Effects of the β-receptor antagonists propranolol, oxprenolol and labetalol on human vascular smooth-muscle contraction

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Summary

1. Spiral strips of human digital arteries have been studied in vitro to investigate whether DL-propranolol, D-propranolol, oxprenolol and labetalol have peripheral vascular effects in man.

2. Labetalol was a potent inhibitor of contractile responses to noradrenaline, but had less effect on responses to 5-hydroxytryptamine and barium chloride.

3. DL- and D-propranolol were equally effective inhibitors of responses to barium chloride. They were only weak antagonists of noradrenaline responses, but stronger, non-competitive antagonists of 5-hydroxytryptamine responses.

4. Oxprenolol was only a weak inhibitor of the responses to both noradrenaline and 5-hydroxytryptamine and had little effect on responses to barium chloride.

5. It is concluded that labetalol has specific α-adrenoreceptor-blocking properties, which are probably relevant to its therapeutic action in man. Propranolol has non-specific inhibitory effects on vascular smooth muscle, which might contribute to its hypotensive activity at high concentrations, but oxprenolol has only slight peripheral effects that are probably therapeutically insignificant.

Key words: adrenergic β-receptor blockade, adrenergic α-receptor blockade, anti-hypertensive agents, smooth muscle.

Introduction

The mechanism of the hypotensive effects of the β-receptor-blocking drugs is still obscure. It has been demonstrated in the past that some of these agents have inhibitory effects on the contractile responses of isolated animal vascular preparations, so peripheral vascular effects may contribute to their activity in man (Gulati, Gokhale, Parikh, Upwadia & Krishnamurty, 1969; Mazurkiewicz-Kwilecki, 1970). A new β-receptor-blocking drug, labetalol, has also specific α-receptor-blocking properties which probably contribute to its hypotensive effect (Farmer, Kennedy, Levy & Marshall, 1972). A recent report suggests that oxprenolol also acts as an α-receptor antagonist in the rabbit ear artery (Law, Rand & Story, 1978). We have therefore used a human arterial preparation to investigate whether peripheral vascular effects in general, and α-receptor-blocking properties in particular, may contribute to the hypotensive activity of propranolol, oxprenolol and labetalol in man.

Materials and methods

The preparation used was the human palmar digital artery obtained post mortem, usually within 24 h of death. This preparation has previously been shown to provide durable and reproducible responses to a variety of pharmacological agonists and antagonists (Jauernig & Moulds, 1978). Spiral strips of the arteries were prepared as has been previously described. Isometric tension was measured by using Grass FTO3 C or Hewlett Packard FTA-100 transducers and a Hewlett Packard 7758 A or
The strips were suspended at 37°C in a bathing fluid of the following composition (mmol/l): Na⁺ 137.4, K⁺ 5.4, Mg²⁺ 1.2, Ca²⁺ 2.5, Cl⁻ 131.5, HCO₃⁻ 15.0, H₂PO₄⁻ 1.2, SO₄⁻ 1.2, and glucose 11.5. For experiments with barium chloride, sulphate ions were omitted from the bath to avoid salt precipitation. Cumulative dose–response curves of the contractile responses to different agonists were performed in the absence of propranolol, oxprenolol and labetalol and then repeated in the presence of increasing concentrations of these drugs. At least five different arterial preparations were used to test the response of each antagonist. Controls consisted of repeated dose–response curves to the agonist performed on a separate preparation from the same digital artery.

Oxprenolol was selected because of its suggested α-receptor-blocking properties, and labetalol because it is a β-receptor-blocking agent with well-accepted α-receptor-blocking properties. Propranolol was selected because it is an effective and widely used β-receptor-blocking agent in the treatment of hypertension. Both the commonly used DL racemic mixture of this drug and the D isomer, which has equal non-specific effects but much less β-receptor-blocking potency, were studied.

The agonists chosen were noradrenaline, 5-hydroxytryptamine and barium chloride. We have previously demonstrated that the digital artery possesses separate receptors for noradrenaline and 5-hydroxytryptamine (Jauernig & Moulds, 1978), so specific receptor-blocking effects of the drugs were tested by comparing their inhibitory effects on the responses to noradrenaline and 5-hydroxytryptamine. The non-specific membrane-stabilizing effects of the drugs were determined by studying their effects on the responses to barium chloride, which is thought to produce smooth-muscle contraction by crossing the cell membrane and substituting for calcium ions in the excitation–contraction coupling mechanism (Bohr, 1964).

