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EFFECTS OF ATENOLOL ON REGIONAL MYOCARDIAL BLOOD FLOW AND ST SEGMENT ELEVATION IN THE CANINE MYOCARDIUM

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1 The effects of atenolol on regional myocardial blood flow (RMBF) and on ST segment elevation were studied both in normal and ischaemic regions of the myocardium in dogs. Some of the experiments were performed with cardiac pacing or after bilateral stellectomy.

2 In the absence of cardiac pacing, atenolol (1 mg/kg, i.v.) induced a marked reduction in heart rate and contractile force and a decrease in RMBF, which was of the same magnitude in normal and ischaemic areas. There was no modification in the endo/epi flow ratio. ST segment elevation in ischaemic areas was significantly reduced.

3 Bilateral stellectomy induced similar effects to those of atenolol. Atenolol after bilateral stellectomy exhibited no additional effects.

4 In dogs submitted to cardiac pacing, atenolol no longer decreased RMBF and ST segment elevation.

5 These results demonstrate that with atenolol, there is no correlation between bradycardia and the endo/epi flow ratio while there is one between bradycardia and reduction in ST segment elevation.

Introduction

In recent years, several authors have shown, by assessing the degree and extent of ischaemic injury from epicardial electrocardiograms and/or creatinine phosphokinase release, that β-adrenoceptor blocking agents are capable of reducing 'infarct size' in dogs (Maroko, Kjekshus, Sobel, Watanabe, Covell, Ross & Braunwald, 1971) and in man (Pelides, Reid, Thomas & Shillingford, 1972). These beneficial effects of β-blockade are thought to be related to a more favourable balance between myocardial oxygen supply and demand in the ischaemic heart. However, other factors may contribute. For instance, Becker, Fortuin & Pitt (1971) and Gross & Winbury (1973) have shown that β-adrenoceptor antagonists can alter the distribution of regional myocardial blood flow (RMBF) and induce its redistribution from the epicardium to the endocardium in the normal and ischaemic heart. Moreover, Marshall & Parratt (1976) have recently shown in the canine myocardium that, while propranolol reduces RMBF in both ischaemic and non-ischaemic regions, practolol induces a more favourable distribution of coronary blood flow since it decreases RMBF only in the non-ischaemic areas; this beneficial effect has been partly attributed to the drug's β_1-selectivity.

In the present work we have investigated these two redistribution possibilities by studying the effects of atenolol, another selective β_1-adrenoceptor antagonist (Barrett, Carter, Fitzgerald, Hull & Le Count, 1973; Harry, Knapp & Linden, 1973) on RMBF assessed by the radioactive microsphere mapping technique (Becker, Ferreira & Thomas, 1973). This study has been performed in an experimental model of canine myocardial ischaemia (Berdeaux, Coutte, Giudicelli & Boissier, 1976a) with simultaneous measurement of ST segment changes. A preliminary account of this work has been published (Berdeaux, Boissier & Giudicelli, 1976).

Methods

Twenty adult mongrel dogs of either sex, weighing 18–29 kg were anaesthetized with sodium pentobarbitone (35 mg/kg, i.v.). Artificial ventilation was performed by means of a Bird Mark 7 respirator using room air through a cuffed endotracheal tube. The chest was opened by means of a left thoractomy and the heart cradled in the open pericardium.

The first diagonal branch of the left anterior

descending coronary artery was dissected free from the adjacent tissues and intermittently occluded with an intracranial arterial clamp. A strain gauge arch was sutured to the epicardial surface of the left ventricle in the area perfused by the circumflex coronary artery (non-ischaemic area) for the measurement of developed force. Silastic catheters were placed in the two femoral arteries and in the left atrial appendage. Blood pressure was measured with a Hewlett-Packard 267 AC transducer and displayed on a Hewlett-Packard 7700 multichannel recorder. The electrocardiogram (lead II) was monitored on a Visocardiette 560 Hewlett-Packard.

