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# Pharmacology of 4-hydroxypropranolol, a metabolite of propranolol

J. D. FITZGERALD AND STELLA R. O'DONNELL*
(WITH THE TECHNICAL ASSISTANCE OF MARILYN AUSTIN)

*Department of Pharmacology, Imperial Chemical Industries Limited, Pharmaceuticals Division, Alderley Park, Macclesfield, Cheshire*

## Summary

1. 4-Hydroxypropranolol, a metabolite produced after oral administration of propranolol, has been shown to be a $\beta$-adrenoceptor blocking drug. It is of similar potency to propranolol in antagonizing the effects of isoprenaline on heart rate and blood pressure in cats and against isoprenaline protection of guinea-pigs from bronchospasm. It is not cardioselective.

2. In rats depleted of catecholamine 4-hydroxypropranolol produced an increase in heart rate, suggesting that it has intrinsic sympathomimetic activity.

3. In anaesthetized dogs 4-hydroxypropranolol produced a decrease in heart rate and $dP/dt$ and an increase in A-V conduction time at doses within the range 0·09–1·25 mg/kg. These effects are a result of $\beta$-adrenoceptor blockade. In dogs depleted of catecholamines these same doses produced an increase in heart rate and $dP/dt$ and a decrease in A-V conduction time. These responses were antagonized by propranolol, and were due to the intrinsic sympathomimetic activity of the compound. At higher doses (5·25 and 13·25 mg/kg) a further dose dependent decrease in heart rate and $dP/dt$ and an increase in A-V conduction time occurred. This trend was also seen in animals depleted of catecholamines. These changes represent membrane stabilizing activity of 4-hydroxypropranolol.

4. 4-Hydroxypropranolol is a potent $\beta$-adrenoceptor blocking drug with both intrinsic sympathomimetic activity and membrane stabilizing activity.

## Introduction

Propranolol is used extensively in man and, after its oral administration as $^{14}$C-labelled propranolol, approximately equal peak concentrations in the blood of propranolol and a metabolite 1-(4-hydroxynaphth-1-yloxy)-3-isopropylamino-2-propanol (hereinafter referred to as 4-hydroxypropranolol; Fig. 1) have been observed (Patterson, Conolly, Dollery, Hayes & Cooper, 1970). At the time of peak concentrations in the blood the maximum degree of $\beta$-adrenoceptor blockade also occurred. In view of the production in man of 4-hydroxypropranolol as a metabolite after oral administration, it was important to know its pharmacological properties. Previous unpublished results (Barrett, personal communication) suggested that

* On study leave from University of Queensland, Brisbane, Australia.

4-hydroxypropranolol is a β-adrenoceptor blocking drug of equivalent potency to propranolol. Further study of the compound was hindered by its instability in aqueous solution. This property has remained a problem in experiments on isolated tissues in the organ bath but this paper describes pharmacological studies on 4-hydroxypropranolol carried out *in vivo* using a solution prepared in citrated saline.

## Methods

### *Studies in cats*

The β-adrenoceptor blocking potency was determined in five cats (2–5 kg) anaesthetized with chloralose (80 mg/kg i.v.). Heart rate was recorded using a cardiotachometer triggered by the R wave Lead II of the electrocardiogram (Horsfall, 1965) and femoral blood pressure using a Devices (type 4) pressure transducer (1 mmHg≡1·333 mbar). Reproducible submaximal chronotropic and depressor responses (diastolic blood pressure) to 0·2 μg/kg isoprenaline, given intravenously by a cannula in the femoral vein, were obtained. 4-Hydroxypropranolol ((1 μg/kg)/min) was then infused into the other femoral vein for 30 min and the isoprenaline responses recorded every 10 min during the infusion. The 4-hydroxypropranolol solution was kept cold in the syringe by means of an ice jacket. The rate of infusion was then increased for further periods of 30 min such that 2 and then 4 (μg/kg)/min were administered. The total dose of 4-hydroxypropranolol infused over each 30 min period was plotted against the inhibition of the isoprenaline control responses at 30 min and the dose of 4-hydroxypropranolol producing 50% inhibition of tachycardia was estimated. The inhibition of the vasodepressor response at this dose was also recorded. Mean doses for 50% inhibition for the five cats with their standard errors were then calculated.

### *Studies in rats*

Heart rates were counted in rats anaesthetized with pentobarbitone sodium (55 mg/kg i.p.) by the method described by Barrett & Carter (1970). The rats were depleted of endogenous catecholamines by pretreatment, 24 h before use, with syrosingopine (5 mg/kg s.c.). 4-Hydroxypropranolol was injected intravenously into the femoral vein as 0·1 ml/100 g rat of the appropriate concentration of drug. In some rats 0·5 mg/kg propranolol was administered 30 min before 4-hydroxypropranolol. The resting heart rate before injection and the maximum heart rate after injection were recorded. Thus the maximum change in heart rate could be obtained. Each dose was injected into at least four rats and the mean value (± standard error) of maximum change in heart rate calculated.