Drugs used were noradrenaline bitartrate (Sigma), 5-hydroxytryptamine creatinine sulphate (Sigma), barium chloride (British Drug Houses), DL-propranolol and D-propranolol (I.C.I.), oxprenolol (Ciba–Geigy) and labetalol (Allen and Hanbury).

Agonist dose ratios were calculated from the dose–response curves in the presence of each of the antagonist concentrations. Any change in the repeated control dose–response curves was calculated identically and experimental results were corrected accordingly. The negative logarithm of the antagonist concentration, whereby twice the dose of the agonist is needed to obtain the same response (pA₂ values), were calculated from a Schild plot (Arunlakshana & Schild, 1959). Comparisons were made by unpaired Student’s t-test.

Results

Intrinsic activity

No intrinsic contractile activity was observed with D-propranolol, DL-propranolol or labetalol in any of the concentrations used. Oxprenolol, however, at high concentrations (usually >3 × 10⁻⁵ mol/l) produced a contractile response. In one single preparation this response was not antagonized by a high concentration of phentolamine (1.28 × 10⁻⁶ mol/l).

Inhibition of responses to noradrenaline

Of the four drugs tested, labetalol (3 × 10⁻⁶ and 3 × 10⁻⁵ mol/l) was the strongest inhibitor of the contractile response to noradrenaline. The dose–response curves were shifted to the right in a dose-dependent, parallel manner with only a slight reduction of the maximum response (Fig. 1). The Schild plot gave a pA₂ value of 6.2. Labetalol had much greater effect on the response to noradrenaline than did either DL- or D-propranolol. Dose ratios in the presence of both concentrations of labetalol were significantly (P < 0.05) greater than those in the presence of the same concentrations of propranolol, and there was no significant difference between the inhibitory potencies of the two forms of propranolol. Oxprenolol was a similarly weak antagonist of noradrenaline, causing only very small, parallel shifts of the dose–response curves. Dose ratios to noradrenaline obtained in the presence of both concentrations of oxprenolol were not significantly different from those obtained in the presence of the same concentrations of either DL- or D-propranolol.

Inhibition of responses to 5-hydroxytryptamine

To decide whether the effects on the responses to noradrenaline represented specific α-receptor antagonism, dose–response curves to 5-hydroxytryptamine were performed in the presence of the four drugs in the same concentrations that were used to antagonize responses to noradrenaline. Labetalol was a much weaker inhibitor of 5-
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hydroxytryptamine than of noradrenaline, causing only small, parallel shifts of the dose–response curves (Fig. 2). Dose ratios to 5-hydroxytryptamine obtained in the presence of both concentrations of labetalol were significantly less (P < 0.05) than those to noradrenaline. Oxprenolol also produced only small, parallel shifts of the 5-hydroxytryptamine dose–response curves, with

FIG. 1. Effect on the responses to noradrenaline of the four drugs (○, 3 × 10^{-6} mol/l; ▲, 3 × 10^{-3} mol/l). ●, Control. Each curve represents the mean (+SEM) responses of at least five different arterial preparations.

FIG. 2. Effect on the responses to 5-hydroxytryptamine of the four drugs (○, 3 × 10^{-6} mol/l; ▲, 3 × 10^{-3} mol/l). ●, Control. Each curve represents the mean (+SEM) responses of at least five different arterial preparations.
dose ratios to 5-hydroxytryptamine in the presence of both concentrations of oxprenolol not being significantly different from those to noradrenaline. Dose ratios to 5-hydroxytryptamine obtained in the presence of oxprenolol were also not significantly different from those in the presence of labetalol. In contrast, DL- and D-propranolol were stronger inhibitors of 5-hydroxytryptamine than of noradrenaline, causing non-parallel shifts of the 5-hydroxytryptamine response curves and reducing the maximum responses. [The maximum response to serotonin was inhibited significantly more (P < 0.05) by the highest dose of propranolol than was the maximum response to noradrenaline.] Both forms of propranolol were equally effective as non-competitive antagonists of 5-hydroxytryptamine.

