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Bisoprolol improves perfusion of ischaemic myocardium in anaesthetized pigs

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1 The ability of the cardioselective β-adrenoceptor antagonist bisoprolol ((±)-1-[4-(2-isopropoxyethoxymethyl)-phenoxy]-3-isopropyl-amino-2-propanol hemifumarate, EMD 33512) to suppress isoprenaline-induced increases in heart rate and maximal rate of rise in left ventricular pressure ($LVdP/dt_{max}$) was studied in 6 anaesthetized pigs given 4 cumulative doses (16, 64, 256 and 1024 $\mu g\,kg^{-1}$). Bisoprolol was about 2 times more effective in suppressing isoprenaline-induced increases in $LVdP/dt_{max}$ than those in heart rate.

2 In 8 animals which had a partial stenosis of the left anterior descending coronary artery (LADCA), the effects of 3 consecutive doses (50, 200 and 750 $\mu g\,kg^{-1}$) of bisoprolol were studied on systemic haemodynamics, regional myocardial perfusion and function. The effects of the drug were compared with those obtained in a group of 9 animals with LADCA stenosis which did not receive any treatment.

3 The lowest dose of bisoprolol (50 $\mu g\,kg^{-1}$) increased perfusion of the ischaemic myocardium (which had been reduced from $123 \pm 20\,ml\,min^{-1}\,100\,g^{-1}$ to $42 \pm 11\,ml\,min^{-1}\,100\,g^{-1}$) by $21 \pm 10\,ml\,min^{-1}\,100\,g^{-1}$ ($P < 0.05$). In particular the subendocardial layers, which were most severely affected by the stenosis (a decrease from $128 \pm 19\,ml\,min^{-1}\,100\,g^{-1}$ to $20 \pm 6\,ml\,min^{-1}\,100\,g^{-1}$) benefited from the administration of the drug (an increase of $30 \pm 10\,ml\,min^{-1}\,100\,g^{-1}$). Perfusion of the subepicardium was not significantly affected. With the higher dose only a minor additional improvement in perfusion of the ischaemic myocardium was observed.

4 The negative chronotropic response is the most likely factor leading to the improvement in perfusion.

5 Myocardial wall thickening, which decreased from 41 ± 2% to 9 ± 4% ($P < 0.05$) due to the hypoperfusion, did not improve after administration of the drug. This lack of improvement may possibly be due to the duration of ischaemia before and the magnitude of the flow deficit after bisoprolol administration.

6 Between 15 and 60 min of ischaemia, 5 of the 9 untreated animals had an episode of ventricular fibrillation compared with only 1 of the 8 animals treated with bisoprolol, in spite of an initially larger flow reduction in the treated animals. The more homogeneous flow distribution after bisoprolol might account for the lower incidence of arrhythmias in this group.

7 It was demonstrated that bisoprolol improves perfusion of ischaemic myocardium in anaesthetized pigs even at doses (50 $\mu g\,kg^{-1}$) that only moderately antagonize isoprenaline-induced cardiostimulatory effects.

Introduction

In spite of the increasing number of drugs of different mechanisms available, β-adrenoceptor antagonists still remain the drugs of choice for the treatment of myocardial ischaemia. Since recent studies have shown that this class of drugs may also be useful in preventing sudden cardiac death after acute myocardial infarction, the area of application of β-adrenoceptor antagonists has widened (Norwegian Multicenter Study Group, 1981; Hjalmarson *et al.*, 1981). One such new drug is bisoprolol

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((±)-1-[4-(2-isopropoxyethoxymethyl)-phenoxy]-3-isopropylamino-2-propanol hemifumarate, EMD 33512) which is devoid of intrinsic sympathomimetic activity (Harting *et al.*, 1986) and exhibits selectivity for β_1-adrenoceptors in several studies performed in anaesthetized dogs, guinea-pigs and cats (Schliep & Harting, 1984; Schliep *et al.*, 1986). In conscious pigs, bisoprolol antagonized isoprenaline-induced increases in the maximum rate of rise in left ventricular pressure ($LVdP/dt_{max}$) more effectively than those in heart rate. In the same model, propranolol was more potent than bisoprolol but inhibited isoprenaline-induced changes in these two parameters equally (Duncker *et al.*, 1987).