The experimental protocol was similar to that previously described (Berdeaux *et al.*, 1976a). In all dogs, a 12 min control occlusion was carried out with a microsphere injection 7 min after clamping and epicardial ST mapping just before and 2, 5 and 10 min after starting the occlusion. A second coronary artery occlusion was then performed usually 55 min later, i.e. at least 10 min after complete recovery from the first occlusion. This second occlusion, with the same ST mapping and blood flow measurements, gave similar results to the first (Berdeaux *et al.*, 1976a).

Four groups of five dogs each were studied. In group I, the animals were given atenolol (1 mg/kg, i.v.) 45 min after the first and 10 min before the second occlusion. In groups II and III, a bilateral stellectomy and a T1–T4 sympathetic chain removal was performed 45 min after the first and 30 min before the second occlusion. In addition in group III, atenolol (1mg/kg, i.v.) was given 10 min before the second occlusion. Group IV was identical to group I but atrial pacing (rectangular pulses, 2 V, 2 ms) at the pre-occlusion heart rate was started immediately before the atenolol injection and continued until the end of the second occlusion.

Myocardial blood flow was measured with radioactive tracer microspheres. The carbonized microspheres used were 15 ± 5 μm in diameter and labelled with the gamma emitting nuclides ^{141}Ce or ^{85}Sr (3 M Company). They were obtained as 1 mCi of nuclide suspended in 10 ml of 10% dextran with 1 drop of benzalkonium (0.5%). Aggregation of the microspheres before injection was prevented by vigorous stirring with a Teflon covered magnet and by applying ultrasound for 10 min (Ultrasonic model NSU 144). Approximately two million beads were injected into the left atrium for each blood flow determination. During each measurement a reference blood sample was collected from the femoral artery. Collection of the reference sample was started simultaneously with the beginning of microsphere injection and continued for 90 s at a rate of 20 ml/min, using a withdrawal pump (Sage Instruments, model 351).

At the end of the experiments, the animals were killed by an overdose of potassium chloride. The heart was then removed and fixed in buffered 4% formaldehyde for 2 days. Maps of the left ventricular myocardial blood flow were made according to the method of Becker *et al.* (1973). The free wall was cut into 40–50 pieces of approximately 0.5–1.5 g and the location of each piece of the free wall was recorded. All samples were divided into endocardial and epicardial halves, weighed and placed in vials for counting. The myocardial and blood samples were counted under similar geometry in a Nuclear Chicago Gamma Spectrometer at window settings that corresponded to the peak energies emitted by each radioactive nuclide (145 keV for ^{141}Ce and 514 keV for ^{85}Sr). The activities recorded in each energy window and the corresponding sample weights were then entered into a digital computer (Hewlett-Packard, Model 9821 A) programmed to correct activity recorded in each window for contaminant activity contributed by the associated nuclides and for background activity and to compute the corrected counts. Details of the design have been published by Schaper, Lewi, Flameng & Gipjen (1973). Knowing the rate of withdrawal of the reference sample ($\dot{Q}r$) and its radioactivity (Cr), we used myocardial activity (Cm) to compute myocardial blood flow ($\dot{Q}m$) as: $\dot{Q}m = \dot{Q}r \times Cm/Cr$. The flow for each sample was then calculated in ml min^{-1} g^{-1}, maps of blood flow were drawn and endo/epi ratios were determined for each occlusion. According to Becker, Ferreira & Thomas (1975), and Berdeaux *et al.* (1976a), an 'ischaemic region' is defined as one in which flow is less than 50% of that of the non-ischaemic posterior wall. Pooled samples from this ischaemic region always had more than 400 spheres. Mean flows in endocardial and epicardial regions of the left ventricular wall and endo/epi ratios for each region were determined and compared before and after treatment, each dog serving as its own control.

Using the method of Maroko *et al.* (1971), tissue injury was assessed by ST segment elevation in epicardial electrocardiograms obtained from 12–15 sites on the anterior surface of the left ventricle. Some of the sites were within, and some outside, the ischaemic zone. Areas adjacent to vessel bifurcations were chosen for easy relocation. The exploring cotton wick epicardial electrode was soaked in saline and attached to lead V of a standard electrocardiograph. Elevation or depression of the ST segment was recorded at a speed of 25 mm/s (range 1 mV/mm) on the Hewlett-Packard 7700 recorder. Sites with ST elevation in excess of 2 mV before coronary occlusion were excluded from the study and mean ST segment elevation ($\overline{ST}$) was used as an index of the severity of ischaemia as described by Pelides *et al.* (1972).