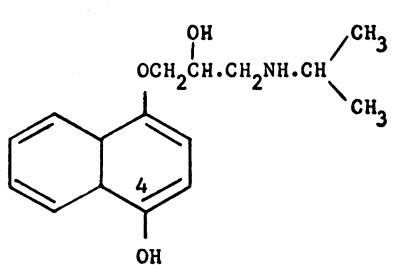


FIG. 1. Hydroxypropranolol.

*Studies in guinea-pigs*

Twenty guinea-pigs of either sex (250–400 g) were divided randomly into five groups of four animals. All were finally exposed in a pressure chamber to an aerosol of histamine (200 mg base in 76 ml distilled water) produced by 45 s spray from a nebulizer operated at 10 kg/cm$^2$ by compressed air. One group received no pretreatment: the other four were given 0·1 mg/kg of isoprenaline subcutaneously 15 min before exposure to the aerosol, and three of these groups received a subcutaneous injection of 4-hydroxypropranolol 15 min before the isoprenaline. The pressure chamber held up to four animals at a time and the experiment was so designed that four animals from different treatment groups selected at random were exposed to the aerosol together. The animals were observed and deaths recorded for 10 minutes. The number of survivors in each group was calculated. The results from several such experiments were combined to give between four and sixteen animals for each dose of 4-hydroxypropranolol; they are expressed as the percentage of animals surviving histamine bronchospasm.

*Studies in dogs*

Seven male beagle dogs (12–16 g) were used in the cardiovascular studies on 4-hydroxypropranolol. Five more dogs (13–16 g) were pretreated with syrosingopine (0·5 mg/kg i.v.) 48 and 24 h before the experiment to deplete the peripheral stores of noradrenaline (Orlans, Finger & Brodie, 1960). Three of these were used to examine the effects of 4-hydroxypropranolol alone, while the others were injected with propranolol intravenously 15 min before 4-hydroxypropranolol. The animals were anaesthetized with 30 mg/kg pentobarbitone sodium intravenously and anaesthesia was maintained with intravenous and intraperitoneal injections of pentobarbitone as required. A cuffed endotracheal tube was inserted and dogs were maintained on artificial respiration with a mixture of 60% air and 40% oxygen. Arterial and venous catheters were inserted for recording systemic blood pressure by means of a transducer and for administration of drugs. The chest was opened by median sternotomy and in some animals the mammary vessels were then cut between double ligatures. The pericardium was opened widely and sewn to the sternum to provide a cradle for the heart. A cuff-type flow probe (Statham) was placed around the aorta, a ventricular electrode sutured to the epicardial surface of the right ventricle and an atrial electrode to the right atrium for measurement of atrioventricular conduction time (Fitzgerald, Wale & Austin, 1971) and also for electrical pacing of the heart. Myocardial contractility was measured by the maximum rate of rise of pressure in the left ventricle ($dP/dt$), recorded through a cannula or a catheter tip transducer inserted into the apical dimple (Fitzgerald, *et al.*, 1971). ECG, from Lead II, and heart rate, using a cardiotachometer (Horsfall, 1965) were also recorded. The vagus nerves were sectioned in the neck and the animals left to settle for 30 min before starting control readings. All responses were recorded on an eight channel ink-writing Elema-Schonander recorder (Mingograph 81B).

From the flow-probe around the aorta flow velocity signals were detected using a Medicon 2000 flow meter. These were integrated to give stroke volume using an analogue integrator which was reset by the amplified ECG ventricular potential (Mellor, unpublished results). The probe was calibrated at the end of the experiment by removing the aorta with the flow probe *in situ* and passing through it measured

volumes of saline. Aortic flow (ml/min) was then calculated from stroke volume (ml)×heart rate (beats/min). This can be considered equivalent to the cardiac output less that volume going to the coronary vessels. Mean blood pressure was calculated from diastolic pressure+one-third pulse pressure.

Eight recordings of resting values were taken over 15 minutes. A record was made every 5 min at the intrinsic heart rate of the animal and 2 min later at a constant paced heart rate. The control values are the mean of these four readings. In all dogs the paced rate was 20–30 beats/min greater than the resting heart rate. 4-Hydroxypropranolol was administered intravenously at 0, 15, 30, 45 and 60 min in fixed doses of 0·09, 0·16, 1·0, 4·0 and 8·0 mg/kg resulting in cumulative doses of 0·09, 0·25, 1·25, 5·25 and 13·25 mg/kg. Recordings were taken at 5 and 10 min (unpaced) and 7 and 12 min (paced) after each injection and values of each parameter were taken as the mean over at least 10 complete cardiac cycles. The effects of each dose of drug were determined on each parameter by comparing the average value of the two recordings taken at 5 and 10 min (unpaced) and 7 and 12 min (paced) with the value obtained in the control recording. Changes are expressed as percentage change from the mean control value. The mean values of percentage change from control from several animals were then calculated, with standard errors, and plotted against cumulative dose. The significance of the difference of these values from zero or of the difference between two values was assessed by Student's *t* test.

### *Drugs*

The drugs used were histamine acid phosphate; 4-hydroxypropranolol hydrochloride; (±) isoprenaline sulphate; propranolol hydrochloride; practolol hydrochloride (Eraldin, I.C.I.); syrosingopine. The doses of drugs refer to the base. The 4-hydroxypropranolol was made up in normal saline containing $x/2$ citric acid, where $x$ is weight of 4-hydroxypropranolol hydrochloride. The solution was prepared immediately before use and was kept in ice during experiments. Syrosingopine was dissolved in glycerol formal and saline (6:4) immediately before use.