The cardiovascular profile of bisoprolol is well documented and it resembles other β-adrenoceptor antagonists (Harting *et al.*, 1986; Verdouw *et al.*, 1987a,b). Although bisoprolol reduces ST-segment elevation during short-lasting intermittent occlusions of the left anterior descending coronary artery (LADCA) in anaesthetized dogs (Harting *et al.*, 1986), information about its effects on blood flow in and function of the ischaemic myocardium is not available. In this study we describe the effects of three consecutively administered doses of bisoprolol on myocardial perfusion and performance in anaesthetized open-chest pigs with a graded coronary artery stenosis. In order to discover whether anaesthesia affected the β-adrenoceptor antagonist potency of bisoprolol we also studied the drug's ability to inhibit isoprenaline-induced changes in heart rate and myocardial contractility.

Methods

General

Studies were performed on Yorkshire pigs (21–35 kg) after an overnight fast. After sedation with 120 mg azaperone i.m. (Heykant *et al.*, 1971), the animals were anaesthetized with 150 mg metomidate i.v. (Dimigen & Reetz, 1970) and subsequently intubated for artificial ventilation with a mixture of oxygen and nitrous oxide (1:2). Respiratory rate and tidal volume were adjusted to control arterial blood gas values (ABL-3, Radiometer, Copenhagen, Denmark). In the present study arterial blood pH, P_{CO_2} and P_{O_2} were 7.44 ± 0.01, 41 ± 2 mmHg and 160 ± 5 mmHg, respectively.

Catheters were placed in the superior vena cava via the jugular vein for the administration of 100 mg kg^{-1} α-chloralose (Merck, Darmstadt, F.R.G.) followed by an infusion of 5 mg kg^{-1} h^{-1} pentobarbitone sodium (Sanofi, Paris, France). When necessary, sodium bicarbonate (8.4%) and Haemaccel (Behringwerke, Marburg, F.R.G.) were infused to correct base deficit and blood loss. Left ventricular and central aortic pressures were monitored with microtipped catheters (Honeywell-Philips, Best, the Netherlands).

After the heart had been exposed by a midsternal split, electromagnetic flow probes (Skalar, Delft, the Netherlands) were placed around the ascending aorta and the LADCA. The vein accompanying the LADCA was cannulated with a polyethylene catheter for the withdrawal of blood samples for determination of coronary venous blood gases. The left atrial appendage was catheterized for the injection of radioactive microspheres. Finally, a commercially available inflatable balloon (R.E. Jones, Silver Spring, Maryland, U.S.A.) was placed around the LADCA distal to the flow probe and connected to a 1 ml syringe (Hamilton Bonaduz, Bonaduz, Switzerland) which was driven by a micrometer.

Regional myocardial function

Regional myocardial function was estimated from myocardial wall thickness recordings obtained with the aid of a 5 MHz ultrasonic transducer (Krautkramer-Branson, Lewistown, Pa, U.S.A.) sutured onto a part of the epicardial surface perfused by the LADCA (Verdouw *et al.*, 1981). From the wall thickness at end-diastole (EDT) and end-systole (EST), percentage systolic wall thickening (SWT) was calculated as: $SWT\ (\%) = 100 \times (EST - EDT)/EDT$.

Regional myocardial blood flows

Just before the injection of about 2×10^6 microspheres (15 ± 1 (s.d.) μm diameter) (NEN Company, Dreieich, F.R.G.), labelled with either ^{46}Sc, ^{95}Nb, ^{103}Ru, ^{113}Sn or ^{141}Ce, the withdrawal of an arterial reference sample was started at a flow rate of 10 ml min^{-1} and continued for a period of about 1 min after injection of the microspheres. After completion of the experiment, the ischaemic myocardium was identified by intra-arterial injection of methylene blue (P4125, Sigma Chemical Company, St Louis, MO, U.S.A.) while the LADCA was completely occluded at the site of the inflatable balloon. Then the animal was killed and the excised heart was fixed in 4% formalin for at least 24 h. Epicardial fat and large vessels were removed. The atria and the right ventricle were separated from the left ventricle, including the intraventricular septum. Radioactivity in the tissues was counted to obtain blood flow data as described previously (see Saxena & Verdouw, 1984).

Experimental protocols

In the first group of six animals the responses of heart rate and $LVdP/dt_{max}$ to isoprenaline (0.025,

0.05, 0.1, 0.2, 0.4 and 0.8 $\mu g\,kg^{-1}$) were evaluated following four doses (16, 64, 256 and 1024 $\mu g\,kg^{-1}$) of bisoprolol. Each of the doses of bisoprolol was administered over a period of 2 min at 60 min intervals. Dose-ratios of isoprenaline after each dose of bisoprolol (cumulative doses 16, 80, 336 and 1360 $\mu g\ kg^{-1}$) were calculated for increases in heart rate of 40 beats min^{-1} and in $LVdP/dt_{max}$ of 2000 mmHg s^{-1}, which is approximately 50% of the maximal response for each of the two parameters.

