Receptor sites of action of clonidine: effects of clonidine and three structural isomers on prejunctional and postjunctional α-adrenoceptors and histamine H_2-receptors in guinea-pig isolated cardiovascular tissues

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The antihypertensive agent clonidine is an imidazolidine derivative: 2-(2,6-dichlorophenylimino)imidazolidine. Its mode of action is thought to be due to activation of α-adrenoceptors in the central nervous system (Schmitt 1977; van Zwieten 1977). Sympathomimetic drugs, including clonidine are capable of preferentially activating either prejunctionally or postjunctionally located α-adrenoceptors in the periphery (Starke et al 1974, 1975; Drew 1976). Recent evidence from structure-activity studies suggests that in the rat, the central α-adrenoceptors are not different from peripheral vascular postjunctional α-adrenoceptors (Timmermans & van Zwieten 1977a). Recently, histamine H_2-receptors have also been implicated in the hypotensive effects of clonidine (Finch et al 1978). The present study examines the effects of clonidine and three structural isomers on pre- and postjunctional α-adrenoceptors and histamine H_2-receptors in guinea-pig isolated cardiovascular tissues.

To investigate effects on guinea-pig vascular postjunctional α-adrenoceptors, cumulative concentration-response curves were constructed to the contractile effects of noradrenaline and the imidazolidines in spirally-cut aortic strips. The method is similar to that for rabbit aortic strips reported previously (Medgett et al 1978). Effects on histamine H_2-receptors mediating positive chronotropic activity were assessed in guinea-pig isolated atria; cumulative concentration-response curves were constructed to histamine and the imidazolidines. Effects on prejunctional α-adrenoceptors were also assessed in guinea-pig isolated atria in transmitter release studies using atria labelled with [^{3}H]noradrenaline (Medgett et al 1978). The four imidazolidine derivatives used in the study were generously supplied by Boehringer Ingelheim: these were the hydrochloride salts of clonidine, St 155 [2-(2,6-dichlorophenylimino)-imidazolidine), St 363 (2,4-dichloro analogue), St 475 (2,5-dichloro) and St 476 (2,3-dichloro).

pD_2 and intrinsic activities for the imidazolidines at postjunctional α-adrenoceptors (guinea-pig aorta), prejunctional α-adrenoceptors (guinea-pig atria) and histamine H_2-receptors (guinea-pig atria) are shown in Table 1. Included in the Table are the pC30 (− log ED 30 in μmol kg^{-1}) values for the hypotensive effects of the compounds in the anaesthetized rat taken from the data of Timmermans & van Zwieten (1977b). In the aorta the concentration-response curves yield pD_2 values which are similar, ranging over only half a log unit; intrinsic activity values are also similar. In atria, the range of pD_2 values for the drugs at prejunctional α-adrenoceptors is much greater (3·3 log units) and the relative intrinsic activity values also differ widely. To calculate a pD_2 value for a drug at prejunctional α-adrenoceptors, a range of drug concentrations was used; each experiment yields the value for inhibition of stimulation-induced efflux at the drug concentration. Values from individual experiments are pooled in order to construct a 'concentration-response curve' and hence calculate a pD_2 value. In all cases except that of St 475 there was a concentration-dependent inhibition of efflux using stimulation of 5 pulses (1 Hz for 5 s). In the case of St 475, maximum inhibition of efflux is obtained at a concentration of 0·1 μM; at higher concentrations, inhibition decreases. St 475 markedly enhances (up to three-fold) resting efflux, simultaneously decreasing the rate and force of beating of the atria. It thus seems likely that the cardiotoxic actions obscure the inhibitory effect of St 475 on stimulation-induced efflux in concentrations above 0·1 μM; hence no pD_2 was calculated for this drug at prejunctional α-adrenoceptors.

Clonidine and histamine produced positive chronotropic effects in atria which were competitively antagonized by the prior administration of the histamine H_2-receptor antagonist cimetidine. In contrast, the other imidazolidines produced only negative chronotropic effects in concentrations above 10 μM.