## Results

### *Potency as a β-adrenoceptor blocking drug*

Increasing doses of 4-hydroxypropranolol admnistered to cats reduced the tachycardia and vasodepression produced by a standard dose of isoprenaline. 4-Hydroxypropranolol (54±10 μg/kg) produced 50% reduction in heart rate response in five cats. At this dose there was 89±3% inhibition of the vasodepressor response.

The drug was similar in potency to propranolol, for which the corresponding figures are 62±10 μg/kg to produce 50% inhibition of heart rate response to isoprenaline (eight cats) at which dose 85±4% inhibition of isoprenaline vasodepression occurred (Barrett, personal communication).

### *Presence of intrinsic sympathomimetic activity*

4-Hydroxypropranolol produced a dose dependent increase in heart rate in rats depleted of catecholamines. The degree of this response resembled that to practolol

(Barrett & Carter, 1970) and was dissimilar from that to propranolol (Fig. 2). Injection of saline containing only citric acid altered the heart rate by less than 5 beats/minute. The response to 4-hydroxypropranolol (10 and 80 $\mu$g/kg) was blocked by pretreatment of the rats with 0·5 mg/kg propranolol.

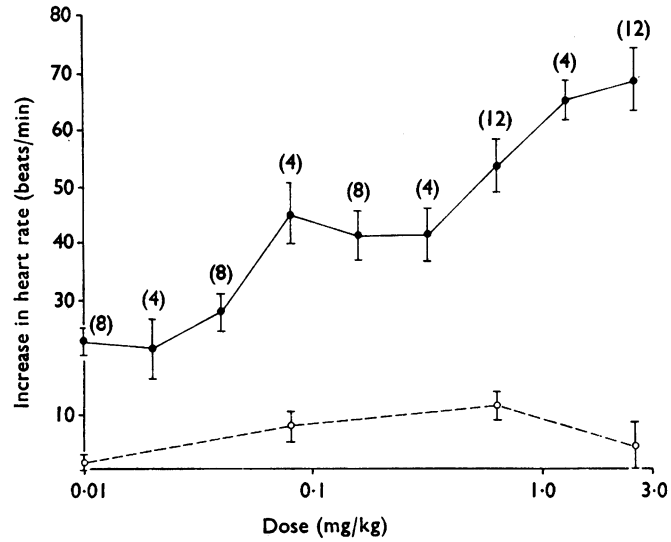


FIG. 2. Increase in heart rate after the intravenous injection of doses of either 4-hydroxypropranolol (●——●) or propranolol (○- - -○) to groups of rats depleted of catecholamines. Each point represents the mean, and one standard error is shown. The number of rats in each group is in parentheses. The mean resting heart rate of the sixty-four rats used for 4-hydroxypropranolol was 300±2·7 beats/minute. The data for propranolol are taken from Barrett (1970).

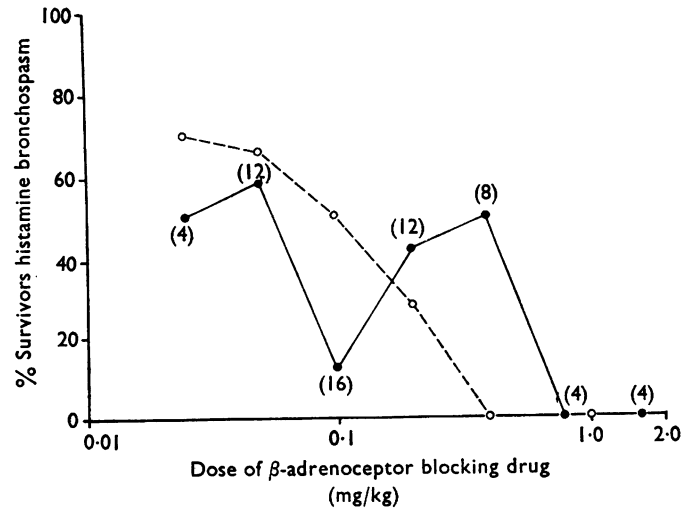


FIG. 3. Protection of guinea-pigs from bronchospasm (histamine aerosol) by injection of isoprenaline after pretreatment with various doses of either 4-hydroxypropranolol (●——●) or propranolol (○- - -○). 100% survivors represents complete protection and no antagonism of isoprenaline. 0% survivors represents no protection and complete antagonism of isoprenaline. The number of guinea-pigs in each group for 4-hydroxypropranolol is shown in parentheses. The data for propranolol are taken from Barrett (1970).

### *Antagonism of the isoprenaline protective effect against histamine-induced bronchospasm in guinea-pigs*

Exposure of control guinea-pigs to histamine aerosol resulted in no survivors, whereas pretreatment with 0·1 mg/kg isoprenaline completely protected the animals from histamine bronchospasm (100% survivors). In animals given a subcutaneous injection of a low dose (0·025 or 0·05 mg/kg) of either propranolol or 4-hydroxypropranolol some antagonism of isoprenaline occurred resulting in only 50–70% survivors. Either propranolol or 4-hydroxypropranolol (0·8 mg/kg) produced complete antagonism of isoprenaline in that no animals survived. Whereas the percentage of animals surviving bronchospasm decreased gradually as the dose of propranolol was increased, the effect of 4-hydroxypropranolol was not related to dose in a similar way (Fig. 3).