The racemic form of atenolol hydrochloride was dissolved in 0.9% w/v NaCl solution (saline) and injected via a catheter in the saphenous vein. All doses refer to the salt. All values quoted in the text are means $\pm$ s.e. mean. Statistical analysis of the results was performed using Student's *t* test for paired data.

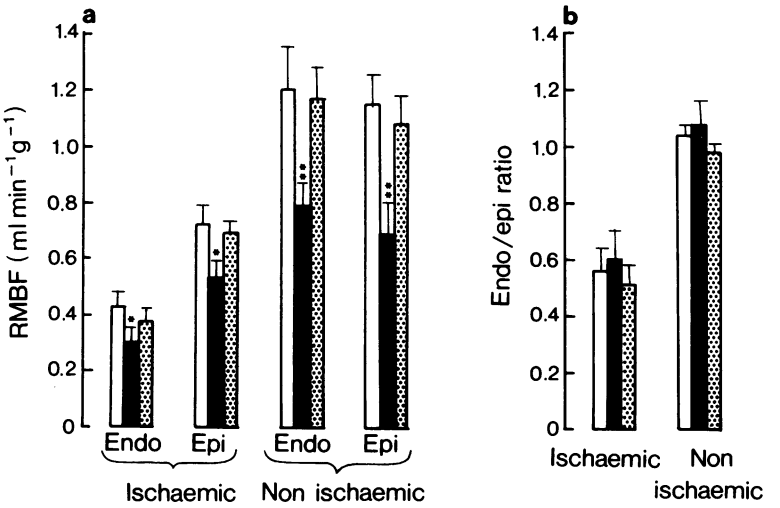

Figure 1 Regional myocardial blood flow (RMBF) (a) in the endocardium and epicardium and (b) endo/epi ratio values measured in non-ischaemic and ischaemic areas of the canine myocardium, during a control coronary occlusion (open columns) and during occlusion performed after atenolol (1 mg/kg, i.v.) without (solid columns) and with atrial pacing (stippled columns). Values are means; vertical lines show s.e. mean. Significantly different from control occlusion values, $*P<0.05$; $**P<0.01$.

Results

Group I (Atenolol)

Table 1 shows the haemodynamic effects induced by atenolol, 1 mg/kg. There were significant decreases in heart rate and contractile force and a slight, but not significant, reduction in blood pressure.

Figure 1 compares the effects of the second occlusion, performed after atenolol, with those of the first occlusion performed before administration of the drug. RMBF values were significantly decreased and to the same extent both in ischaemic ($-32.5 \pm 6.2\%$) and non-ischaemic areas ($-37.6 \pm 7.5\%$). This decrease was similar in both epicardial and endocardial regions and this resulted in an unchanged endo/epi flow ratio in both ischaemic and non-ischaemic areas. Atenolol induced a significant reduction in the ST segment elevation during the second occlusion as compared to the first (Figure 2).

Table 1 The effect of atenolol, bilateral stellectomy and atrial pacing on heart rate, contractile force and systemic arterial pressure

	Heart rate (beats/min)		*Contractile force (arbitrary units)*		*Systolic blood pressure (mmHg)*		*Diastolic blood pressure (mmHg)*	
Treatment	*Pre*	*Post*	*Pre*	*Post*	*Pre*	*Post*	*Pre*	*Post*
Atenolol (1 mg/kg) $n=6$	174.1 ±1.5	126.2‡ ±3.1	14.3 ±0.3	12.3* ±0.3	146.2 ±5.5	142.5 ±6.6	97.5 ±2.5	96.5 ±3.2
Bilateral stellectomy $n=5$	164.1 ±1.7	110.0‡ ±6.1	14.5 ±0.8	10.6† ±0.4	161.2 ±5.9	135.5† ±5.4	103.2 ±5.2	88.7 ±8.7
Atenolol (1 mg/kg) after bilateral stellectomy $n=5$	154.1 ±1.7	107.0‡ ±7.2	14.5 ±0.8	9.7† ±0.8	160.2 ±5.2	133.7† ±8.6	100.2 ±6.4	89.5 ±8.8
Atenolol (1 mg/kg) during atrial pacing $n=5$	153.0 ±1.3	151.0 ±7.1	16.0 ±3.3	13.8 ±3.6	149.0 ±8.4	138.0 ±6.3	86.0 ±6.6	79.0 ±6.1