Inhibition of responses to barium chloride

Dose–response curves to barium chloride were performed in the presence of the same drug concentrations (3 × 10⁻⁶ and 3 × 10⁻⁵ mol/l) that were used with noradrenaline and 5-hydroxytryptamine, and also a higher concentration (10⁻⁴ mol/l). At the higher concentrations, DL- and D-propranolol produced near-parallel shifts of the dose–response curves to barium chloride (Fig. 3). Since the concentrations of barium chloride in the tissue bath could not be increased any further, the nature of this inhibition could not be identified unequivocally. It was, however, different from the effects of the other β-receptor antagonists tested.

Labetalol in the same three concentrations produced hardly any shift in the dose–response curves, but reduced the maximum responses by approximately 40%, which suggests non-competitive inhibition. Oxprenolol could not be shown to antagonize the responses to barium chloride, since in high concentrations (>3 × 10⁻⁵ mol/l) it caused contractures upon which the effect of barium chloride was superimposed. An accurate determination of possible inhibitory effects was therefore impossible. However, it can be seen from the curves that any such effect could only be small (Fig. 3).

Discussion

Pharmacological considerations

This study confirms that labetalol has significant α-receptor-blocking properties in this preparation of human blood vessels. This can be concluded from the higher potency of labetalol for inhibiting responses to noradrenaline than to 5-hydroxytryptamine. It does not confirm that either propranolol or oxprenolol has any specific α-receptor-

![Figure 3](image-url)
blocking activity. Indeed propranolol had somewhat greater potency for inhibiting responses to 5-hydroxytryptamine than to noradrenaline, although this may not reflect specific antagonism at the 5-hydroxytryptamine receptor site. Since the nature of the inhibition was obviously non-competitive. 5-Hydroxytryptamine antagonism by propranolol has also been reported in other tissues (Schechter & Weinstock, 1974). The inhibition of responses to barium chloride probably represents non-specific 'membrane-stabilizing' properties of these drugs. Propranolol was the most potent drug in this regard, and although slightly higher concentrations were required than to inhibit responses to noradrenaline and 5-hydroxytryptamine, it is probable that the 'membrane-stabilizing' properties of propranolol were responsible for all the effects observed. This explanation is supported by the finding that both D- and DL-propranolol were equally effective inhibitors of all contractile responses studied.

Such a mechanism would also explain the effects of oxprenolol, which could not be demonstrated effectively to antagonize responses to barium chloride, and accordingly was only a weak inhibitor of responses to noradrenaline and 5-hydroxytryptamine. In contrast to propranolol, labetalol had only slight effects on responses to barium chloride, thus providing further evidence that it antagonizes the effects of noradrenaline at the a-receptor.

The nature of the contractile responses produced by high concentrations of oxprenolol was not pursued. However, they probably do not represent intrinsic sympathomimetic activity of that drug as they were not antagonized by phentolamine in a higher concentration than that previously shown to inhibit responses to noradrenaline in this preparation (Jauernig & Moulds, 1978).

**Clinical significance**

The effects of labetalol have been demonstrated at concentrations similar to the plasma levels occurring in the therapeutic use of the drug (Richards, Maconochie, Bland, Hopkins, Woodings & Martin, 1977). Therefore, we suggest that a-receptor antagonism within peripheral blood vessels is likely to be an important component of labetalol action in man. In contrast, however, it is unlikely that peripheral effects contribute to the hypotensive activity of oxprenolol, since the effects we have observed were only minor and were observed only at concentrations considerably greater than those obtained in the clinical use of the drug (Brunner, Imhof & Jack, 1975). It is also unlikely that the intrinsic contractile activity of oxprenolol is of clinical relevance, since the effects observed again occurred only at much higher concentrations than the usual plasma levels of the drug.

The finding that there was no difference between the actions of DL- and D-propranolol in this study, whereas only DL-propranolol is thought to possess hypotensive activity in man (Rahn, Hawlina, Kersting & Planz, 1974; Waal-Manning, 1970), would also make it unlikely that the effects we have observed are predominantly responsible for the hypotensive effects of DL-propranolol. However, it is well known clinically that some patients require very high doses of propranolol to obtain a satisfactory hypotensive effect. The plasma concentrations of the drug may then approach those that produced the non-specific effects observed in this study (Johnsson & Regardh, 1976; Nies & Shand, 1975). So unless it is clearly demonstrated that even high doses of D-propranolol lack any hypotensive activity in man, the possibility must remain that a non-specific effect on vascular smooth muscle contributes to the hypotensive effect of propranolol when it is given in high doses.

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**References**