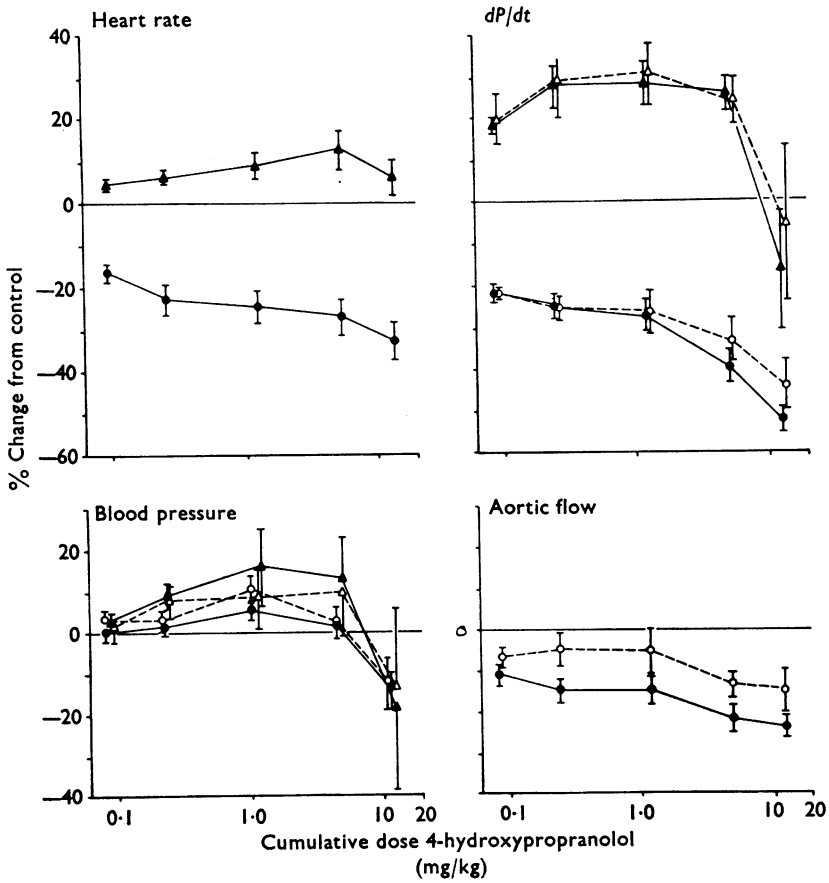


FIG. 4. Mean responses, expressed as % change from control values, to cumulative doses of 4-hydroxypropranolol. Heart rate, *dP/dt*, blood pressure and aortic flow were measured in seven normal dogs (circles) and in three dogs pretreated with syrosingopine, that is catecholamine-depleted (triangles). Values were obtained before (full lines) and after (broken lines) electrical pacing of the heart at a constant rate. Vertical bars represent standard errors of the mean.

## *Cardiovascular effects in dogs*

### *Haemodynamic effects*

The effects of 4-hydroxypropranolol on haemodynamic functions are illustrated in Fig. 4. Mean control values for the various parameters studied are given in Table 1. Pacing the heart at constant rate caused no significant change in the control values of $dP/dt$, mean blood pressure, stroke volume or aortic flow in either normal animals or animals treated with syrosingopine. However, the animals pretreated with syrosingopine had significantly lower control values of $dP/dt$ and mean blood pressure than normal animals, both in paced and unpaced conditions.

*Normal dogs.* Administration of 0·09 mg/kg 4-hydroxypropranolol, when the heart was unpaced, produced a significant decrease in heart rate (−16·5±2·1%, $P<0·001$) and in $dP/dt$ (−21·6±2·2%, $P<0·001$) accompanied by a decrease in aortic flow (−10·9±2·1%, $P<0·01$) but no change in mean blood pressure. After 0·25 mg/kg an additional 6.5% decrease in heart rate occurred, but after 1·25 mg/kg the heart rate remained constant at this value of about −24% control value. Aortic flow and $dP/dt$ were not decreased further after administration of 0·25 and 1·25 mg/kg. A slight increase in blood pressure occurred after 1·25 mg/kg. Doses of 5·25 and 13·25 mg/kg 4-hydroxypropranolol produced dose dependent, additional decreases in $dP/dt$, the value after 13·25 mg/kg (−52·0±3·10) being significantly greater than that after 1·25 mg/kg (−27·0±4·00, $P<0·001$). A dose dependent decrease in blood pressure and additional slight decreases in heart rate and aortic flow also occurred. After all doses of 4-hydroxypropranolol a slight increase in stroke volume occurred.

With the exception of aortic flow, values obtained when the heart was being paced at a constant rate (23·3±3·1 beats/min above resting rate) were not significantly different from values obtained without pacing and the trend of the effects of 4-hydroxypropranolol observed during pacing were similar to those described above for unpaced conditions. When the heart was paced, the effect of 4-hydroxypropranolol in reducing aortic flow was significantly less at all dose levels, with the exception of 0·09 mg/kg. Stroke volume was slightly decreased.

