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Involvement of peripheral presynaptic inhibition in the reduction of sympathetic tone by moxonidine, rilmenidine and UK 14304

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Abstract

We studied the possibility that presynaptic inhibition of transmitter release from postganglionic sympathetic neurons contributes to the overall reduction of sympathetic tone produced by moxonidine, rilmenidine and 5-bromo-6-(2-imidazolin-2-ylamino)-quinoxaline tartrate (UK 14304). In pithed rabbits without electric stimulation, moxonidine, rilmenidine and UK 14304 caused a long-lasting, > 10 min, increase in arterial pressure. Heart rate was not changed. In pithed rabbits in which sympathetic tone was created by electric stimulation through the pithing rod (2 Hz), the same doses of moxonidine, rilmenidine and UK 14304 caused only a brief, < 10 min, blood pressure rise. Heart rate was decreased, as were the plasma concentrations of noradrenaline and adrenaline. Dose-response curves for the effects on the plasma noradrenaline concentration (stimulated pithed rabbits) were compared with previously obtained dose-response curves for depression of renal sympathetic nerve activity (conscious rabbits). For each drug, the curve describing peripheral presynaptic inhibition and the curve describing central sympathoinhibition were very similar. Both the power and the dose dependence of the peripheral inhibitory effect support its contribution to the overall decrease in sympathetic tone produced by clonidine-like drugs in intact animals. The peripheral effect in all likelihood consists in activation of presynaptic α_2-autoreceptors. The agreement of the dose-response curves for the peripheral and for the central effect supports the view that the central effect, like the peripheral one, is mediated through α_2-adrenoceptors.

Keywords: α_2-Adrenoceptor; Imidazoline receptor; Presynaptic inhibition; Moxonidine; Rilmenidine; UK 14304

1. Introduction

Clonidine and clonidine-like drugs such as moxonidine and rilmenidine are lipophilic α_2-adrenoceptor agonists that lower blood pressure. The blood pressure decrease is due to a decrease of sympathetic tone and an increase in vagal tone (see Schmitt, 1977; Kobinger, 1986; Kobinger and Pichler, 1990). Sympathetic tone of cardiovascular effector tissues may be defined as the degree of postsynaptic adrenoceptor activation. Clonidine-like drugs can change this tone by at least three mechanisms: reduction of sympathetic outflow from the central nervous system (CNS), i.e. of action potential frequency in preganglionic sympathetic fibres; reduction of the average release of noradrenaline and its cotransmitters per action potential by means of activation of presynaptic α_2-autoreceptors; and direct activation of postsynaptic α_2-and α_1-adrenoceptors. The first two mechanisms tend to reduce cardiac output, total peripheral resistance and, hence, blood pressure; the third mechanism tends to increase total peripheral resistance and, hence, blood pressure.

In most discussions on clonidine-like drugs, only the reduction of central sympathetic outflow is held responsible for the fall in blood pressure, and the drugs therefore are classified as centrally acting antihypertensives (Schmitt, 1977; Kobinger and Pichler, 1990; moxonidine and rilmenidine: Armah et al., 1988; Verbeuren et al., 1990; Chrisp and Faulds, 1992; Ernsberger et al., 1993). The nature of their central receptors is debated. The original idea that they act as agonists on central α_2-adrenoceptors (Schmitt, 1977; Kobinger and Pichler, 1990; moxonidine: Armah et al., 1988; rilmenidine: Van Zwieten, 1988) has recently been strengthened (Hieble and Kolpak, 1993; Allen

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and Guyenet, 1993; Szabo et al., 1993; Guyenet et al., 1994; Urban et al., 1994, 1995). Alternatively, they may lower central sympathetic outflow through the so-called imidazoline receptors, specifically I_1 receptors (Bousquet et al., 1984; Verbeuren et al., 1990; Chrisp and Faulds, 1992; Ernsberger et al., 1993). Clonidine, moxonidine and rilmenidine possess a higher affinity for I_1 than for α_2 binding sites (Ernsberger et al., 1993; but for moxonidine see Bricca et al., 1994).

Despite the universal operation of presynaptic α_2-autoreceptors in cardiovascular tissues (see Starke, 1977, 1987; Westfall, 1977; Langer, 1981; Starke et al., 1989), the possibility that they contribute to the decrease in sympathetic tone caused by clonidine-like drugs has received little attention and even less support (Haeusler, 1976; Pichler and Kobinger, 1978; Garty et al., 1990; Kobinger and Pichler, 1990). One reason may be that the direct experimental test, selective peripheral α_2-autoreceptor activation or blockade, is not practicable: even if an α_2-adrenoceptor ligand does not cross the blood-brain barrier it will reach both the (tone-decreasing) autoreceptors and the (tone-increasing) smooth muscle α-adrenoceptors. Disregard of this inevitable duality may lead to error. From the finding that only those α_2-adrenoceptor agonists that penetrate into the brain lower blood pressure it has been concluded that peripheral presynaptic inhibition is negligible (Pichler and Kobinger, 1978; Kobinger and Pichler, 1990). However, the direct postsynaptic vasoconstrictor action may have balanced the presynaptic inhibition in these experiments. A second reason for neglect of the peripheral inhibition may be the success of direct tests for CNS involvement. For example, selective CNS application of α_2-adrenoceptor antagonists prevents the hypotension caused by clonidine-like drugs, and selective CNS application of α_2-adrenoceptor agonists produces hypotension (Kobinger, 1967; Armah et al., 1988; Feldman et al., 1990; Sannajust and Head, 1994; Urban et al., 1995; and see Schmitt, 1977; Kobinger and Pichler, 1990). However, the latter finding does not imply that the CNS action alone produces hypotension when clonidine-like drugs are given *systemically*: systemic application brings them into contact with the (tone-increasing) vascular postsynaptic α-receptors, and in this – therapeutically relevant – situation the (tone-decreasing) peripheral presynaptic action may be a second necessary condition for the hypotension. A specific reason for failures to detect presynaptic inhibition by clonidine-like drugs may be high rates of sympathetic impulse traffic: the higher the rate, the narrower is the scope for this inhibition (Starke, 1977, 1987). For example, clonidine did not presynaptically inhibit the renal release of noradrenaline in anaesthetized rats (Garty et al., 1990). However, the arterial plasma noradrenaline level in that study, > 1 ng ml^{-1}, indicates a high sympathetic firing rate.