Values are mean ± s.e. mean.
Significantly different from pre-treatment value $*P<0.05$; $†P<0.01$; $‡P<0.001$.

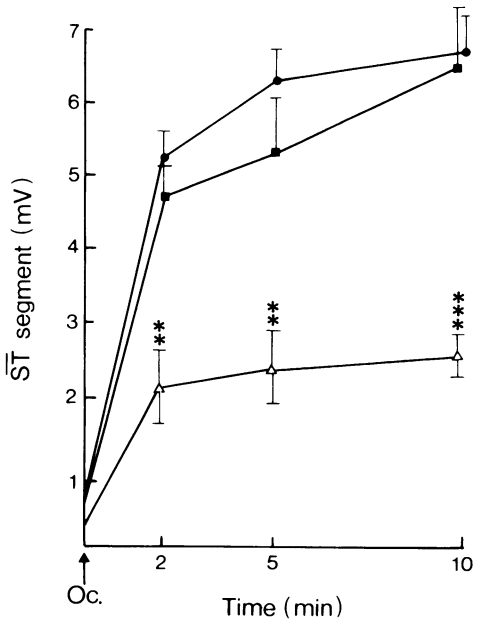

Figure 2 Mean ST segment elevation ($\overline{ST}$) measured 2, 5 and 10 min after left anterior descending coronary artery ligation during control occlusion (●) and during occlusion performed after atenolol (1 mg/kg, i.v.) without (△) and with atrial pacing (■). Vertical lines show s.e. mean. Significantly different from control occlusion values, $^{**}P < 0.01$; $^{***}P < 0.001$.

Group II (Bilateral stellectomy)

Table 1 shows the haemodynamic effects induced by bilateral stellectomy. There were significant decreases in heart rate, contractile force and systolic blood pressure.

Figure 3 compares the effects of the second occlusion, performed after bilateral stellectomy, with those of the first occlusion performed before stellectomy. RMBF values were again significantly decreased, and to the same extent, both in ischaemic ($-31.9 \pm 6.4\%$) and non-ischaemic areas ($-36.2 \pm 8.7\%$). As in the group I dogs, this decrease was again similar in both epicardial and endocardial regions and the endo/epi flow ratio was thus not changed. After bilateral stellectomy, there was a significant reduction in the $\overline{ST}$ segment elevation during the second occlusion as compared to the first (Figure 4). This reduction was of a similar magnitude to that observed in the group I dogs.

Group III (Bilateral stellectomy and atenolol)

Table 1 summarizes the results obtained when atenolol was administered 10 min after bilateral stellectomy. Atenolol did not further reduce either heart rate or contractile force.

Figure 3 compares the effects of the second occlusion performed after bilateral stellectomy and atenolol to those of the first (control) occlusion. There were no differences between the effects observed in groups II and III: reduction in RMBF values was