The effects of 4-hydroxypropranolol on $dP/dt$ in normal dogs at constant heart rates are compared in Fig. 5 with results obtained from similar dogs given propranolol or practolol.

TABLE 1. *Control readings in seven normal dogs and three anaesthetized dogs pretreated with syrosingopine at unpaced and paced heart rate*

| Parameter | Mean value±standard error | | | |
|---|---|---|---|---|
| | Normal | | Pretreated with syrosingopine | |
| | Unpaced | Paced | Unpaced | Paced |
| Heart rate (beats/min) | 137·7± 8·8 | 161·0± 7·3 | 89·7± 7·2 | 111·3± 7·7 |
| $dP/dt$ (mmHg/s) | 1956·2±134·3 | 2152·8±148·3 | 1369·3±310·3 | 1461·3±339·1 |
| Mean blood pressure (mmHg) | 76·5± 4·1 | 78·7± 3·8 | 57·7± 3·9 | 61·3± 4·2 |
| Stroke volume (ml) | 7·4± 1·8 | 6·3± 1·0 | — | — |
| Aortic flow (ml/min) | 989·0±174·9 | 1003·0±164·4 | — | — |
| AV conduction time (ms) | 107·8± 5·0 | 144·8± 5·0* | 133·7± 14·8 | 197·3± 10·0 |

* Sig. different from unpaced control value.

*Dogs depleted of catecholamines.* Administration of 0·09 mg/kg 4-hydroxypropranolol, when the heart was unpaced, produced an increase in heart rate (+4·5±1·2%), a significant increase in $dP/dt$ (+19·0±2·1%, $P$=0·05−0·01) and a slight increase in blood pressure. These parameters remained increased until after administration of 13·25 mg/kg when heart rate returned towards control values, blood pressure was reduced, and $dP/dt$ was reduced to below the control value (−15·8±14·2%). Values obtained when the heart was paced were not significantly different from values obtained before pacing and the same trends as described above for unpaced conditions were observed.

*A-V conduction time*

The effects of 4-hydroxypropranolol on A-V conduction time are illustrated in Fig. 6, and mean control values are given in Table 1. Pacing the heart at constant rate caused a significant increase in control A-V conduction time in both normal animals and animals pretreated with syrosingopine. The control A-V conduction time in animals pretreated with syrosingopine was significantly higher than in normal dogs both before and during pacing.

*Normal dogs.* After all doses of 4-hydroxypropranolol, the increase in A-V conduction time was greater during pacing than without pacing. Administration of 0·09 mg/kg caused an increase of 4·8±2·9% in conduction time without pacing

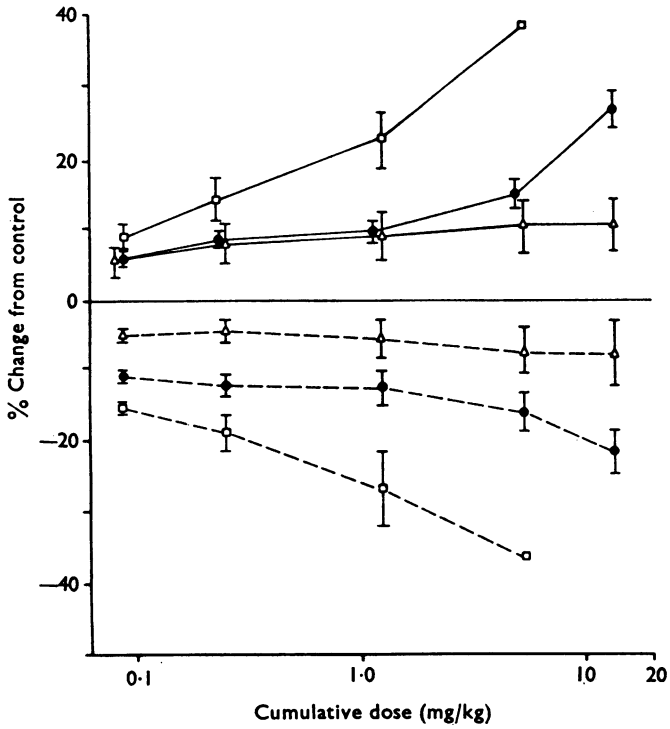


FIG. 5. Percentage change in A-V conduction time (full lines) and in $dP/dt$ (broken lines) from control values in dogs with a fixed heart rate after cumulative doses of practolol (△), propranolol (□) or 4-hydroxypropranolol (●). Each point represents the mean value and vertical bars the standard error of the mean from seven dogs given 4-hydroxypropranolol, three dogs given practolol and three given propranolol. The data for 4-hydroxypropranolol are taken from Figs. 4 and 6.

and of 11·9±2·0% ($P<0·01$) during pacing. Conduction time remained at these increased levels until after 5·25 and 13·25 mg/kg when additional marked, dose dependent increases occurred.

The effects of 4-hydroxypropranolol on A-V conduction time in normal dogs with paced heart rate are compared in Fig. 5 with results from similar dogs given propranolol or practolol.

