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Clinical investigation of an antagonist at α- and β-adrenoceptors-AH 5158A

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Summary

1. The α- and β-adrenoceptor blocking action of AH 5158A was investigated in man using the veins of the hand, the arterial bed of the forearm, and certain responses of the circulation as a whole.

2. In the veins, locally infused AH 5158A resulted in specific and competitive antagonism of the constrictor response to locally infused noradrenaline and of the dilator response to isoprenaline.

3. Brachial artery infusions of AH 5158A resulted in competitive antagonism of the arterial blood flow changes produced by local infusions of noradrenaline and isoprenaline.

4. Systemic infusion of AH 5158A (0·5–0·9 mg/kg) produced clear blockade of the heart rate response to systemic infusion of isoprenaline. It also attenuated the response to exercise at 80 watts for 4 min; mean arterial pressure during exercise was reduced by 16% and heart rate by 18%. Blockade lasted at least 1 hour.

5. AH 5158A caused small changes in arterial pressure and heart rate at rest supine, but had no effect on the response of pressure and rate to tilting.

Introduction

Studies in animals have shown AH 5158A (5-[1-hydroxy-2-[(1-methyl-3-phenyl propyl) amino] ethyl] salicylamide hydrochloride) to be a specific competitive antagonist at both α- and β-adrenoceptors (Farmer, Kennedy & Levy, 1971). We report here an investigation of some of its actions in man. The activity of the substance as both an α- and β-adrenoceptor blocking agent has been assessed in the forearm arterial bed and the superficial veins of the hands. We have also studied the effect of systemic administration of the drug on some of the circulatory changes occurring during exercise, tilting to 70° and the infusion of isoprenaline.

Methods

The studies were carried out in medical students and doctors, and each subject received a full explanation of the nature of the experiment before giving his consent.

Forearm blood flow was measured in both experimental and control arms by venous occlusion plethysmography using mercury-in-rubber strain gauges; the wrist cuff occlusion pressure was 200 mmHg and the congesting cuff pressure was 40 mmHg (1 mmHg≡1·333 mbar). Flows were recorded for 10 s in each 15 seconds.

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Drugs were infused into the brachial artery through a 24 or 26 SWG unmounted needle previously introduced under local anaesthesia (1% lignocaine); the infusion was maintained by means of a constant rate infusion pump.

Superficial hand vein compliance was estimated by the technique previously described (Nachev, Collier & Robinson, 1971). The distension of the vein under study was measured at a standard congesting cuff pressure of 45 mmHg by focussing a microscope on a marker dot on the skin at the summit of the distended vein; the microscope was then refocussed on the dot when the hand vein had been allowed to collapse. The distance the microscope moved gave an index of vein size at the selected pressure. The distensibility of the vein at a cuff pressure of 45 mmHg was referred to as HVD[45].

Drugs were infused into the vein at 0·25 ml/min through a 26 SWG needle which was introduced so that its tip was 5–10 mm upstream from the point of measurement.

Arterial pressure was recorded by the auscultatory method using a standard sphygmomanometer cuff. In studies of the response to exercise, the cuff was inflated and deflated by a semiautomatic system; cuff pressure was recorded continuously by means of a pressure transducer, and the observer marked the appearance and disappearance of the Korotkoff sounds on the trace without knowing the level of pressure at the time. Mean arterial pressure was calculated by adding one-third of the pulse pressure to the diastolic pressure.

Heart rate was recorded electrocardiographically.

Forearm flow, arterial pressure and electrocardiogram were recorded on a Devices M8 recorder. The temperature of the laboratory varied from 22°–26° C, but remained essentially constant throughout each experiment.

Drugs were made up in physiological saline (0·9% w/v) containing ascorbic acid (approximately 5–10 μg/ml) to prevent oxidation of catecholamines. Doses of drugs were expressed as the salt except noradrenaline and angiotensin II which were expressed as the base and amide respectively. The drugs used were acetylcholine chloride (Roche), AH 5158A (5-[1-hydroxy-2-[(1-methyl-3-phenylpropyl) amino] ethyl] salicylamide hydrochloride) (Allen & Hanburys), angiotensin II amide (Hypertensin, Ciba), ascorbic acid (Evans Medical), synthetic bradykinin (Sandoz), histamine acid phosphate (Savory & Moore), isoprenaline sulphate, noradrenaline acid tartrate (Levophed-Bayer), propranolol hydrochloride (Inderal, I.C.I.) and 5-hydroxytryptamine creatinine phosphate (Burroughs Wellcome).