We investigated the possibility of a peripheral presynaptic contribution to the depression of sympathetic

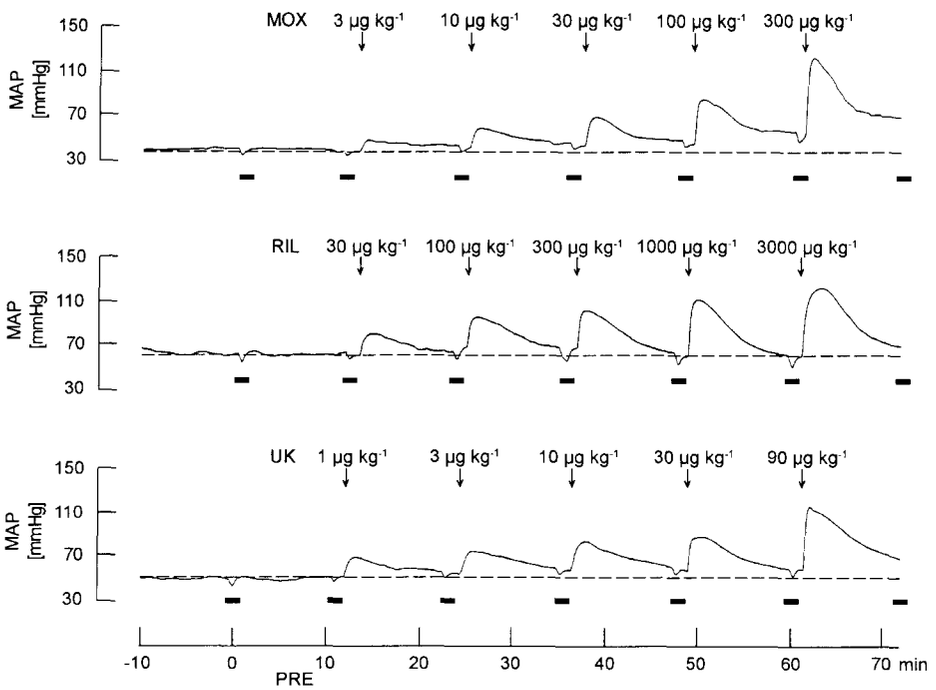

Fig. 1. Mean arterial pressure in single experiments with moxonidine (MOX), rilmenidine (RIL) and UK 14304 (UK) in pithed rabbits without electric stimulation. Blood withdrawal at the 7 measurement points, first point at $t = 0$ min and subsequent points at intervals of about 12 min (bars below curves), caused mean arterial pressure falls. The 0-and 12-min measurement points were averaged in each experiment to give the PRE values (dashed horizontal lines). The drugs were injected i.v. after the 2nd to 6th measurement point as indicated.

tone produced by moxonidine and rilmenidine in rabbits. The direct test mentioned not being practicable, postsynaptic vasoconstrictor effects were tested in pithed rabbits. The presynaptic effect was examined in pithed rabbits in which an artificial sympathetic tone was created by electric stimulation through the pithing rod. The dose-response curves thus obtained were compared with dose-response curves, taken from our previous studies (Urban et al., 1994, 1995), for inhibition of the firing rate of renal sympathetic nerves in conscious rabbits. The effect of 5-bromo-6-(2-imidazolin-2-ylamino)-quinoxaline tartrate (UK 14304), an α_2-adrenoceptor agonist with very low affinity for imidazoline I_1 receptors (Ernsberger et al., 1992), was studied for comparison.

2. Materials and methods

Thirty-seven rabbits of mixed breed and either sex were used (1.8–2.5 kg). The preparation has been described previously (Szabo et al., 1987; see also Majewski et al., 1983a). Briefly, the rabbits were anaesthetized with sodium pentobarbitone 75 mg kg^{-1} i.v. and artificial respiration with air enriched with O_2 was applied. Both carotid arteries and jugular veins were cannulated. One artery served for blood pressure measurement, the other for blood sampling. Heart rate was integrated from the pressure pulse. Veins served for drug administration. After muscle relaxation by gallamine triethiodide 5 mg kg^{-1} i.v., a non-insulated steel rod was advanced through a hole in the parietal bone approximately 25 cm down the spinal canal. The rod destroyed the central nervous system.

Two kinds of experiments were carried out: on pithed rabbits without electric stimulation ('non-stimulated'), and on pithed rabbits in which the preganglionic sympathetic nerves were continuously stimulated through the pithing rod by square wave pulses of 0.5 ms width, 140 mA current strength and a frequency of 2 Hz ('stimulated').

There were 7 (non-stimulated) or 5 (stimulated) measurement points, the first one about 90 min after surgery ($t = 0$ min) and the following ones at intervals of about 12 min. Carotid blood, 2 ml, was withdrawn at each measurement point; re-suspended erythrocytes from the preceding withdrawal were injected as substitution; the sampling procedure took about 2 min. Although blood should be almost catecholamine-free in non-stimulated rabbits, samples were also taken from these animals to make experiments comparable. Blood pressure and heart rate were read immediately before blood sampling in order to avoid artifacts (see Fig. 1 and Fig. 3). Saline or increasing doses of moxonidine, rilmenidine or UK 14304 were injected i.v. briefly after the erythrocyte re-injections of the 2nd to the penultimate measurement points, so that the 3rd to the last measurement points represent values about 10 min after the preceding injection.

