar to that of clonidine, further studies and careful analysis of potential side effects are required to establish the clinical utility of azepexole.

Acknowledgements

The generous gifts of azepexole by Boehringer Ingelheim and idazoxan by Reckitt and Colman are gratefully acknowledged. The author is also thankful to Dr R.K. Varma for timely advice and Mr. P. Narayana Rao for skilled technical assistance. Communication No. 196 from IDPL Research Centre, Hyderabad, India.

References

Al-Damluji, S., G. Ross, R. Touzel, D. Perret, A. White and G.M. Besser, 1988, Modulation of the actions of tyrosine by alpha$_2$ adrenoceptor blockade, Br. J. Pharmacol. **95**, 405.

Andén, N.E., K. Golembiowska-nikitin and U. Thornstrum, 1982, Selective stimulation of dopamine and noradrenaline autoreceptors by B-HT 920 and B-HT 933 respectively, Naunyn-Schmied. Arch. Pharmacol. 321, 100.

Cagin, W.A., J.C. Somberg, H. Bounous, T. Mittag, A. Raines and B. Levitt, 1974, The influence of spinal cord transection on the capacity of digitoxin to induce cardiotoxicity, Arch. Int. Pharmacodyn. Ther. 207, 340.

Ciofalo, F.R. and G. Treece, 1970, Ouabain induced arrhythmias in the rabbit: effects of alpha methyl-metatyrosine, Eur. J. Pharmacol. 9, 297.

Deniards, M.J., J. Meiguen and F. De Fendis, 1983, Reversal of reserpine induced ptosis in the mouse by alpha adrenoceptor agonists, Psychopharmacology 80, 243.

Dogget, N.S. and G. Case, 1975, Some observations on the interaction between cardiac glycosides and reserpine in the heart and nervous system, Toxicol. Appl. Pharmacol. 33, 87.

Dohadwalla, A.N., A.S. Freedberg and E.M. Vaughan Williams, 1969, The relevance of beta blockade to ouabain induced cardiac arrhythmias, Br. J. Pharmacol. 36, 257.

Drew, G.M., 1976, Effects of alpha adrenoceptor agonists and antagonists on pre- and post-synaptically located alpha adrenoceptors, Eur. J. Pharmacol. 36, 313.

Ferrier, G.R., 1977, Digitalis arrhythmias. Role of oscillatory afterpotentials, Prog. Cardiovasc. Dis. 19, 459.

Gillis, R.A., H. Jolson, H. Thibodeaux and B. Levitt, 1975, Antagonism of deslanoside induced cardiotoxicity by combined nicotinic and muscarinic blockade of autonomic ganglia, J. Pharmacol. Exp. Ther. 195, 126.

Gillis, R.A. and J.A. Quest, 1980, The role of the nervous system to the cardiovascular effects of digitalis, Pharmacol. Rev. 31, 20.

Hamilton, J.C., and S.D. Longman, 1982, A comparison of the cardiovascular and sedative action of the alpha adrenoceptor agonist FLA 136 and clonidine in the rat, Br. J. Pharmacol. 75, 13.

Isaac, L., 1980, Clonidine in the central nervous system: site and mechanism of hypotensive action, J. Cardiovasc. Pharmacol. 2, Suppl. 1, 515.

Kobinger, W., 1978, Central alpha adrenergic systems as target for hypotensive drugs, Rev. Physiol. Biochem. Pharmacol. 81, 39.

Kobinger, W. and L. Pichler, 1977, Pharmacological characterisation of B-HT 933 (2-amino-6-ethyl-4,5,7,8-tetrahydro-6*H*-oxazolo-(5,4-*d*)-azepine dihydrochoride) as hypotensive agent of the clonidine type, Naunyn-Schmied. Arch. Pharmacol. 300, 39.

Kobinger, W. and L. Pichler, 1980a, Investigation into different types of post and presynaptic alpha adrenoceptors at cardiovascular sites in rats, Eur. J. Pharmacol. 65, 393.

Kobinger, W. and L. Pichler, 1980b, Relation between central sympathoinhibitory and peripheral pre- and post-synaptic adrenoceptors on evaluated by different clonidine like substances in rats, Naunyn-Schmied. Arch. Pharmacol. 315, 21.

Lechat, P. and H. Schmitt, 1982, Interactions between the autonomic nervous system and the cardiovascular effects of ouabain in guinea-pigs, Eur. J. Pharmacol. 78, 212.

Papp, J.G. and E.M. Vaughan Williams, 1969, The effect of bretylium on intracellular cardiac action potentials to its antiarrhythmic and local anaesthetic activity, Br. J. Pharmacol. 37, 380.