Assessment of drug action in forearm arterial bed

The agonist, either noradrenaline or isoprenaline, was infused into the brachial artery at progressively increasing rates; each rate was maintained for 3–4 min, and the average of the last five flow readings recorded during each rate of infusion was taken as the response. In some studies the antagonist was then infused for 10 min, after which a further dose-response curve to the agonist alone was recorded. In most studies, however, the antagonist was mixed with the second series of infusions of the agonist. The degree of blockade was assessed from the log dose-response curves. Changes in flow were expressed as percentages of the maximum change recorded during the control infusion.

Assessment of drug action in superficial hand veins

Noradrenaline infusions into the vein under study were made at progressively increasing rates, and the resulting venoconstriction recorded; each dose rate was maintained for 5 minutes. A continuous infusion of 5-hydroxytryptamine was given before and during each administration of isoprenaline since the dilator effect of this drug can only be demonstrated if the vein is first constricted (Collier, Nachev & Robinson, 1970). In some experiments the antagonist was infused for 10 min before the second dose-response curve was recorded. In others, the blocking drug was given mixed with increasing doses of the agonist. The degree of blockade was assessed as in the arterial studies.

Assessment of response to systemic infusion of isoprenaline

Isoprenaline was infused intravenously in increasing doses and the heart rate recorded; each dose level was maintained for 4–5 minutes. AH 5158A was then given intravenously over 10 min in a dose varying from 0·5–0·9 mg/kg. The infusion of isoprenaline was repeated, the dose level being increased until a clear response was seen.

Assessment of response to tilting to 70°

Arterial pressure and heart rate were recorded with the subject lying supine on a tilt table and again after 1 min at 70°, head up tilt. Two control tilt studies were made on each subject, and two further studies were made after systemic administration of AH 5158A.

Assessment of response to exercise

The subjects exercised in the upright position on a constant work-load bicycle ergometer (Lode Instrumenten). Heart rate and arterial pressure were recorded at rest and during the 4th minute of exercise at 80 watts. After two exercise periods separated by a rest of at least 15 min, AH 5158A (0·5 mg/kg) was infused over 10 min with the subject recumbent. The exercise was repeated approximately 5 min after the end of the infusion, and again after a rest of about 10 minutes.

Results

α-Adrenoceptor antagonism

In three experiments in which AH 5158A was infused into a superficial hand vein at 500 ng/min for 10 min, there was little or no reduction in the constriction induced by a subsequent infusion of noradrenaline; AH 5158A itself had no effect on the compliance of the resting vein. In four experiments, in which AH 5158A was infused at the same rate mixed with the noradrenaline, there was clear competitive α-adrenoceptor blockade as shown by the parallel shift to the right of the log dose-response curve. The blockade was dose dependent (three experiments) and increased progressively up to a dose rate of 32 μg/min (one experiment; Fig. 1). Further infusions of noradrenaline were given at intervals after discontinuing AH 5158A (four experiments); the blockade induced by infusion of the drug at dose rates varying from 8–32 μg/min persisted for 8–33 minutes.

In one subject the specificity of the α-adrenoceptor blockade was demonstrated by infusing noradrenaline, 5-hydroxytryptamine and angiotensin in turn mixed

with AH 5158A (10 μg/min); the constriction of the vein induced by noradrenaline was abolished but there was no attenuation of the constrictor effect of the other substances.

In five experiments on forearm flow in which AH 5158A was infused into the brachial artery at 40 μg/min either for 10 min before infusion of noradrenaline, or

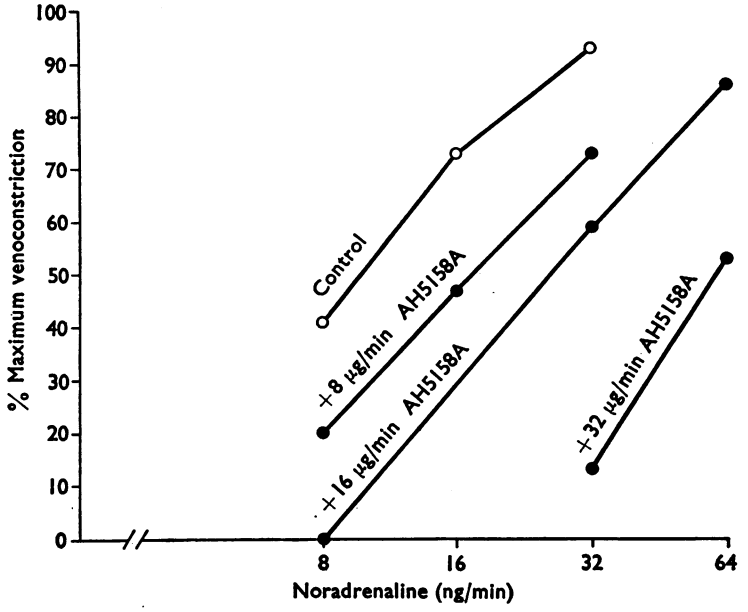

FIG. 1. Effects of increasing doses of AH 5158A on the venoconstrictor response to noradrenaline in one subject. The agonist and antagonist were infused simultaneously.