Noradrenaline and adrenaline were determined in the 2-ml blood samples (only 0- and 12-min samples in non-stimulated rabbits). Catecholamines were measured by alumina chromatography followed by high pressure liquid chromatography with electrochemical detection (Szabo and Schultheiss, 1990); dihydroxybenzylamine 5 ng was added to each sample as internal standard. When the concentration was below the detection limit (twice baseline noise; 9.5 ± 3.2 pg ml^{-1} plasma for noradrenaline and 10.6 ± 3.5 pg ml^{-1} plasma for adrenaline; $n = 21$), the detection limit was taken as the catecholamine value (stimulated rabbits: 1 from 105 noradrenaline measurements and 3 from 105 adrenaline measurements).

In each experiment, the blood pressure, heart rate and catecholamine values of the 1st and 2nd measurement points, $t = 0$ and 12 min, before saline or drugs, were averaged to yield the PRE value; all values are expressed as percentages of PRE. Means ± S.E.M. are given throughout. Groups were compared by means of the Mann-Whitney test. $P < 0.05$ was taken as the limit of significance, and only this level is indicated even where $P < 0.01$. Logistic curves were fitted to dose-response data (means) using equation No. 25 of Waud (1976) (Fig. 5).

Drugs were moxonidine (Beiersdorf, Hamburg, Germany), rilmenidine dihydrogenphosphate (Servier,

Table 1
Mean arterial pressure, heart rate and plasma catecholamine concentration

	Non-stimulated pithed rabbits ($n = 16$)	Stimulated pithed rabbits ($n = 21$)
Mean arterial pressure (mm Hg)	48.8 ± 2.0	68.1 ± 3.5 [a]
Heart rate (min^{-1})	267 ± 9	250 ± 7
Plasma noradrenaline concentration (pg ml^{-1})	Not detectable in 25 of 32 samples	435 ± 32
Plasma adrenaline concentration (pg ml^{-1})	Not detectable in 29 of 32 samples	159 ± 18

In one group there was no electric stimulation, in the other the preganglionic sympathetic nerves were stimulated electrically (2 Hz). Values are the PRE values, i.e. averages of the values determined at $t = 0$ and 12 min. [a] Significant difference from non-stimulated rabbits ($P < 0.05$).

Courbevoie, France) and UK 14304 tartrate (Pfizer, Sandwich, UK). They were dissolved in saline. Doses refer to the salts. Intravenous injection volumes were 1 ml kg^{-1}.

3. Results

3.1. Pithed rabbits without electric stimulation

Averages of the blood pressure and heart rate values at the first two measurement points, $t = 0$ and 12 min, before administration of saline or drugs, are presented in Table 1. As expected, noradrenaline and adrenaline were rarely detectable in blood from these animals.

In rabbits which received 5 injections of saline, mean arterial pressure and heart rate remained approximately constant or decreased very slightly (see triangles in Fig. 2). Moxonidine 3–300 $\mu g\ kg^{-1}$, rilmenidine 30–3000 $\mu g\ kg^{-1}$ and UK 14304 1–90 $\mu g\ kg^{-1}$ all caused an immediate blood pressure rise. Representative experiments are shown in Fig. 1. Mean arterial pressure then declined from the peak but was

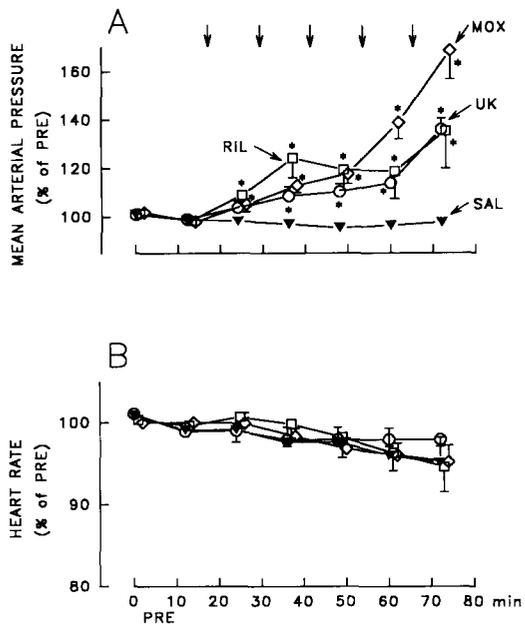

Fig. 2. Effect of saline (SAL), moxonidine (MOX), rilmenidine (RIL) and UK 14304 (UK) on A, mean arterial pressure, and B, heart rate in pithed rabbits without electrical stimulation. Protocol is illustrated in Fig. 1. There were 7 measurement points. After the 2nd to 6th measurement points, the rabbits received i.v. injections (arrows) of either saline (triangles) or increasing doses of MOX (3, 10, 30, 100 and 300 $\mu g\ kg^{-1}$; diamonds) or increasing doses of RIL (30, 100, 300, 1000 and 3000 $\mu g\ kg^{-1}$; squares) or increasing doses of UK (1, 3, 10, 30 and 90 $\mu g\ kg^{-1}$; circles). Post-injection measurements represent values at about 10 min after the preceding injection. All values are expressed as percentages of PRE (ordinates). Means ± S.E.M., $n = 4$–6 each. * Significant difference from SAL ($P < 0.05$).

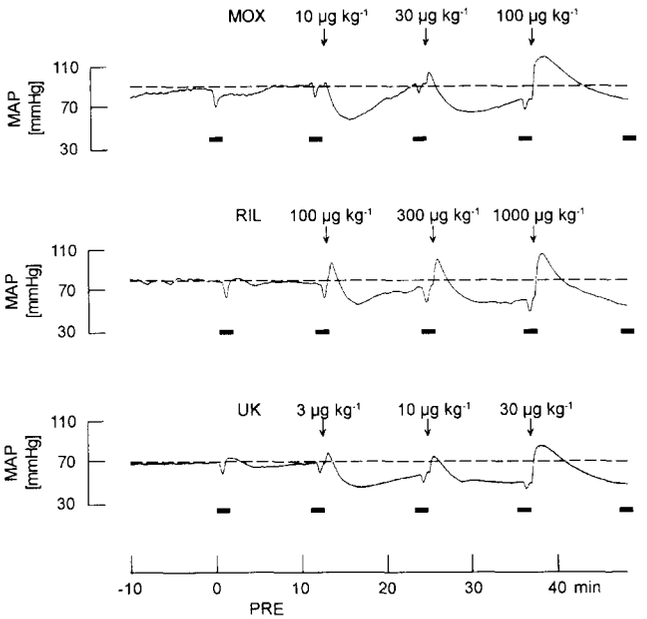

Fig. 3. Mean arterial pressure in single experiments with moxonidine (MOX), rilmenidine (RIL) and UK 14304 (UK) in pithed rabbits with electrically stimulated sympathetic outflow (2 Hz). Blood withdrawal at the 5 measurement points, first point at $t = 0$ min and subsequent points at intervals of about 12 min (bars below curves), caused mean arterial pressure falls. The 0-and 12-min measurement points were averaged in each experiment to give the PRE values (dashed horizontal lines). The drugs were injected i.v. after the 2nd to 4th measurement point as indicated.