In the second group of 21 animals, baseline values were obtained for systemic haemodynamics, regional myocardial function and arterial and coronary venous blood gases, while a batch of microspheres was injected after systemic haemodynamic parameters had been stable for at least 30 min. Subsequently, the flow in the LADCA was reduced gradually by a slow inflation of the balloon until regional systolic wall thickening had decreased to approximately 20% of baseline. The presence of ischaemia was also confirmed by the reduction of LADCA blood flow and the appearance of myocardial proton release. In this model the latter is an accurate reflection of myocardial lactate production (Schamhardt *et al.*, 1981a). When necessary, minor adjustments in the degree of flow reduction were performed during the first 5 min of ischaemia, but thereafter, the degree of stenosis was kept constant. After post-ischaemic (15 min) measurements the animals were divided into two groups. Nine animals did not receive any treatment and served for evaluation of the stability of the preparation. In these animals all measurements were repeated after 30, 45 and 60 min of flow reduction. Eight other animals received 3 consecutive doses of bisoprolol (50, 200 and 750 $\mu g\,kg^{-1}$), administered over a period of two min at 15 min intervals. In these animals all measurements were repeated 10–12 min after the administration of each dose of bisoprolol. Because of technical failure, in one animal regional blood flows could not be determined after the highest two doses.

Since the magnitude of the systolic wall thickening depends on the location of the transducer on the myocardium (Verdouw *et al.*, 1980), we did not determine the regional function of the adjacent control segment, but evaluated the effects of bisoprolol on systolic wall thickening of the LADCA perfused area in a separate group of 10 animals with an intact coronary circulation.

Four animals had an episode of ventricular fibrillation in the first 15 min of ischaemia and were excluded from further study. When, however, ventricular fibrillation occurred after 15 min of ischaemia, the animals were included in the study until precipitation of this arrhythmia. Antiarrhythmic drugs were not administered during the course of these experiments.

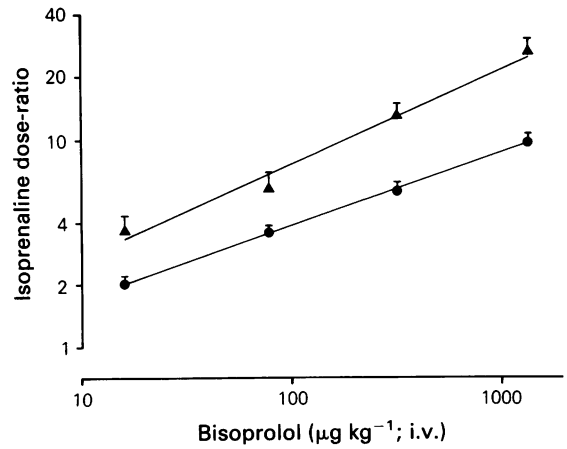

Figure 1 Effect of bisoprolol on isoprenaline-induced increases in heart rate (●) and left ventricular (LV) dP/dt_{max} (▲) in 6 anaesthetized pigs. Note that bisoprolol was about 2 times more effective in antagonizing $LVdP/dt_{max}$ changes than heart rate changes induced by isoprenaline. Data have been presented as means with vertical lines showing s.e.mean.

Drugs

Except for the anaesthetic drugs only (−)-isoprenaline sulphate (Pharmacy Department, Erasmus University, Rotterdam) and bisoprolol hemifumarate (Merck, Darmstadt, F.R.G.) were used. Both drugs were dissolved in isotonic saline and doses refer to the respective salts.

Data presentation and statistical analysis

All data are presented as mean ± s.e.mean. The significance of the changes produced by the reductions in LADCA flow in the untreated and bisoprolol-treated animals was evaluated by Duncan's new multiple range test once an analysis of variance had revealed that the samples represented different populations (Steel & Torrie, 1980). The significance of the bisoprolol-induced changes was determined by comparing these changes with those observed in the untreated animals at comparable points of time by use of Student's unpaired *t*-test.

Results

Antagonism of cardiac responses to isoprenaline

Modifications of isoprenaline-induced changes in heart rate and $LVdP/dt_{max}$ by bisoprolol are presented in Figure 1. It is clear that bisoprolol was approximately twice as effective in antagonizing the

effects of isoprenaline on LV$dP/dt_{max}$ compared with those on heart rate.