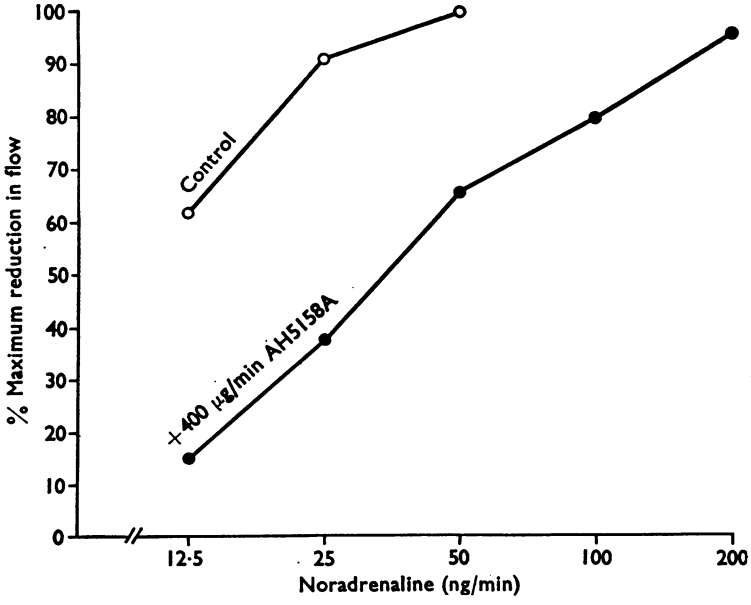

FIG. 2. Effect of AH 5158A on the response to noradrenaline in the forearm arterial bed in one subject. The agonist and antagonist were infused simultaneously.

mixed with the noradrenaline, no consistent evidence of blockade was obtained. However, when AH 5158A was given at 400 μg/min mixed with noradrenaline it induced a consistent competitive blockade of the constrictor action of the agonist (three experiments) as shown by a parallel shift to the right of the log dose-response curve (Fig. 2). AH 5158A given by itself in a dose of 40 or 400 μg/min for 3 min caused little or no increase in resting flow. After prolonged infusions at 400 μg/min (mixed with noradrenaline) large increases in flow were seen ; the total dose infused, however, reached 8 mg.

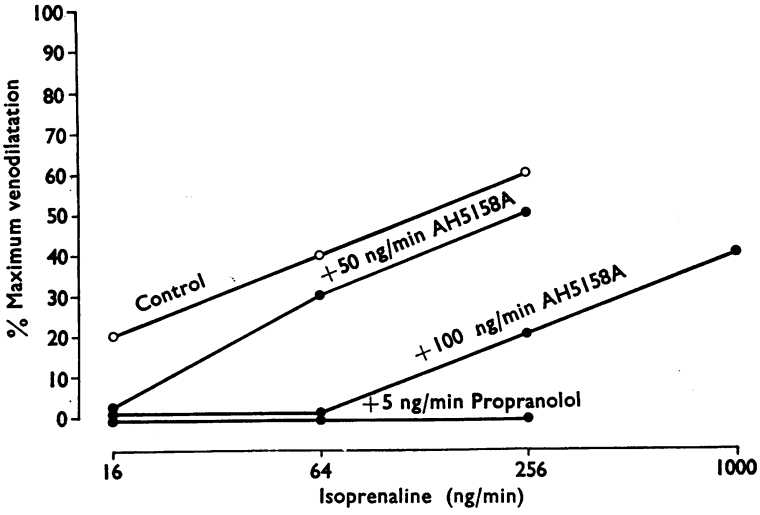

FIG. 3. Effect of AH 5158A and propranolol on the venodilator response to isoprenaline in a vein preconstricted by a continuous infusion of 5-hydroxytryptamine. The agonist and antagonist were infused simultaneously.