still higher than after saline 10 min later (Fig. 1 and Fig. 2A). Heart rate was not changed (Fig. 2B) except for occasional small transient decreases following injections of high doses of the drugs (not shown).

3.2. Pithed rabbits with electrically stimulated sympathetic outflow

The 0-and 12-min values (Table 1) indicate that mean arterial pressure was higher than without electric stimulation but the heart rate was not higher on average, although in individual experiments the stimulation caused cardioacceleration. Noradrenaline and adrenaline were always detectable. The values resemble those measured previously under similar conditions (Szabo et al., 1987).

In rabbits receiving 3 injections of saline, blood pressure fell during the experiments, by 11% at the last measurement point (see triangles in Fig. 4A). Heart rate and the plasma noradrenaline and adrenaline concentration remained approximately constant or decreased very slightly (see Fig. 4B–D). Moxonidine 10–100 $\mu g\ kg^{-1}$, rilmenidine 100–1000 $\mu g\ kg^{-1}$ and UK 14304 3–30 $\mu g\ kg^{-1}$ again caused a sharp initial blood pressure rise. Representative experiments are shown in Fig. 3. The pressure peaks tended to be lower than in

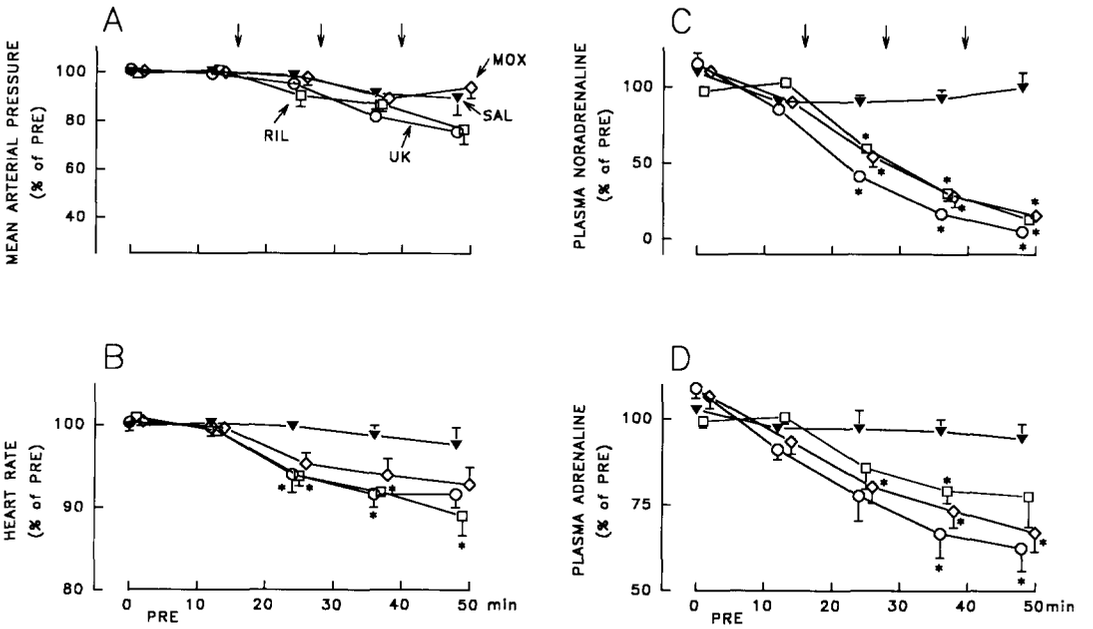

Fig. 4. Effect of saline (SAL), moxonidine (MOX), rilmenidine (RIL) and UK 14304 (UK) on A, mean arterial pressure, B, heart rate, C, plasma noradrenaline concentration and D, plasma adrenaline concentration in pithed rabbits with electrically stimulated sympathetic outflow (2 Hz). Protocol is illustrated in Fig. 3. There were 5 measurement points. After the 2nd to 4th measurement points, the rabbits received i.v. injections (arrows) of either saline (triangles) or increasing doses of MOX (10, 30 and 100 μg kg^{-1}; diamonds) or increasing doses of RIL (100, 300 and 1000 μg kg^{-1}; squares) or increasing doses of UK (3, 10 and 30 μg kg^{-1}; circles). Post-injection measurements represent values at about 10 min after the preceding injection. All values are expressed as percentages of PRE (ordinates). Means ± S.E.M., $n = 4$–6 each. * Significant difference from SAL ($P < 0.05$).

non-stimulated pithed rabbits (compare Fig. 1 and Fig. 3). After the peaks, the blood pressure fell much more steeply than in animals without electric stimulation (compare Fig. 1 and Fig. 3) and reached a level no higher than, or even (non-significantly) below, that with saline after 10 min (Fig. 3 and Fig. 4A). Again, in contrast to the effect in non-stimulated rabbits, moxonidine, rilmenidine and UK 14304 significantly lowered heart rate, by up to about 10% maximally (Fig. 4B). The plasma noradrenaline concentration was reduced to almost zero at the highest doses (Fig. 4C). The plasma adrenaline concentration was also reduced, by 33, 23 and 38% at the highest doses of moxonidine, rilmenidine and UK 14304, respectively (Fig. 4D).