Ventricular arrhythmias between 15 and 60 min after LADCA stenosis

In 5 of the 9 animals in the untreated group the protocol was not completed because of ventricular fibrillation (in 2 animals between 30 and 45 min and in 3 animals between 45 and 60 min). The incidence of this fatal arrhythmia was considerably lower in the animals which received bisoprolol since ventricular fibrillation occurred in only 1 of the 8 treated animals after administration of the second dose between 30 and 45 min.

Myocardial proton release

Within two minutes of LADCA blood flow reduction, the coronary arterio-venous difference in pH increased from 0.05 ± 0.01 to 0.28 ± 0.04 in the untreated animals and from 0.06 ± 0.01 to 0.27 ± 0.03 in the animals which later received bisoprolol. These data did not change significantly during the first 15 min but started to decline gradually thereafter. This pattern has been well described and excluded myocardial proton release as a variable to evaluate the stability of the preparation (Verdouw *et al.*, 1978). Additionally, the rate of decline in myocardial proton release also depends on factors unrelated to the severity of ischaemia. Therefore, this variable is not suitable for the evaluation of the effects of pharmacological interventions (Verdouw *et al.*, 1978) and was not further studied during the course of experiments.

Regional myocardial blood flow

During the first 15 min of ischaemia, perfusion of the myocardium supplied by the LADCA in the untreated animals decreased from 132 ± 14 ml min^{-1} 100 g^{-1} to 71 ± 14 ml min^{-1} 100 g^{-1} (Figure 2). This decrease was not equally distributed over all myocardial layers since the deficit in perfusion of the subendocardial layers (65%) was more severe than that in the subepicardial layers (25%). Up to 45 min of flow reduction, perfusion values did not change significantly; the data obtained at 60 min showed a tendency for the ischaemic area to deteriorate further. As a result of the small number of observations ($n = 4$), we could not determine if there was an additional significant decrease in perfusion compared with that observed after 15 min of ischaemia. Perfusion of the control segment tended to increase during the first 30 min of ischaemia, but a significant increase could only be demonstrated for

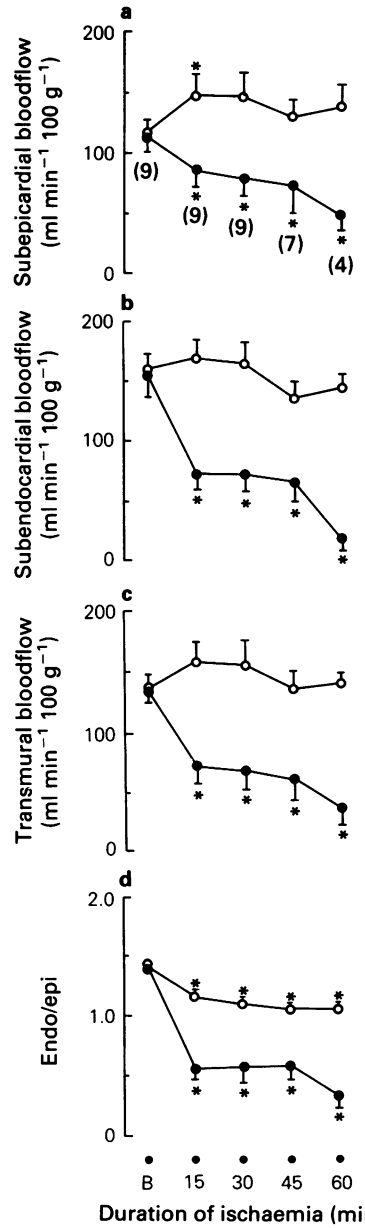

Figure 2 Myocardial perfusion in 9 open-chest anaesthetized pigs with a graded stenosis in the left anterior descending coronary artery. (a) Subepicardial blood flow, (b) subendocardial blood flow and (c) transmural blood flow. (d) Endo/epi = ratio of normalized subendocardial and subepicardial blood flows. (○) Non-ischaemic segment, (●) ischaemic segment. The numbers in parentheses indicate the number of observations. *$P < 0.05$ vs baseline (B). Data have been presented as means with vertical lines showing s.e.mean.