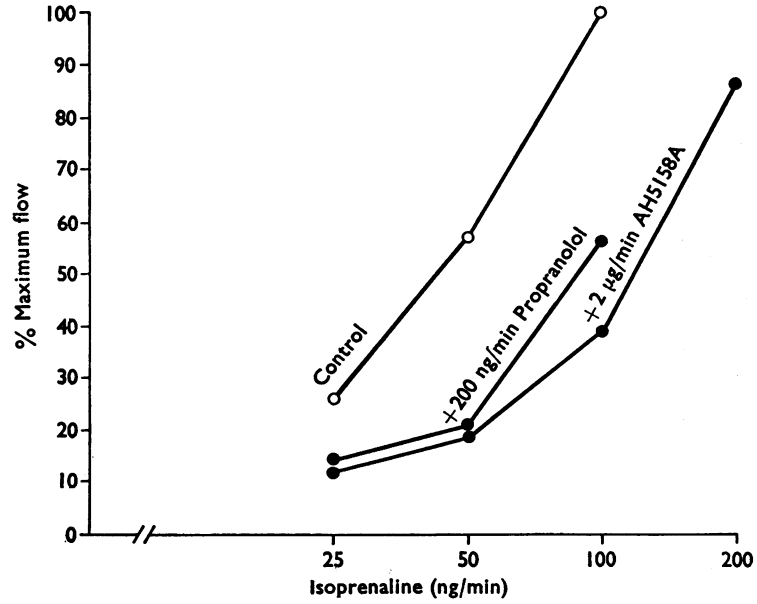

FIG. 4. Effect of AH 5158A and propranolol on the response to isoprenaline in the forearm arterial bed. The agonist and antagonist were infused simultaneously.

β-Adrenoceptor blockade

In three experiments in two subjects in which AH 5158A (50 ng–2 μg/min) was infused with isoprenaline into a preconstricted superficial vein, clear β-adrenoceptor blockade was observed. In one study in which the lowest doses of AH 5158A had been used, the blockade was dose dependent and appeared to be competitive in type (Fig. 3). In this experiment propranolol (5 ng/min) given mixed with isoprenaline was more potent than AH 5158A at 100 ng/minute. The β-adrenoceptor blockade induced by AH 5158A disappeared within 21 min of discontinuing the infusion.

The blockade of the venodilator effect of isoprenaline was specific for there was no attenuation of the dilator response to acetylcholine, histamine or bradykinin when these drugs were infused into a vein mixed with AH 5158A (10 μg/min) (one experiment). In two experiments AH 5158A was infused into the brachial artery at 40 μg/min for 10 min before infusion of isoprenaline; there was no reduction in the dilator response. In three experiments, however, in which AH 5158A was given in doses of 2–40 μg/min mixed with isoprenaline there was clear evidence of blockade (Fig. 4). In these experiments, subsequent administration of propranolol in one-tenth the dose of AH 5158A produced a similar degree of blockade. The β-adrenoceptor blockade produced by AH 5158A had largely disappeared within 15–20 minutes.

The increase in heart rate induced by systemic isoprenaline was antagonized by AH 5158A (0·5 mg/kg in two subjects; 0·9 mg/kg in one). In each experiment isoprenaline (16 μg/min) was required after AH 5158A to produce an effect on heart rate similar to that produced by 2 μg/min beforehand. In contrast to the short duration of blockade after local administration of AH 5158A, the blockade induced by systemic administration persisted for at least 48–64 min after completion of the infusion.

Effect of AH 5158A on heart rate and arterial pressure at rest supine and during tilting to 70°

In the supine position, intravenous infusions of AH 5158A (0·5 mg/kg in four subjects; 0·9 mg/kg in one) had no statistically significant effect on heart rate which averaged 71/min before and 68/min after; calculated mean arterial pressure fell from 93 to 89 mmHg ($P<0·05$). In the upright position AH 5158A caused a significant reduction in heart rate from 85 to 80/min ($P<0·02$) and a reduction in mean arterial pressure from 97 to 89 mmHg which was also significant ($P<0·05$). However, AH 5158A had no significant effect on the change in heart rate or arterial pressure induced by tilting.

Effect of AH 5158A on heart rate and arterial pressure during exercise

In five experiments there was no significant difference between the two control studies in the heart rate and arterial pressure achieved during exercise at 80 watts. When the average of the two control studies was compared with the average of the two studies after AH 5158A, there was a highly significant reduction in heart rate and arterial pressure (Table 1); the reduction in heart rate averaged 18% and in arterial pressure 16%.

Subjective sensations after systemic administration of AH 5158A

Five of the seven subjects who received a systemic dose of AH 5158A reported a tingling sensation in the scalp. This occurred towards the end of the infusion and thereafter came intermittently over the next 10–20 minutes. No other side effects were reported.