4. Discussion

Moxonidine (Armah, 1988), rilmenidine (Laubie et al., 1985; Van Zwieten et al., 1986) and UK 14304 (Van Meel et al., 1981) have previously been shown to increase arterial pressure in pithed rats. Our experiments demonstrated the same in pithed rabbits. The increase, typical for clonidine-like drugs, results from activation of vascular postsynaptic α-adrenoceptors.

Moxonidine (Armah, 1988; Schlicker et al., 1990; Göthert and Molderings, 1991; Bohmann et al., 1994), rilmenidine (Verbeuren et al., 1986) and UK 14304 (Levitt and Hieble, 1986) decrease the overflow of noradrenaline evoked from isolated tissues. They also reduce effector cell responses to sympathetic nerve stimulation in whole animal preparations (Van Meel et al., 1981; Laubie et al., 1985; Van Zwieten et al., 1986; Armah, 1988). We applied the chemical approach to pithed rabbits with electrically stimulated sympathetic outflow: all three drugs reduced the plasma noradrenaline level. The mode of action, although not examined here, surely is presynaptic α_2-adrenergic inhibition: clonidine also reduces the release of noradrenaline in stimulated pithed rabbits, as does the α_2-selective agonist (Starke et al., 1975b) α-methyl-noradrenaline, and the effect of both drugs is blocked by the α_2-selective antagonist (Starke et al., 1975a) yohimbine (Majewski et al., 1983a; Ensinger et al., 1985). Inhibition of sympathetic ganglia seems unlikely because clonidine, the prototype, lacks ganglionic action (Kobinger, 1967). UK 14304 has been shown to inhibit noradrenaline release in rabbit isolated tissues through the α_{2A} subtype to which presynaptic α_2-autoreceptors belong in this species (Limberger et al., 1991,1995; Trendelenburg et al., 1993; Starke et al., 1995). For moxonidine and rilmenidine, an action at presynaptic imidazoline receptors, which have been demonstrated in rabbit tissues (Göthert and Molderings, 1991; Molderings et al., 1991), might be an alternative; however, moxoni-

dine does not act on the presynaptic imidazoline receptors (Molderings et al., 1991).

The decrease of the plasma adrenaline level in the stimulated rabbits is another feature that moxonidine, rilmenidine and UK 14304 share with clonidine (Majewski et al., 1983a). The mechanism is not clear (see Majewski et al., 1983a).

Does peripheral presynaptic inhibition, shown here directly as a fall in plasma noradrenaline, contribute to the overall inhibition by moxonidine, rilmenidine and UK 14304 of sympathetic tone in animals with an intact CNS? Two observations suggest an affirmative answer: the depression of cardiovascular neuroeffector transmission resulting from the presynaptic effect, and the occurrence of the presynaptic effect at appropriate doses.

In non-stimulated rabbits, in which the postganglionic sympathetic neurons were silent (note absence of noradrenaline from blood), moxonidine, rilmenidine and UK 14304 increased blood pressure and did not change the heart rate. In the stimulated preparations, in which presynaptic inhibition became possible, their effect was greatly changed. They decreased the heart rate, demonstrating inhibition of *cardiac* sympathetic neuroeffector transmission. The decrease was small, perhaps because few cardioaccelerator fibres were excited: PRE heart rate was not higher in stimulated than non-stimulated animals (Table 1). The decrease supports the idea that peripheral presynaptic inhibition contributes to the fall in *cardiac* sympathetic tone produced by clonidine-like drugs in intact animals. The initial blood pressure peak became smaller and briefer in the stimulated rabbits, and 10 min after injection the blood pressure had returned to its control levels, whereas it was still above the control in the non-stimulated preparations. The change of the blood pressure response demonstrates inhibition of *vascular* neuroeffector transmission and supports the idea that peripheral presynaptic inhibition contributes to the fall in *vascular* sympathetic tone produced by clonidine-like drugs in intact animals (compare the analysis in rabbit isolated pulmonary artery: Starke et al., 1974).

Dose-response curves of moxonidine, rilmenidine and UK 14304 for their peripheral presynaptic effect (decrease of plasma noradrenaline in stimulated pithed rabbits) are compared with dose-response curves for their central effect (decrease of renal sympathetic nerve activity in conscious rabbits) in Fig. 5. It is obvious – and this is a new finding for clonidine-like drugs as far as we are aware – that each of the three drugs exerts its peripheral and its central effect over the same dose range. Both central and peripheral sympathoinhibition develop after administration of sympathoinhibitory doses of clonidine-like drugs. Similarly, dose-response curves for the peripheral presynaptic effect of moxonidine, rilmenidine and UK 14304 (in stimulated pithed rabbits) can also be compared with curves for their hypotensive effect (in conscious rabbits; from Urban et al., 1994, 1995). The result is the same: each of the three compounds produces peripheral presynaptic inhibition and hypotension over the same dose range (not shown).