*Catecholamine depleted dogs.* In these, in contrast to normal dogs, 0·09 mg/kg 4-hydroxypropranolol produced a significant decrease in A-V conduction time (−10·5±2·2%, $P=0·05–0·01$) when the heart was unpaced. Additional slight decreases occurred after 0·25 and 1·25 mg/kg but A-V conduction time then increased towards control values after 5.25 and 13·25 mg/kg. When the heart was paced at constant rate the A-V conduction time was still decreased after 0·09 mg/kg (−5·7±1·3%, $P=0·05–0·01$) but the decrease was not as great as in the unpaced preparation, that is the actual values for A-V conduction time were higher. The A-V conduction time remained at this constant, decreased level until after 13·25 mg/kg, when it increased towards control values . After 0·09, 0·25 and 1·25 mg/kg A-V conduction time was decreased less during pacing than without pacing but at the two high doses (5·25 and 13·25 mg/kg) there was no significant difference between unpaced and paced values.

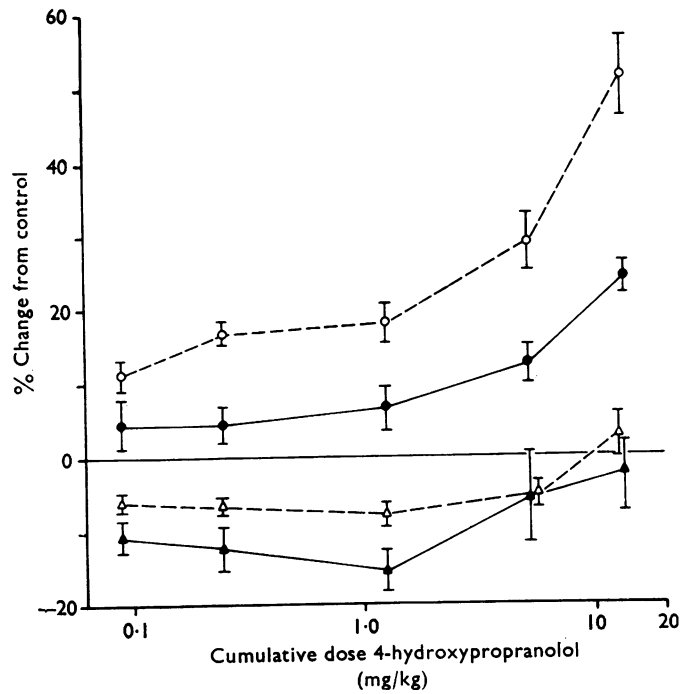


FIG. 6. Percentage change in A-V conduction time from control values after cumulative doses of 4-hydroxypropranolol. Values were obtained in seven normal dogs (circles) and three dogs pretreated with syrosingopine (triangles) both before (full lines) and after (broken lines) electrical pacing of the heart at constant rate. Each point represents the mean value and vertical bars the standard errors of the mean.

### *Effect of propranolol on responses to 4-hydroxypropranolol in catecholamine depleted dogs*

In two dogs which were given either 0·25 or 0·5 mg/kg propranolol the effects of 4-hydroxypropranolol on heart rate, $dP/dt$ and A-V conduction time were blocked. Propranolol (0·5 mg/kg) also antagonized the effects of 4-hydroxypropranolol (1·25 and 5·25 mg/kg). The reduction in heart rate and $dP/dt$ observed after 13·25 mg/kg 4-hydroxypropranolol was even greater in the presence of propranolol. Also, A-V conduction time was increased above control values whereas, without propranolol, it only increased towards control values.

## Discussion

The results obtained confirm previous unpublished observations of Barrett that the 4-hydroxylated metabolite of propranolol is approximately equipotent with the parent drug as a β-adrenoceptor blocking agent. In anaesthetized cats the dose of 4-hydroxypropranolol required to produce 50% reduction in isoprenaline tachycardia was similar to that of propranolol. 4-Hydroxypropranolol is also non-selective in its β-blocking potency in that there was no marked difference in the dose required to produce block of tachycardia and of vasodepression produced by isoprenaline in cats. Also, 4-hydroxypropranolol antagonized the protective effect of isoprenaline against histamine bronchospasm in essentially the same dose range as propranolol. However, the effects of 4-hydroxypropranolol in this test were dissimilar to those of propranolol in that they were not clearly dose dependent. It is difficult to explain why the antagonism of 4-hydroxypropranolol to isoprenaline is greater at 0·1 mg/kg than at 0·25 and 0·5 mg/kg. Subsequent doses of 0·8 and 1·5 mg/kg completely antagonized the fixed dose of isoprenaline. A similar response has been observed with oxprenolol and clearly additional studies are required before an explanation of the findings can be given.