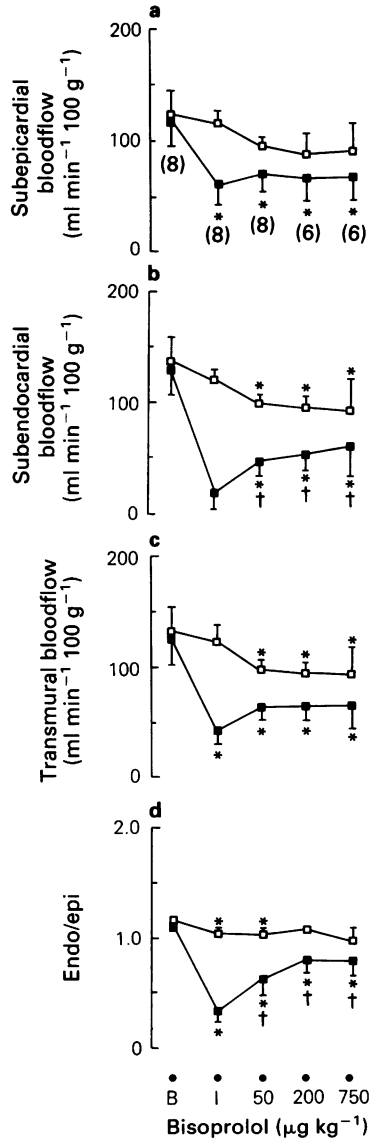

Figure 3 Effect of bisoprolol on myocardial perfusion in 8 open-chest anaesthetized pigs with a graded stenosis in the left anterior descending coronary artery. (a) Subepicardial blood flow, (b) subendocardial blood flow and (c) transmural blood flow. (d) Endo/epi = ratio of normalized subendocardial and subepicardial blood flows. The three doses of bisoprolol were administered over a period of 2 min at 15 min intervals. The first dose was given after 15 min of ischaemia (I); (□) non-ischaemic segment; (■) ischaemic segment. The numbers in parentheses indicate the number of observations. * $P < 0.05$ vs baseline (B). † $P < 0.05$ vs I. Data have been presented as means with vertical lines showing s.e.mean.

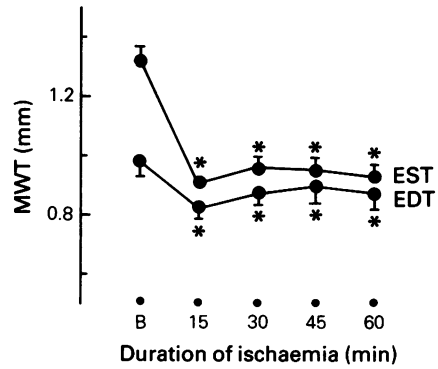

Figure 4 Myocardial wall thickness (MWT) of the ischaemic segment in 9 open-chest anaesthetized pigs with a graded stenosis in the left anterior descending coronary artery. EDT and EST are the wall thickness at end-diastole and end-systole, respectively. * $P < 0.05$ vs baseline (B). Data have been presented as means with vertical lines showing s.e.mean.

the subepicardial layers after 15 min of ischaemia. Although the perfusion of the individual layers was not grossly affected, there was a slight decrease in the ratio of the normalized subendocardial to subepicardial blood flow (endo/epi ratio).

In animals which were later given bisoprolol, the initial reduction in LADCA flow was larger (−65%) than in the untreated animals (−45%): the perfusion of myocardium supplied by the LADCA was reduced from $123 \pm 20\,ml\,min^{-1}\,100\,g^{-1}$ to $42 \pm 11\,ml\,min^{-1}\,100\,g^{-1}$ before administration of bisoprolol (Figure 3). The subendocardium was more severely affected than the subepicardium (−81% vs −50%, respectively). Administration of $50\,\mu g\,kg^{-1}$ bisoprolol improved the perfusion of the ischaemic segment by $21 \pm 10\,ml\,min^{-1}\,100\,g^{-1}$ ($P < 0.05$). This improvement was limited, however, to the subendocardial layers as perfusion of the subepicardial layers was not significantly modified (Figure 3). With the higher doses of bisoprolol, there were only minor additional changes. It has to be noted, however, that there was no indication of further deterioration in the ischaemic segment; this is in contrast to that in the control group at 60 min of flow reduction.

Since perfusion of the non-ischaemic myocardium decreased slightly after administration of bisoprolol, in contrast with the control group, the normalized flow ratio of the ischaemic and non-ischaemic myocardium was calculated in order to gain a better insight into the flow deficit of the ischaemic segment after bisoprolol administration. Table 1 clearly demonstrates that bisoprolol distinctly levelled out the perfusion differences between epicardium and endocardium while these remained unchanged in the control group.