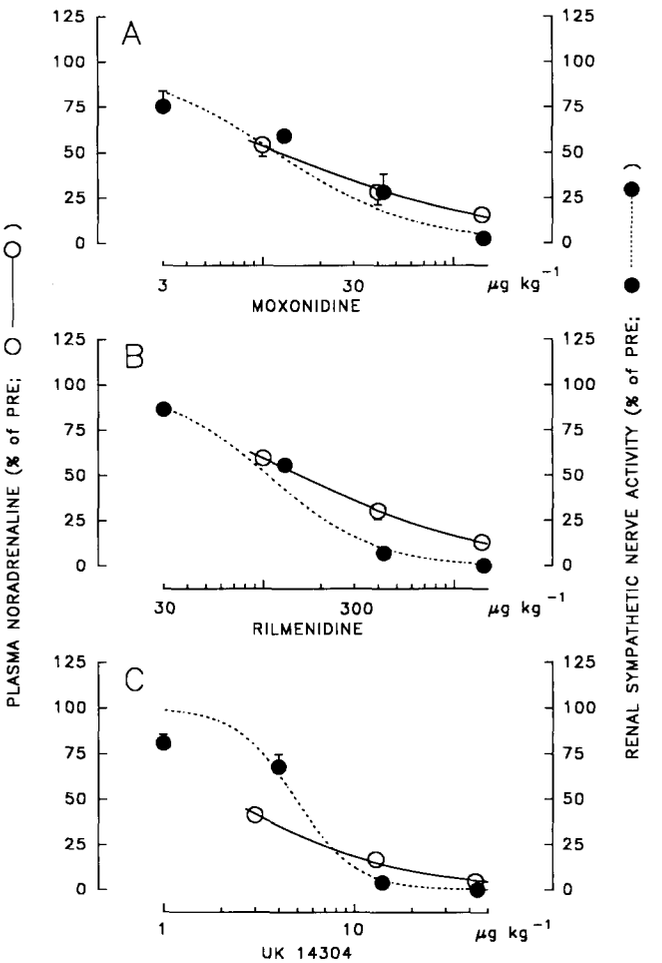

Fig. 5. Comparison of effects of A, moxonidine, B, rilmenidine and C, UK 14304 on the plasma noradrenaline concentration in pithed rabbits with electrically stimulated sympathetic outflow (left-hand ordinates) and renal sympathetic nerve activity in conscious rabbits (right-hand ordinates). Pithed rabbit data are from Fig. 4C. Conscious rabbit data are from Urban et al. (1994,1995). Drugs were injected i.v. in increasing doses at intervals of 12–13 min. Abscissae show cumulative doses. All values were obtained about 10 min after the respective dose and are expressed as percentages of initial values (PRE). Means ± S.E.M., $n = 4–7$ each. Logistic curves were fitted to mean values.

Some limitations of the study must be pointed out. It was necessary to stimulate the sympathetic outflow artifically to study peripheral presynaptic inhibition. We cannot rule out the possibility that transmitter release elicited by the natural irregular sympathetic firing is more resistant to presynaptic α_2-adrenoceptor inhibition, although this inhibition of course also operates during normal impulse flow (Majewski et al., 1983b;

Szabo et al., 1989; Grossman et al., 1991). Also, we compared peripheral activity to central inhibition of *renal* sympathetic firing (Fig. 5). Clonidine-like drugs may inhibit central sympathetic outflow to other organs more potently than that to the kidney (Ramage and Wilkinson, 1989). However, this does not seem to be the case in rabbits, at least for cardiac sympathetic fibres: higher, not lower doses of moxonidine, rilmenidine and UK 14304 are required for a decrease in heart rate than for renal sympathoinhibition (Urban et al., 1994, 1995). Another limitation is the duration of our observations: only 10 min after injection of a given dose. The impact of peripheral as compared to central inhibition may decline later. On the positive side, other studies are also compatible with, or have suggested, a peripheral contribution (De Jonge et al., 1981; Brown and Harland, 1984; Laubie et al., 1985; Van Zwieten et al., 1986; Warren et al., 1991; Sannajust et al., 1992; Urban et al., 1995). For example, in conscious rabbits, elimination of the central sympathoinhibition by yohimbine given into the cisterna magna blocked the hypotensive effect of systemically administered moxonidine and UK 14304 but blocked neither the bradycardia nor the fall in the plasma noradrenaline level (Urban et al., 1995).

The comparison of the dose-response curves for central and peripheral inhibition (Fig. 5) supplies an argument for the involvement of α_2-adrenoceptors in the central effect. Moxonidine and rilmenidine have higher affinity for I_1 than α_2 binding sites whereas the reverse holds true for UK 14304 (see Introduction). If I_1 receptors were responsible for the central inhibition and α_2-adrenoceptors for the peripheral inhibition, then rilmenidine and moxonidine should be more potent centrally than peripherally; the reverse should hold true for UK 14304. This was not found: the central and peripheral curves were closely similar for each drug, supporting the identity of the central with the peripheral receptor and, hence, α_2 identity of the former.

In conclusion, peripheral inhibition of sympathetic transmitter release by moxonidine, rilmenidine and UK 14304 *alone* is able to decrease heart rate and to entirely compensate for their postsynaptic vasoconstrictor effect. The dose-response curves for the peripheral presynaptic and the central sympathoinhibitory effect of these drugs can almost be superimposed. Both the power and the dose dependence of the peripheral effect support its contribution to the overall decrease in sympathetic tone. The designation 'centrally acting antihypertensives' for clonidine-like drugs refers to only part of their mechanism of action: the central action is necessary, but perhaps not sufficient, for the blood pressure fall. The results also support the view that, in the CNS, clonidine-like drugs act through α_2-adrenoceptors.

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References

Allen, A.M. and P.G. Guyenet, 1993, α_2-Adrenoceptor-mediated inhibition of bulbospinal barosensitive cells of rat rostral medulla, Am. J. Physiol. 265, R1065.

Armah, B.I., 1988, Unique presynaptic α_2-receptor selectivity and specificity of the antihypertensive agent moxonidine, Arzneim.-Forsch./Drug Res. 38, 1435.

Armah, B.I., E. Hofferber and W. Stenzel, 1988, General pharmacology of the novel centrally acting antihypertensive agent moxonidine, Arzneim.-Forsch./Drug Res. 38, 1426.

Bohmann, C., P. Schollmeyer and L.C. Rump, 1994, Effects of imidazolines on noradrenaline release in rat isolated kidney, Naunyn-Schmied. Arch. Pharmacol. 349, 118.

Bousquet, P., J. Feldman and J. Schwartz, 1984, Central cardiovascular effects of alpha adrenergic drugs: differences between catecholamines and imidazolines, J. Pharmacol. Exp. Ther. 230, 232.

Bricca, G., H. Greney, J. Zhang, M. Dontenwill, J. Stutzmann, A. Belcourt and P. Bousquet, 1994, Human brain imidazoline receptors: further characterization with [^{3}]clonidine, Eur. J. Pharmacol. 266, 25.