Propranolol has no agonist activity (Black, Duncan & Shanks, 1965; Barrett & Carter, 1970). 4-Hydroxypropranolol is a partial agonist in that it has intrinsic sympathomimetic activity as well as β-adrenoceptor blocking potency. This sympathomimetic activity was shown by the increase in heart rate produced after its administration to rats of catecholamines. The dose-response relationship resembled that to practolol (Barrett & Carter, 1970) and the response was blocked by propranolol. The intrinsic sympathomimetic activity was also observed during the cardiovascular studies on dogs depleted of catecholamines, in that 4-hydroxypropranolol increased heart rate and contractility and decreased conduction time (*vide infra*). These effects were also blocked by propranolol. It has been suggested that β-adrenoceptor blocking drugs which are partial agonists might be less likely to aggravate cardiac failure than agents with no agonist properties (Ablad, 1967). Thus, the practical implications of the finding that a propranolol metabolite has agonist activity may appear to be attractive. However, when the blocking drug itself is a partial agonist, the initial effect of the drug will be β-adrenoceptor stimulation before β-adrenoceptor block. 4-Hydroxypropranolol is produced as a result of the metabolism of propranolol and, since free propranolol blocks β-adrenoceptors, it is reasonable to assume that the 4-hydroxy derivative will be unable to exert its agonist activity when produced as a metabolite in the body.

The effects of 4-hydroxypropranolol on haemodynamic function and on A-V conduction time were studied in anaesthetized dogs. The rate of rise of left ventricular pressure ($dP/dt$) was used as a quantitative index of contractility. However, various factors can affect $dP/dt$ besides the direct effects on the cardiac muscle of the drug being administered, for example changes in aortic pressure or in heart rate (Furnival, Linden & Snow, 1970). For each 10 mmHg change in mean aortic pressure, Furnival *et al.* describe about 120 mmHg/s change in $dP/dt$. In our experiments mean aortic pressure was not kept constant but the maximum change in blood pressure was 12 mmHg and would introduce only a trivial error in the $dP/dt$ readings. Changes in heart rate have much greater effect on $dP/dt$ but if the heart rate is kept constant then any changes in $dP/dt$ observed after the drug can be assumed to result from effects of the drug on the heart which are not related to changes in rate. Thus $dP/dt$ recordings were also made with the heart rate kept at a constant value by electrical pacing. In normal dogs this mean pacing rate was twenty-three beats/min above the animals' resting heart rate. This higher rate of pacing was selected to ensure that the intrinsic heart rate would not compete with the pacing rate. The increase in rate had no significant effect on the control values of $dP/dt$, blood pressure or aortic flow but it caused a significant rise in the resting values for A-V conduction time. The values of $dP/dt$ obtained after administration of 4-hydroxypropranolol were also not different before and during electrical pacing. This may be because the changes in rate were not of sufficient degree to bring out the difference, or the difference may not be detectable in the presence of 4-hydroxypropranolol.

The dose-response relationships obtained for 4-hydroxypropranolol on the various parameters examined (see Figs. 4 and 6) might be explained on the hypothesis that 4-hydroxypropranolol has three properties: (a) $\beta$-adrenoceptor blockade, (b) intrinsic sympathomimetic activity, and (c) membrane stabilizing activity.

The initial fall in heart rate and reduction in $dP/dt$ observed in normal dogs after 0·09 mg/kg 4-hydroxypropranolol were not observed in animals depleted of catecholamines. The spontaneous release of endogenous catecholamines increases heart rate and contractility by an action on the sino-atrial node and the ventricular muscle. Thus the responses to 4-hydroxypropranolol in normal dogs result from its $\beta$-adrenoceptor blocking action, that is antagonism of endogenous catecholamines at $\beta$-adrenoceptors in the heart. Further decreases in heart rate and $dP/dt$ only occurred after much higher doses of drug suggesting that the lowest dose examined (0·09 mg/kg) produced complete $\beta$-adrenoceptor blockade. The reduction in $dP/dt$ still occurred when the negative chronotropic effect of 4-hydroxypropranolol was prevented by electrical pacing. This suggests that the reduction in $dP/dt$ observed at fixed rates is due to antagonism of $\beta$-adrenoceptors concerned with inotropic activity and does not result from the decreased heart rate. In normal dogs there was also an increase in A-V conduction time after 0·09 mg/kg 4-hydroxypropranolol. This was not observed in dogs depleted of catecholamines and is probably also a consequence of $\beta$-adrenoceptor blockade. Cardiac sympathetic nerve stimulation (Wallace & Sarnoff, 1964) or exogenous catecholamines (Kabela & Mendez, 1966) facilitate atrioventricular conduction, that is decrease conduction time. Kabela & Mendez (1966) have shown that propranolol reverses the effects of adrenaline and isoprenaline, that is increases A-V conduction time, as a result of $\beta$-adrenoceptor blockade.

The administration of a β-adrenoceptor blocking drug to animals depleted of catecholamines, in which there is no existing sympathetic tone or circulating catecholamines, should produce no changes in haemodynamic function or A-V conduction time. However, the administration of the lower doses (0·09, 1·25 and 5·25 mg/kg) of 4-hydroxypropranolol to dogs depleted of catecholamines produced a consistent increase in heart rate and $dP/dt$ and a decrease in A-V conduction time, that is it facilitated conduction. These effects mimic the effects of endogenous catecholamines and can be explained by the intrinsic sympathomimetic action of 4-hydroxypropranolol. These effects of the drug were prevented by the prior administration of propranolol, a β-adrenoceptor blocking drug without intrinsic sympathomimetic activity, and can thus be assumed to result from an agonist action on β-adrenoceptors. In dogs depleted of catecholamines there was a lesser decrease in A-V conduction time in paced than unpaced conditions. This is possibly because, without electrical pacing, the intrinsic sympathomimetic activity of 4-hydroxypropranolol affects both the S-A node, to increase heart rate, and the A-V node, to facilitate conduction, and both tend to decrease conduction time. With electrical pacing, only the agonist effects on the A-V node are observed, resulting in a lesser effect on conduction time. After high doses of the drug, when no separation of the paced and unpaced values occurred, the membrane stabilizing activity (*vide infra*) may have become predominant.