In conclusion, the data obtained in the present study indicate that, of the four imidazolidine derivatives, only clonidine activates histamine H_2-receptors. The effects of the other derivatives in causing decreases in the rate of beating of the atria in high concentrations is probably a local anaesthetic effect, as has been demonstrated for clonidine itself (Schmitt 1977; van Zwieten 1977) and such an action for clonidine may contribute to its apparently low intrinsic activity at histamine H_2-receptors. In contrast to their effects on histamine H_2-receptors, all four derivatives induced hypotension in the anaesthetized rat (Timmermans & van Zwieten 1977b). If histamine H_2-receptors are similar in different species, a postulate which is supported in the literature by similar pA_2 values obtained for histamine H_2-receptor antagonists in different species and tissues (Black et al 1972), then the present results argue against the involvement of histamine H_2-receptors in the hypotensive effects of clonidine.

In the study of Timmermans & van Zwieten (1977b), the rank order of effectiveness for the hypotensive

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Table 1. pD_2 and intrinsic activity (I.A.) values for the four imidazolidine derivatives on postjunctional α-adrenoceptors (guinea-pig atria), prejunctional α-adrenoceptors (guinea-pig atria) and histamine H_2 receptors (guinea-pig atria). Included for comparison are the pC30 (—log ED30 μmolkg^{-1}) values of the compounds for their hypotensive activities in the anaesthetized normotensive rat taken from the data of Timmermans & van Zwieten (1977b). N.C. indicates not calculated; N.A. indicates not activated. Values are given as mean ± standard error of the mean; n is the number of experiments. In guinea-pig aorta, noradrenaline (pD_2 = 6·74, s.e.m. = 0·04, n = 66) was taken as the full agonist (I.A. = 100); in guinea-pig atria, for effects on prejunctional α-adrenoceptors "I.A." = 100 was taken arbitrarily as complete (100%) inhibition of stimulation-induced efflux. n is relatively large in these experiments since in a single experiment the effect of only one drug concentration could be assessed. In guinea-pig atria, for effects on histamine H_2-receptors, histamine (pD_2 = 6·21, s.e.m. = 0·16, n = 6) was taken as the full agonist (I.A. = 100).

	Postjunctional α-adrenoceptors			Prejunctional α-adrenoceptors			Histamine H_2-receptors			Hypotensive activity
	pD_2	I.A.	n	pD_2	"I.A."	n	pD_2	I.A.	n	pC30
Clonidine (2,6-di-Cl)	6·03 ± 0·10	61·3 ± 18·8	6	8·81 ± 0·19	87 ± 10	19	5·31 ± 0·11	30 ± 4	3	1·99
St 476 (2,3-di-Cl)	5·41 ± 0·21	58·5 ± 8·4	4	6·08 ± 0·27	70 ± 13	14		N.A.	3	1·31
St 363 (2,4-di-Cl)	5·61 ± 0·18	70·8 ± 3·0	3	5·53 ± 0·27	43 ± 5	12		N.A.	3	0·64
St 475 (2,5-di-Cl)	6·18 ± 0·23	74·0 ● 9·7	4	N.C.	N.C.	12		N.A.	3	0·25

effects in the rat was: clonidine > St 476 > St 363 > St 475; these values range over about 2 log units. For guinea-pig aortic postjunctional α-adrenoceptors, the rank order of pD_2 values is different: the values are all very similar, ranging over only half a log unit. On the other hand, for guinea-pig atrial prejunctional α-adrenoceptors, with the exception of St 475, for which a pD_2 value could not be calculated, the pD_2 values follow the same order of potency: the range of values is 3·3 log units. It should also be pointed out that all four imidazolidines possess nearly identical lipid solubility and pKa values (data provided by Boehringer Ingelheim), and their molecular volumes are equal, since they are isomers; thus it is unlikely that purely physicochemical differences give rise to the different orders of potency in the different receptor systems.

The results then suggest that the central receptor activated by clonidine resembles more closely peripheral prejunctional, rather than peripheral postjunctional α-adrenoceptors.

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