Table 1 Flow deficit in ischaemic myocardium, expressed as the ratio of the normalized flows to the ischaemic and non-ischaemic myocardium, in untreated (U) and bisoprolol-treated (B) pigs before (baseline) and after partial stenosis of LADCA

			Time after ischaemia (min)			
		Baseline	*15*	*30*	*45*	*60*
			Dose ($\mu g\,kg^{-1}$)			
	U		0 ($n = 9$)	0 ($n = 9$)	0 ($n = 7$)	0 ($n = 4$)
	B		0 ($n = 8$)	50 ($n = 8$)	250 ($n = 7$)	750 ($n = 7$)
Transmural	U	0.98 ± 0.02	0.46 ± 0.10*	0.51 ± 0.12*	0.46 ± 0.12*	0.27 ± 0.10*
	B	0.96 ± 0.03	0.34 ± 0.08*	0.64 ± 0.11*†	0.71 ± 0.11*†	0.74 ± 0.11*†
Subendocardial	U	0.99 ± 0.05	0.33 ± 0.08*	0.39 ± 0.12*	0.39 ± 0.13*	0.14 ± 0.07*
	B	0.96 ± 0.05	0.19 ± 0.07*	0.52 ± 0.16*†	0.60 ± 0.13*†	0.66 ± 0.13*†
Subepicardial	U	0.97 ± 0.04	0.62 ± 0.12*	0.60 ± 0.12*	0.59 ± 0.14*	0.38 ± 0.12*
	B	1.03 ± 0.03	0.49 ± 0.09*	0.72 ± 0.09*†	0.78 ± 0.09*†	0.80 ± 0.10*†

LADCA, Left anterior descending coronary artery. * $P < 0.05$ vs baseline; † $P < 0.05$ vs changes in untreated group. Data have been presented as means ± s.e.mean.

Regional myocardial function

The decrease in perfusion led to decreases in end-diastolic and, in particular, in end-systolic wall thickness. Consequently, absolute systolic wall thickening (EST − EDT) decreased from 3.5 ± 0.2 mm to 0.8 ± 0.2 mm and percentage systolic wall thickening (SWT) fell from 36 ± 3% to 10 ± 3% ($P < 0.05$) after the first 15 min of flow reduction in the untreated animals (Figure 4). There were no additional changes during the remainder of the experiment.

In animals that were later treated with bisoprolol the reduction in LADCA blood flow also caused an immediate decrease in absolute systolic wall thickening from 4.6 ± 0.2 mm to 0.8 ± 0.3 mm and in

Table 2 Systemic haemodynamics in untreated (U) and bisoprolol-treated pigs (B) before (baseline) and after partial stenosis of LADCA

			Time after ischaemia (min)			
		Baseline	*15*	*30*	*45*	*60*
			Dose ($\mu g\,kg^{-1}$)			
	U		0 ($n = 9$)	0 ($n = 9$)	0 ($n = 7$)	0 ($n = 4$)
	B		0 ($n = 8$)	50 ($n = 8$)	250 ($n = 7$)	750 ($n = 7$)
CO ($l\,min^{-1}$)	U	2.23 ± 0.16	1.97 ± 0.18	2.06 ± 0.17	1.94 ± 0.23	1.83 ± 0.03
	B	2.50 ± 0.34	2.04 ± 0.16*	1.84 ± 0.17*	1.84 ± 0.12*	1.67 ± 0.10*†
HR (beats min^{-1})	U	102 ± 4	123 ± 9*	127 ± 9*	116 ± 7	115 ± 10
	B	111 ± 10	113 ± 12	94 ± 8†	91 ± 7*†	84 ± 6*†
SV (ml)	U	22 ± 2	16 ± 1*	16 ± 1*	17 ± 1*	16 ± 1*
	B	22 ± 1	19 ± 1*	20 ± 1	21 ± 1	20 ± 1
MAP (mmHg)	U	91 ± 5	78 ± 6*	74 ± 6*	66 ± 5*	60 ± 2*
	B	79 ± 5	64 ± 3*	61 ± 3	59 ± 3*	57 ± 3*
LV$dP/dt_{max}$ ($mmHg\,s^{-1}$)	U	2360 ± 180	2070 ± 260*	2020 ± 280	1810 ± 280*	1800 ± 160*
	B	2640 ± 440	1860 ± 380*	1230 ± 190*†	1240 ± 210*†	1070 ± 150*†
LVEDP (mmHg)	U	9 ± 1	13 ± 2*	12 ± 2	11 ± 2	14 ± 3
	B	9 ± 1	15 ± 1*	15 ± 2*	15 ± 3*	15 ± 3*

LADCA, Left anterior descending coronary artery; CO, cardiac output; HR, heart rate; SV, stroke volume; MAP, mean arterial blood pressure; LV$dP/dt_{max}$, maximum rate of rise in left ventricular pressure; LVEDP, left ventricular end diastolic pressure. * $P < 0.05$ vs baseline; † $P < 0.05$ vs changes in untreated group. Data have been presented as means ± s.e.mean.