Brown, M.J. and D. Harland, 1984, Evidence for a peripheral component in the sympatholytic effect of clonidine in rats, Br. J. Pharmacol. 83, 657.

Chrisp, P. and D. Faulds, 1992, Moxonidine, Drugs 44, 993.

De Jonge, A., P.B.M.W.M. Timmermans and P.A. Van Zwieten, 1981, Participation of cardiac presynaptic α_2-adrenoceptors in the bradycardiac effects of clonidine and analogues, Naunyn-Schmied. Arch. Pharmacol. 317, 8.

Ensinger, H., H. Majewski, L. Hedler and K. Starke, 1985, Neuronal and postjunctional components in the blood pressure effects of dopamine and bromocriptine in rabbits, J. Pharmacol. Exp. Ther. 234, 681.

Ernsberger, P.R., K.L. Westbrooks, M.O. Christen and S.G. Schäfer, 1992, A second generation of centrally acting antihypertensive agents act on putative I_1-imidazoline receptors, J. Cardiovasc. Pharmacol. 20 (Suppl. 4), S1.

Ernsberger, P., H.L. Elliot, H.-J. Weimann, A. Raap, M.A. Haxhiu, E. Hofferber, A. Löw-Kröger, J.L. Reid and H.-J. Mest, 1993, Moxonidine: a second-generation central antihypertensive agent, Cardiovasc. Drug Rev. 11, 411.

Feldman, J., E. Tibirica, G. Bricca, M. Dontenwill, A. Belcourt and P. Bousquet, 1990, Evidence for the involvement of imidazoline receptors in the central hypotensive effect of rilmenidine in the rabbit, Br. J. Pharmacol. 100, 600.

Garty, M., A. Deka-Starosta, P. Chang, I.J. Kopin and D.S. Goldstein, 1990, Effects of clonidine on renal sympathetic nerve activity and norepinephrine spillover, J. Pharmacol. Exp. Ther. 254, 1068.

Göthert, M. and G.J. Molderings, 1991, Involvement of presynaptic imidazoline receptors in the α_2-adrenoceptor-independent inhibition of noradrenaline release by imidazoline derivatives, Naunyn-Schmied. Arch. Pharmacol. 343, 271.

Grossman, E., P.C. Chang, A. Hoffman, M. Tamrat and D.S. Goldstein, 1991, Evidence for functional α_2-adrenoceptors on vascular sympathetic nerve endings in the human forearm, Circ. Res. 69, 887.

Guyenet, P.G., R.L. Stornetta, T. Riley, F.R. Norton, D.L. Rosin and K.R. Lynch, 1994, Alpha$_{2A}$-adrenergic receptors are present in lower brainstem catecholaminergic and serotonergic neurons innervating spinal cord, Brain Res. 638, 285.

Haeusler, G., 1976, Studies on the possible contribution of a peripheral presynaptic action of clonidine and dopamine to their vascular effects under in vivo conditions, Naunyn-Schmied. Arch. Pharmacol. 295, 191.

Hieble, J.P. and D.C. Kolpak, 1993, Mediation of the hypotensive action of systemic clonidine in the rat by α_2-adrenoceptors, Br. J. Pharmacol. 110, 1635.

Kobinger, W., 1967, Über den Wirkmechanismus einer neuen antihypertensiven Substanz mit Imidazolinstruktur, Naunyn-Schmied. Arch. Pharmakol. Exp. Pathol. 258, 48.

Kobinger, W., 1986, Drugs as tools in research on adrenoceptors, Naunyn-Schmied. Arch. Pharmacol. 332, 113.

Kobinger, W. and L. Pichler, 1990, Centrally acting drugs (clonidine, methyldopa, guanfacine), in: Handbook of Experimental Pharmacology, Pharmacology of Antihypertensive Therapeutics, Vol. 93, eds. D. Ganten and P.J. Mulrow (Springer Verlag, Berlin) p. 227.

Langer, S.Z., 1981, Presynaptic regulation of the release of catecholamines, Pharmacol. Rev. 32, 337.

Laubie, M., J.-C. Poignant, J. Scuvée-Moreau, H. Dabire, A. Dresse and H. Schmitt, 1985, Pharmacological properties of (*N*-dicyclopropylmethyl) amino-2-oxazoline (S 3341), an α-2 adrenoceptor agonist, J. Pharmacol. (Paris) 16, 259.

Levitt, B. and J.P. Hieble, 1986, Prejunctional α_2-adrenoceptors modulate stimulation-evoked norepinephrine release in rabbit lateral saphenous vein, Eur. J. Pharmacol. 132, 197.

Limberger, N., L. Späth and K. Starke, 1991, Subclassification of the presynaptic α_2-autoreceptors in rabbit brain cortex, Br. J. Pharmacol. 103, 1251.

Limberger, N., L. Funk, A.U. Trendelenburg and K. Starke, 1995, Subclassification of presynaptic α_2-adrenoceptors: α_{2A}-autoreceptors in rabbit atria and kidney, Naunyn-Schmied. Arch. Pharmacol. 352, 31.

Majewski, H., L. Hedler and K. Starke, 1983a, Evidence for a physiological role of presynaptic α-adrenoceptors: modulation of noradrenaline release in the pithed rabbit, Naunyn-Schmied. Arch. Pharmacol. 324, 256.

Majewski, H., L. Hedler and K. Starke, 1983b, Modulation of noradrenaline release in the conscious rabbit through α-adrenoceptors, Eur. J. Pharmacol. 93, 255.

Molderings, G.J., F. Hentrich and M. Göthert, 1991, Pharmacological characterization of the imidazoline receptor which mediates inhibition of noradrenaline release in the rabbit pulmonary artery, Naunyn-Schmied. Arch. Pharmacol. 344, 630.

Pichler, L. and W. Kobinger, 1978, Presynaptic activity at peripheral adrenergic sites and blood pressure effect of α-adrenoceptor stimulating drugs, Eur. J. Pharmacol. 52, 287.