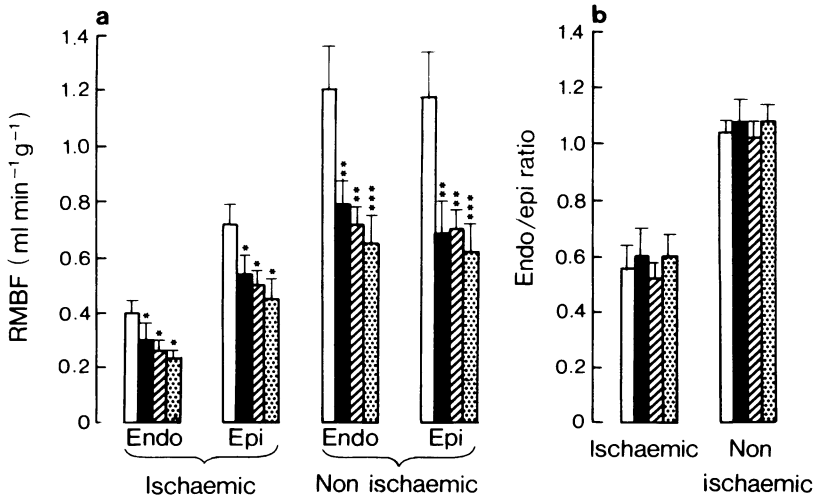

Figure 3 Regional myocardial blood flow (RMBF) (a) in the endocardium and epicardium and (b) endo/epi ratio values measured in non-ischaemic and ischaemic areas of the canine myodardium, during a control coronary occlusion (open columns) and during occlusions performed either after bilateral stellectomy (hatched columns), after atenolol (1 mg/kg, i.v.) alone (solid columns), or after a combination of bilateral stellectomy and atenolol (stippled columns). Values are means; vertical lines show s.e. mean. Significantly different from control occlusion values, $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$.

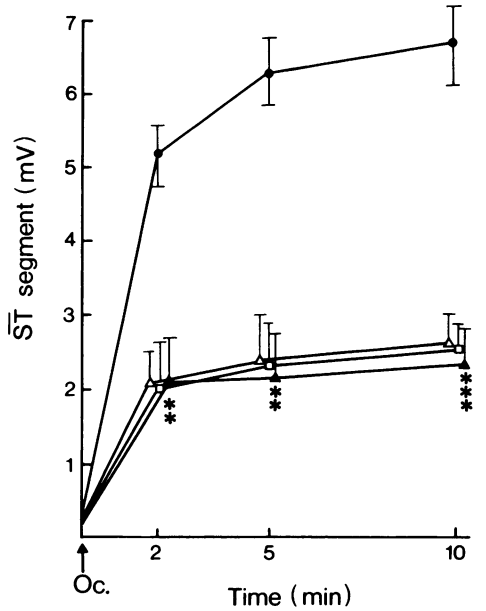

Figure 4 Mean ST segment elevation ($\overline{ST}$) measured 2, 5 and 10 min after a control occlusion of the left anterior descending coronary artery (●) and during an occlusion performed after bilateral stellectomy (□), after atenolol (1 mg/kg, i.v.) (△) or after a combination of bilateral stellectomy and atenolol (▲). Significantly different from control occlusion values, $^{**}P<0.01$; $^{***}P<0.001$.

observed in both ischaemic and non-ischaemic areas and was homogeneous, i.e. endo/epi flow ratios remained unchanged. The reduction in $\overline{ST}$ segment elevation during the second occlusion as compared to the first was similar in groups III and II (Figure 4).

Group IV (Atrial pacing and atenolol)

Table 1 shows that the haemodynamic changes induced by atenolol during atrial pacing were not significant and almost negligible. During atrial pacing, atenolol no longer reduced RMBF either in the ischaemic or non-ischaemic areas and the endo/epi ratio remained unchanged (Figure 1). The reduction in $\overline{ST}$ segment elevation observed in group I was abolished when atenolol was given during atrial pacing (Figure 2).

Discussion

Our results show that atenolol reduces regional myocardial blood flow both in ischaemic and non-ischaemic regions and that it does this without inducing any change in the endo/epicardial distribution of coronary flow. The reduction in regional flow is probably closely and directly related to the atenolol-induced decrease in myocardial oxygen consumption. It is logical then that the same phenomenon should be observed following bilateral stellectomy. This suggests that the decrease in contractile force, and particularly in heart rate, induced by atenolol and bilateral stellectomy is of greater relevance in reducing oxygen consumption than the drop in blood pressure observed after bilateral stellectomy alone. It is noteworthy that when atenolol-induced bradycardia was abolished by pacing, all changes in regional flow were suppressed. The fact that atenolol exerted no additional effects after bilateral stellectomy suggests that this drug reduces regional blood flow only by decreasing heart rate and contractile force (i.e. a reduction in oxygen consumption resulting from cardiac β_1-adrenoceptor blockade).