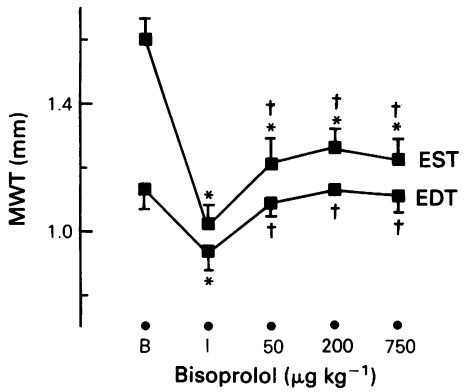

Figure 5 Effect of bisoprolol on myocardial wall thickness of the ischaemic segment in 8 open-chest anaesthetized pigs with a graded stenosis in the left anterior descending coronary artery. The three doses of bisoprolol were administered over a period of 2 min at 15 min intervals. The first dose was given after 15 min of ischaemia (I). EDT and EST are the wall thickness at end-diastole and end-systole, respectively. * $P < 0.05$ vs baseline (B). † $P < 0.05$ vs I. Data have been presented as means with vertical lines showing s.e.mean.

percentage systolic wall thickening from 41 ± 2% to 9 ± 4% ($P < 0.05$) (Figure 5). Although bisoprolol increased both EDT and EST, it failed to improve absolute as well as percentage systolic wall thickening.

As stated before (see experimental protocol) the effects of bisoprolol (cumulative doses: 16, 80, 336 and 1024 $\mu g\,kg^{-1}$) on the LADCA perfused myocardium were evaluated in a separate group of 10 animals with an intact coronary circulation. Systolic wall thickening values (%) before and after the four doses of bisoprolol were 48 ± 2, 52 ± 4, 45 ± 4, 44 ± 3 and 34 ± 3, respectively. Therefore, bisoprolol only slightly reduced systolic wall thickening and that too after the highest dose.

Systemic haemodynamics

The reduction in LADCA blood flow caused a number of immediate changes in systemic haemodynamic variables: decreases in cardiac output (10%), stroke volume (25%), mean arterial blood pressure (15%) and LV$dP/dt_{max}$ (12%), while both heart rate (20%) and left ventricular filling pressure (45%) increased (Table 2). These changes lasted for the entire 60 min period of observation in the untreated group. In the group treated with bisoprolol similar effects were noticed except that heart rate decreased (instead of a moderate increase) and LV$dP/dt_{max}$ decreased more prominently (Table 2).

Discussion

The reduction in LADCA flow produced by the stenosis led to immediate changes in regional myocardial blood flow and wall motion and in systemic haemodynamics. These effects remained stable during the first 45 min of ischaemia, but between 45 and 60 min, the untreated preparations tended to show signs of further deterioration (see Figure 2). However, because of the low number of observations at the end of the 60 min period (4 animals died because of ventricular fibrillation), statistical significance was difficult to ascertain. Therefore, it appears that systemic and regional haemodynamic variables and systolic myocardial wall thickening and perfusion are suitable for evaluating the effects of pharmacological agents during this period.

The present study demonstrated that bisoprolol improves perfusion of the ischaemic myocardium in anaesthetized open-chest pigs. This is, however, not surprising since a large number of other β-adrenoceptor antagonists also possess this property (Vatner *et al.*, 1977; Tomoike *et al.*, 1978; Berdeaux *et al.*, 1979; Buck *et al.*, 1979; 1981; Gross *et al.*, 1979; Saxena, 1983; Verdouw *et al.*, 1986). On the other hand, negative results have also been obtained in earlier studies (Becker *et al.*, 1975; Kloner *et al.*, 1976; Berdeaux *et al.*, 1977). It is interesting that bisoprolol increases blood flow to the ischaemic myocardial segment, in particular to the subendocardium, even at doses which only moderately inhibit isoprenaline-induced increases in heart rate and contractility (Duncker *et al.*, 1987; present results). Using other β-adrenoceptor antagonists or specific bradycardiac drugs, several investigators have shown that the endocardium, especially, benefits from a prolongation of the duration of diastole (Berdeaux *et al.*, 1979; Gross *et al.*, 1979; Buck *et al.*, 1981; Schamhardt *et al.*, 1981b; Saxena, 1983; Daemmgen *et al.*, 1985; Verdouw *et al.*, 1986; Guth *et al.*, 1987).