Ramage, A.G. and S.J. Wilkinson, 1989, Evidence that different regional sympathetic outflows vary in their sensitivity to the sympathoinhibitory actions of putative 5-HT$_{1A}$ and α_2-adrenoceptor agonists in anaesthetized cats, Br. J. Pharmacol. 98, 1157.

Sannajust, F. and G.A. Head, 1994, Rilmenidine-induced hypotension in conscious rabbits involves imidazoline-preferring receptors, J. Cardiovasc. Pharmacol. 23, 42.

Sannajust, F., C. Cerutti, E. Koenig-Bérard and J. Sassard, 1992, Influence of anaesthesia on the cardiovascular effects of rilmenidine and clonidine in spontaneously hypertensive rats, Br. J. Pharmacol. 105, 542.

Schlicker, E., B.I. Armah and M. Göthert, 1990, Central presynaptic α_2-autoreceptors are involved in the blood pressure-lowering effect of moxonidine, J. Cardiovasc. Pharmacol. 16, 15.

Schmitt, H., 1977, The pharmacology of clonidine and related products, in: Handbook of Experimental Pharmacology, Antihypertensive Agents, Vol. 39, ed. F. Gross (Springer Verlag, Berlin) p. 299.

Starke, K., 1977, Regulation of noradrenaline release by presynaptic receptor systems, Rev. Physiol. Biochem. Pharmacol. 77, 1.

Starke, K., 1987, Presynaptic α-autoreceptors, Rev. Physiol. Biochem. Pharmacol. 107, 73.

Starke, K., H. Montel, W. Gayk and R. Merker, 1974, Comparison of the effects of clonidine on pre-and post-synaptic adrenoceptors in the rabbit pulmonary artery, Naunyn-Schmied. Arch. Pharmacol. 285, 133.

Starke, K., E. Borowski and T. Endo, 1975a, Preferential blockade of presynaptic α_2-adrenoceptors by yohimbine, Eur. J. Pharmacol. 34, 385.

Starke, K., T. Endo and H.D. Taube, 1975b, Relative pre-and postsynaptic potencies of α-adrenoceptor agonists in the rabbit pulmonary artery, Naunyn-Schmied. Arch. Pharmacol. 291, 55.

Starke, K., M. Göthert and H. Kilbinger, 1989, Modulation of neurotransmitter release by presynaptic autoreceptors, Physiol. Rev. 69, 864.

Starke, K., A.U. Trendelenburg and N. Limberger, 1995, Presynaptic α_2-adrenoceptors: subtype determination, Pharmacol. Commun. 6, 99.

Szabo, B. and A. Schultheiss, 1990, Desipramine inhibits sympathetic nerve activity in the rabbit, Naunyn-Schmied. Arch. Pharmacol. 342, 469.

Szabo, B., L. Hedler, K. Lichtwald and K. Starke, 1987, ACTH increases noradrenaline release in pithed rabbits with electrically stimulated sympathetic outflow, Eur. J. Pharmacol. 136, 391.

Szabo, B., L. Hedler and K. Starke, 1989, Peripheral presynaptic and central effects of clonidine, yohimbine and rauwolscine on the sympathetic nervous system in rabbits, Naunyn-Schmied Arch. Pharmacol. 340, 648.

Szabo, B., R. Urban and K. Starke, 1993, Sympathoinhibition by rilmenidine in conscious rabbits: involvement of α_2-adrenoceptors, Naunyn-Schmied. Arch. Pharmacol. 348, 593.

Trendelenburg, A.-U., N. Limberger and K. Starke, 1993, Presynaptic α_2-autoreceptors in brain cortex: α_{2D} in the rat and α_{2A} in the rabbit, Naunyn-Schmied. Arch. Pharmacol. 348, 35.

Urban, R., B. Szabo and K. Starke, 1994, Is the sympathoinhibitory effect of rilmenidine mediated by alpha-2 adrenoceptors or imidazoline receptors?, J. Pharmacol. Exp. Ther. 270, 572.

Urban, R., B. Szabo and K. Starke, 1995, Involvement of α_2-adrenoceptors in the cardiovascular effects of moxonidine, Eur. J. Pharmacol. 282, 19.

Van Meel, J.C.A., A. De Jonge, P.B.M.W.M. Timmermans and P.A. Van Zwieten, 1981, Selectivity of some alpha adrenoceptor agonists for peripheral alpha-1 and alpha-2 adrenoceptors in the normotensive rat, J. Pharmacol. Exp. Ther. 219, 760.

Van Zwieten, P.A., 1988, Pharmacology of the alpha$_2$-adrenoceptor agonist rilmenidine, Am. J. Cardiol. 61, 6D.

Van Zwieten, P.A., M.J.M.C. Thoolen, F.A.M. Jonkman, B. Wilffert, A. De Jonge and P.B.M.W.M. Timmermans, 1986, Central and peripheral effects of S 3341 [(*N*-dicyclopropylmethyl)-amino-2-oxazoline] in animal models, Arch. Int. Pharmacodyn. Ther. 279, 130.

Verbeuren, T.J., F.H. Jordaens, L.L. Zonnekeyn and A.G. Herman, 1986, Effect of the amino-oxazoline derivative S-3341 on pre- and postjunctional α-adrenoceptors in isolated blood vessels, Arch. Int. Pharmacodyn. 284, 38.

Verbeuren, T.J., A.T.D. Xuan, E. Koenig-Bérard and P. Vitou, 1990, Rilmenidine, Cardiovasc. Drug Rev. 8, 56.

Warren, J.B., C.T. Dollery, D. Sciberras and M.R. Goldberg, 1991, Assessment of MK-467, a peripheral α_2-adrenergic receptor antagonist, with intravenous clonidine, Clin. Pharmacol. Ther. 50, 71.

Waud, D.R., 1976, Analysis of dose-response relationships, in: Advances in General and Cellular Pharmacology, Vol. 1, eds. T. Narahashi and C.P. Bianchi (Plenum Press, New York) p. 145.

Westfall, T.C., 1977, Local regulation of adrenergic neurotransmission, Physiol. Rev. 57, 659.