The intermediary doses of 4-hydroxypropranolol (0·25 mg and 1·25 mg/kg) produced no significant further changes in the cardiovascular parameters, but after 5·25 mg/kg and 13·25 mg/kg a further decrease in heart rate and $dP/dt$ and a sharp increase in A-V conduction time occurred. Since this same pattern occurred in the catecholamine-depleted dogs after the higher doses of drug, the effects must be due to some other property of the drug. This could be membrane stabilization (Langslet, 1970; Fitzgerald *et al.*, 1971). There was no statistically significant change in blood pressure until after 13·25 mg/kg in either normal or depleted dogs and the fall then observed presumably reflects the marked depression of ventricular function. High doses of 4-hydroxypropranolol (5·25 and 13·25 mg/kg) caused a greater increase in A-V conduction time in the presence of propranolol (0·5 mg/kg). This is probably due to the greater membrane activity when both drugs are present.

Cardiac output, as reflected by aortic flow, was decreased after 4-hydroxypropranolol. This was probably because the slight increase in stroke volume which occurred did not compensate for the larger decrease in heart rate. When the heart rate was maintained constant by electrical pacing there was little reduction in cardiac output after any dose of 4-hydroxypropranolol, emphasizing the importance of its negative chronotropic action in reducing cardiac output.

Comparison of the effects of propranolol, 4-hydroxypropranolol and practolol in decreasing $dP/dt$ and increasing A-V conduction time reflects the different pharmacological properties of these compounds (Fig. 5). Low doses (0·09–1·25 mg/kg) of all three drugs cause an increase in A-V conduction time and decrease in $dP/dt$. Subsequent doses of propranolol cause further dose-dependent changes in both parameters, whilst in contrast, practolol has little further effect. 4-Hydroxypropranolol behaves like practolol in the low and intermediate dose range (0·09–1·25 mg/kg) but resembles propranolol at the highest dose (13·25 mg/kg). This difference between propranolol and 4-hydroxypropranolol at intermediate doses.

(1·25–5·25 mg/kg) may be due to the presence of intrinsic activity in 4-hydroxypropranolol, which is also present in practolol.

The production of 4-hydroxypropranolol as a potent, active metabolite of propranolol may have some significance in the clinical use of the latter drug. Detectable circulating levels of 4-hydroxypropranolol are produced only after oral administration of propranolol and not after intravenous injection. Coltart & Shand (1970) demonstrated in man that the concentration of propranolol in the blood required to obtain maximum reduction of the tachycardia of strenuous exercise after oral administration of the drug was 40% of the concentration after intravenous administration. The production of 4-hydroxypropanolol as a $\beta$-adrenoceptor blocking metabolite after oral propranolol would explain this finding. Hayes & Cooper (1971) have shown that the hydroxylation of propranolol occurs in the liver since, after intraportal administration of propranolol in the dog, concentrations of the 4-hydroxy metabolite in the blood were equivalent to those seen after oral administration. When propranolol is administered orally to man all the drug must pass through the liver, which will be presented with a sudden " surge " of the drug. Hayes & Cooper suggest that under these circumstances, either (a) the main metabolic pathway for propranolol is saturated, allowing hydroxylation to occur or (b) the high concentrations of 4-hydroxypropranolol produced from propranolol overload the pathway for its further metabolism resulting in free 4-hydroxypropranolol in the blood. It is thus possible that the production of 4-hydroxypropranolol could be related to the dose of propranolol administered orally. It has frequently been reported that a wide range of doses of propranolol is required to achieve adequate clinical response in different patients (Gillam & Pritchard), 1966; Wolfson & Gorlin, 1967). It is possible that the pattern of metabolism varies between patients. Some may convert it only to inactive metabolites, others may produce the active 4-hydroxypropranolol. Both propranolol and 4-hydroxypropranolol are bound to a significant extent by tissues, particularly the lung (Hayes & Cooper, 1971). This may explain why 4-hydroxypropranolol is not found after intravenous administration since the liver itself will receive a reduced surge of drug to metabolite. However, the binding of propranolol and 4-hydroxypropranolol by tissues for quite some time after concentrations in the blood are undetectable, could have relevance in the pharmacological effects observed, particularly in maintaining the response for a longer duration.

This study has shown that 4-hydroxypropranolol is a $\beta$-adrenoceptor antagonist similar to propranolol in potency and non-selectivity but differing in having intrinsic sympathomimetic activity. This information may be useful in understanding the effects of propranolol administered orally in man. Furthermore the availability of an analogue of propranolol which resembles it so closely in chemical structure but differs in possessing intrinsic activity may be helpful in ascertaining the role of intrinsic sympathomimetic activity.

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