In non-ischaemic regions, adaptation of myocardial flow is accounted for by the phenomenon of metabolic autoregulation. In ischaemic regions, however, the reduction in local dilatory reserve impairs autoregulation capacity, and myocardial blood flow under these conditions is no longer dependent only on oxygen consumption (James, 1970). However, although at the centre of the lesion the capacity for dilatation is almost nil, numerous investigations have shown that the ischaemic region is in fact very heterogeneous with increasing flow gradients and autoregulation capacity from the centre to the periphery of the infarcting area (Becker *et al.*, 1973; Lubbe, Peisach, Pretorius, Bruyneel & Opie, 1974; Berdeaux *et al.*, 1976). The fact that our definition of the ischaemic region encompasses these various compartments accounts for the fact that we observed a reduction in regional flow under the influence of atenolol and bilateral stellectomy both in non-ischaemic regions and at the periphery of ischaemic areas. This reduction was of the same order of magnitude in these two regions. These results, which are consistent with those obtained with propranolol (Marshall & Parratt, 1976), contrast with the effects of practolol which does not reduce total regional blood flow in ischaemic regions. Marshall & Parratt (1976) have suggested two main reasons to account for this difference between propranolol and practolol. Firstly, only propranolol blocks coronary β_2-adrenoceptors (Parratt & Wadsworth, 1970; Gross & Feigl, 1975), and this alters the balance in favour of α-vasoconstrictor receptors, hence reducing flow. Practolol, which does not block β_2-receptors, does not modify the α–β_2 balance in the coronary arteries, thus leaving coronary flow unchanged. Secondly, practolol increases to a greater extent than propranolol, the diastolic effective perfusion period for the endocardium. Atenolol is a selective β_1-adrenoceptor inhibitor and therefore should not, like practolol, interfere with the α–β_2 balance in the coronary

arteries, and should thus leave myocardial flow unchanged. However, in practice, it reduces it. Two main reasons may explain this phenomenon. The first is that atenolol may lengthen to a lesser degree than practolol the diastolic effective perfusion period of the endocardium, a parameter we have not investigated. The second is that another factor is involved in the favourable effect of practolol, this factor possibly being its intrinsic β stimulant effect (Dunlop & Shanks, 1968), a property of which atenolol and propranolol are devoid. This could exert a beneficial effect at the periphery of the ischaemic zone and thus limit the drop in myocardial flow.

In our experience, neither atenolol nor bilateral stellectomy redistributed blood flow between the epicardial and endocardial layers and the endo/epi ratio remained unchanged both in ischaemic and non-ischaemic regions. This distinguishes atenolol from propranolol since many investigators have reported a redistribution phenomenon with the latter drug (Becker *et al.*, 1971; Gross & Winbury, 1973; Becker *et al.*, 1975; Warltier, Gross & Hardman, 1976) although this has not been confirmed by other workers (Kloner, Reimer &Jennings, 1976). These results tend to demonstrate that there is no correlation between bradycardia and an increase in the coronary endo/epicardial flow ratio. Indeed, although bradycardia and redistribution are observed concomitantly with propranolol and other substances such as verapamil (Berdeaux, Coutte, Giudicelli & Boissier, 1976b), they are not seen after administration of atenolol or following bilateral stellectomy. In contrast, our experience has shown that there is a close relationship between bradycardia and reduction in ST segment elevation during coronary occlusion. Either atenolol or bilateral stellectomy, or a combination of both, simultaneously reduced heart rate and the degree of myocardial ischaemia. It would appear that this phenomenon may be applied more broadly to other substances that decrease heart rate, such as propranolol (Maroko *et al.*, 1971; Becker *et al.*, 1975), practolol (Libby, Maroko, Covell, Malloch, Ross & Braunwald, 1973; Marshall & Parratt, 1976) and verapamil (Berdeaux *et al.*, 1976b).

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