In contrast with the increase in blood flow, there was no improvement in segmental myocardial function. Several arguments can be put forward to explain this apparent discrepancy. Firstly, the improvement in flow may be too small to guarantee an improvement in contractile function. In the same preparation, we have shown that reduction of LADCA flow to 50% of baseline values causes an almost complete loss of systolic wall thickening (Verdouw *et al.*, 1980). Based on the data presented in Figure 3, some slight improvement might have been expected. Secondly, most β-adrenoceptor antagonists, and bisoprolol is no exception, exhibit negative inotropic properties which could, but do not necessarily, decrease systolic wall thickening of the normally perfused myocardium. We therefore

studied systolic wall thickening of the myocardium perfused by the LADCA in an additional group of animals with an intact coronary circulation. Since, in animals with an intact coronary circulation, doses lower than $1000\,\mu g\,kg^{-1}$ did not affect systolic wall thickening, it is unlikely that the drug-induced negative inotropic effect prevented an improvement in regional function. Moreover, a number of β-adrenoceptor antagonists have been shown to improve the regional contractile function of ischaemic myocardium despite their negative inotropic action on the normal myocardium (Tomoike *et al.*, 1978; Gross *et al.*, 1979, Buck *et al.*, 1981). A third reason may be the duration of ischaemia preceding the administration of bisoprolol: numerous studies in dogs and pigs have shown that even after complete restoration of blood flow subsequent to partial or complete occlusion of a coronary artery lasting up to 20 min, recovery of contractile function takes place only after a considerable delay (Heyndrickx *et al.*, 1975; Ramanathan *et al.*, 1978; van der Giessen *et al.*, 1986). During bisoprolol treatment recovery of perfusion is still incomplete, therefore, it is not surprising that regional contractile function of the ischaemic segment did not improve. It is of interest, however, that although regional contractile function did not concomitantly return to normal, both end-diastolic and end-systolic wall thickness improved slightly. This implies that 'bulging' of the ischaemic myocardium became less, which may be a first step towards the return of contractile function.

In the present experiments using a model with partially restricted coronary blood flow, the incidence of ventricular fibrillation was considerably lower in the bisoprolol-treated than in the untreated animals, despite a more severe reduction in blood flow during the first 15 min before drug administration in the bisoprolol group. In recent experiments using repeated, complete coronary occlusions, bisoprolol was shown to be ineffective in preventing ventricular fibrillation in pigs (Verdouw *et al.*, 1987b). This is not surprising because most β-adrenoceptor antagonists, including propranolol, pindolol and sotalol, fail to prevent ventricular fibrillation after ligation of a coronary artery in this species (Frank *et al.*, 1978; Bergey *et al.*, 1984; Benfey *et al.*, 1984; Muller *et al.*, 1986; Verdouw & Hartog, 1986). Metoprolol has been shown to be effective against ventricular fibrillation (Muller *et al.*, 1986), but the dose used ($20\,mg\,kg^{-1}$) is not clinically relevant as it far exceeded that required to obtain adequate β-adrenoceptor blockade, and it is therefore doubtful that the antifibrillatory action was related to β-adrenoceptor blockade. With complete vascular occlusion, the differences in perfusion of the normal and the acutely ischaemic myocardium, and in perfusion of the different layers of the ischaemic segment, are not affected by pharmacological agents since the porcine heart has only few native collaterals (Schaper & Wüsten, 1979; Millard, 1980. However, in the conditions used in the present study, where only a partially obstructed vascular supply was used, the flow differences between the various myocardial areas were attenuated (Table 1). This might have contributed to the reduction in the incidence of fatal ventricular arrhythmias in bisoprolol-treated animals.

In conclusion, bisoprolol appears to produce beneficial effects in an animal model of acute myocardial ischaemia. The finding that the drug improves the perfusion of ischaemic myocardium and prevents ventricular arrhythmias at doses that have relatively moderate effects on β-adrenoceptors, may be clinically advantageous.

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