Discussion

AH 5158A blocked both α- and β-adrenoceptors in the peripheral veins and arteries of man. The dose dependent parallel shift of the log dose-response curves in the veins and the surmountable nature of the blockade are compatible with a competitive antagonism. The results confirm those obtained in animal studies (Farmer *et al.*, 1971). We have not, however, found any evidence to support their observation that the α-adrenoceptor blockade may be self-limiting with increasing drug dosage. The lack of either constrictor or dilator effects of AH 5158A on the veins suggests that the drug has no intrinsic α or β sympathomimetic action over the dose range studied. However, the subjective feeling in the scalp may indicate piloerection which has been observed with tolazoline and is a sensitive indicator of α receptor stimulation. The blockade induced by AH 5158A appeared specific for α- and β-adrenoceptors since the drug had no effect on constrictor or dilator substances operating through other mechanisms.

AH 5158A was much more active an antagonist of β-adrenoceptors than of α-adrenoceptors. In veins the drug was 40 times more potent as a β-adrenoceptor antagonist than as an α-adrenoceptor antagonist; in the arteries it was about 200 times more potent. It is of interest that the established β-adrenoceptor blocking agents are active at significantly lower dose levels than α-adrenoceptor blocking agents.

When compared with propranolol as a β-adrenoceptor antagonist, AH 5158A was about 10–20 times less potent in the peripheral vessels. When the results obtained with AH 5158A were compared with those previously obtained with thymoxamine using identical techniques (Collier, Nachev & Robinson, 1972) the two drugs were found to be of similar potency in the veins, but AH 5158A was less potent than thymoxamine in the arteries. AH 5158A is thus less potent weight for weight in the peripheral vessels than the most active β-adrenoceptor blocking agents available, but is of comparable potency to existing α-adrenoceptor blocking agents.

The blockade of isoprenaline-induced tachycardia by systemic administration of AH 5158A demonstrates that the drug is an effective antagonist of β-adrenoceptors

TABLE 1. *Response to exercise before and after AH 5158A (0·5 mg/kg)*

Subject	Control				AH 5158A			
	Heart rate (beats/min)		Mean arterial pressure (mmHg)		Heart rate (beats/min)		Mean arterial pressure (mmHg)	
	1st study	2nd study	1st study	2nd study	1st study	2nd study	1st study	2nd study
1	144	144	99	95	110	120	70	88
2	140	130	123	121	110	—	110	—
3	120	120	105	107	96	96	97	93
4	96	95	93	95	84	78	76	87
5	110	108	87	93	100	90	78	77
Mean	122	119	102	102	100	98	86	86

The fall in both heart rate and arterial blood pressure after AH 5158A was significant ($P<0·001$)

in the heart as well as those in the periphery; the degree of blockade was similar to that previously reported after administration of propranolol in a dose of 0·15 mg/kg (Epstein, Robinson, Kahler & Braunwald, 1965). Thus, on systemic administration, AH 5158A is about one-quarter as potent as propranolol. The reduction in exercise tachycardia shows that the drug is also an effective antagonist of sympathetically mediated adrenoceptor stimulation.

The antagonism by AH 5158A of the rise in arterial pressure during exercise was similar to that seen after propranolol in doses causing a similar reduction in heart rate. Furthermore, AH 5158A had only a minimal effect on supine arterial pressure and no effect at all on the change in pressure with tilting. These results suggest that blockade of sympathetically mediated stimulation of α-adrenoceptors was no more than slight despite substantial blockade of β-adrenoceptors. Thus, in clinical use, β-adrenoceptor blockade is likely to prove the dominant effect of the drug. When there is combined stimulation of α- and β-adrenoceptors in an arterial bed, administration of AH 5158A in moderate doses should potentiate the α-effects in the same way as propranolol.

The duration of action of AH 5158A after local administration was short, but after systemic administration there was no evidence of waning of the blockade up to an hour. This contrasts with the behaviour of many blocking drugs, such as phentolamine and propranolol, whose effects persist for up to 1 h after local infusion. The short duration of local blockade after infusion of AH 5158A may reflect weak affinity for the receptors. The duration of action when given locally is clearly determined by quite different factors to those influencing the duration of action when given systemically.

Differences in duration of action may be important in making comparisons between drugs, since substances that are long acting may be cumulating during continuous infusions to a much greater extent than those that are relatively short acting. An effect of this sort may explain in part the different potency ratios between AH 5158A and propranolol found after local and systemic administration.

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