Pichler, L., P. Plachetta and W. Kobinger, 1980, Effects of azepexole (B-HT-933) on pre and post synaptic alpha adrenoceptors at peripheral and central nervous sites, Eur. J. Pharmacol. 65, 233.

Plunkett, L.M. and R.L. Tackett, 1983, Central alpha receptors and their role in digoxin cardiotoxicity, J. Pharmacol. Exp. Ther. 227, 683.

Raines, A., D. Moros and B. Levitt, 1968, The effect of guanethidine on ouabain induced ventricular arrhythmia in the cat, Arch. int. Pharmacodyn. Ther. 174, 373.

Rhodes, K.F., 1986, The effects of some alpha adrenoceptor antagonists at alpha$_1$ and alpha$_2$ sites in isolated smooth muscle preparations, Ph.D. Thesis, University of London.

Saito, H., J. Otai, I. Shudo and T. Tanabe, 1974, Effect of 6-hydroxy dopamine on cardiotoxicity of ouabain in guinea-pigs, Jpn. J. Pharmacol. 24, 923.

Saxena, P.R. and K.P. Bhargava, 1975, The importance of central adrenergic mechanism in the cardiovascular responses to ouabain, Eur. J. Pharmacol. 31, 332.

Sekiya, A. and E.M. Vaughan Williams, 1963, The effects of pronethalol, dichloroisoprenaline and disopyramide on the toxicity to the heart of ouabain and anaesthetics, Br. J. Pharmacol. 21, 462.

Sivam, S.P., S.D. Seth, S. Nayar and S.C. Manchanda, 1980, Neurally induced digitalis arrhythmias and adrenoceptors, Eur. J. Pharmacol. 68, 107.

Smith, T.W., E.M. Antman, P.L. Friedman, C.M. Blatt and J.D. Marsh, 1984, Digitalis glycosides: mechanisms and manifestations of toxicity, Prog. Cardiovasc. Dis. 26, 495.

Thomas, G.P. and P.M. Stephen, 1993, Effects of two imidazolines (St-91 and St-93) on the cardiac arrhythmias and lethality induced by ouabain in guinea-pig, Asia Pac. J. Pharmacol. 8, 109.

Thomas, G.P. and R.M. Tripathi, 1986, Effects of alpha adrenoceptor agonists and antagonists on ouabain induced arrhythmias and cardiac arrest in guinea-pig, Br. J. Pharmacol. 89, 385.

Thomas, G.P. and R.K. Varma, 1991, Isolated paced guinea-pig left atrium: A new ouabain-induced arrhythmia model, Meth. Find. Exp. Clin. Pharmacol. 13, 459.

Thomas, G.P. and R.K. Varma, 1993, Effect of flutonidine on ouabain-induced arrhythmias and lethality in guinea-pig, Pharmacol. Res. 27, 365.

Timmermans, P.B.M.W.M. and P.A. Van Zwieten, 1980, Postsynaptic alpha$_1$ and alpha$_2$ adrenoceptors in the circulatory system of the pithed rat. Selective stimulation of the alpha$_2$ subtype by B-HT 933, Eur. J. Pharmacol. 63, 199.

Tripathi, R.M., G.P. Thomas and D.R. Shridhar, 1984, Influence of vagal tone on adrenaline-induced ventricular tachycardia in rats and guinea pigs, Indian J. Physiol. Pharmacol. 28, 237.

Trzeciakowski, J.P., 1985, Protective action of cimetidine against ouabain induced pressor effects, arrhythmias and lethality in guinea-pigs, J. Cardiovasc. Pharmacol. 7, 622.

Van Zwieten, P.A. and P.B.M.W.M. Timmermans, 1980, Centrally mediated hypotensive activity of B-HT 933 upon infusion via the cat's vertebral artery, Pharmacology 21, 327.

Van Zwieten, P.A. and P.B.M.W.M. Timmermans, 1983, Cardiovascular alpha$_2$ receptors, J. Mol. Cell. Cardiol. 15, 717.

Veniant, M., J.-P. Clozel, P. Hess, and M. Clozel, 1994, Endothelin plays a role in the maintenance of blood pressure in normotensive guinea-pigs, Life Sci. 55, 445.

Weitzell, R., T. Tanaka and K. Starke, 1979, Pre and post synaptic effects of yohimbine stereoisomers on noradrenergic transmission in the pulmonary artery of the rabbit, Naunyn. Schmied. Arch. Pharmacol. 308, 127.