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SPECIAL REPORT

Local intracoronary infusions of bradykinin profoundly reduce the severity of ischaemia-induced arrhythmias in anaesthetized dogs

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Bradykinin in a dose (25 ng kg^{-1} min^{-1}) which did not alter coronary flow, or saline, were infused into a small branch of the left anterior descending coronary artery in dogs anaesthetized with chloralose and urethane, for 10 min prior to coronary artery occlusion and throughout the 25 min occlusion period. The degree of inhomogeneity of conduction and epicardial ST-segment changes were measured in the ischaemic zone with a composite electrode. In control dogs, coronary artery occlusion led to severe arrhythmias with an incidence of ventricular fibrillation of 47% and tachycardia of 80% and with a mean of 528 ± 140 ventricular premature beats. In marked contrast, those dogs administered bradykinin had no ventricular fibrillation or tachycardia and the number of premature beats was significantly less (53 ± 19). ST-segment changes were also much less in these dogs. These results raise the possibility that bradykinin might contribute to the protective effects of preconditioning and acts as an 'endogenous myocardial protective substance'.

Keywords: Bradykinin; myocardial ischaemia; arrhythmias; ventricular fibrillation; ST-segment changes; reperfusion; preconditioning

Introduction Brief periods of coronary artery occlusion reduce the severity of the ultrastructural changes (Murry *et al.*, 1986) and the ventricular arrhythmias (Vegh *et al.*, 1990) that occur when the same artery is later occluded for a longer period. This phenomenon is known as preconditioning. The mechanisms remain to be elucidated but one of several possibilities is that brief preconditioning periods of ischaemia, trigger the release from the heart of 'endogenous myocardial protective substances' which then 'precondition' the myocardium by protecting it against a later, more severe, ischaemic stress (Vegh *et al.*, 1991).

We have recently described, in cardiac and coronary vascular tissue, an acid-optimum enzyme capable of releasing kinins from kininogen under conditions of ischaemia (Zeitlin *et al.*, 1989). This raises the possibility that plasma kinins may be involved, as endogenous myocardial protective substances in preconditioning. However, although inhibition of breakdown by angiotensin converting enzyme (ACE) inhibitors results in cardiac protection during ischaemia (Linz *et al.*, 1990) we could find no report of the effects of bradykinin on the severity of the early arrhythmias that occur during the first minutes of coronary artery occlusion. In the present paper we describe the results, in anaesthetized dogs, of infusing a low concentration of bradykinin directly into a small branch of a coronary artery on arrhythmias, on the degree of inhomogeneity of conduction and on the ST-segment changes that result from a prolonged occlusion of that artery.

Methods Twenty-four adult mongrel dogs, with a mean weight of 23.8 ± 1.6 kg, were anaesthetized with a mixture of chloralose and urethane (60 and 200 mg kg^{-1} intravenously) and ventilated with room air by use of a Harvard respirator at a rate and stroke volume sufficient to maintain arterial blood gases and pH within normal limits (Vegh *et al.*, 1990). The temperature was maintained at 37 ± 0.5°C by means of a heating pad. A thoracotomy was performed at the fifth intercostal space and the left anterior descending (LAD) coronary artery dissected free just proximal to the main diagonal branch and a thread placed loosely around it. A smaller branch of the LAD proximal to this proposed site of occlusion was catheterized for the intracoronary infusion of bradykinin or saline. Epicardial ST-segment changes and the degree of inhomogeneity of conduction were measured in the area distal to the occlusion with the composite electrode described by Vegh *et al.* (1987). In some of the experiments coronary blood flow was measured on either the LAD or on the left circumflex coronary artery (or both) with an electromagnetic flow probe (Statham SP 2202; 2.0 mm) and/or a 2.4 mm Doppler flow probe (Triton Technology, San Diego, U.S.A.). Arterial and left ventricular (LV) pressures, and LV$dP/dt$ were measured with Statham P23Db pressure transducers and all parameters, together with a limb lead ECG, were recorded on an 8 channel Medicor R-81 Recorder. The area at risk was measured with patent blue V dye at the end of the experiment and expressed as a percentage of the free left ventricular wall. The mean value was 39.1 ± 1.7%. Ventricular arrhythmias during ischaemia and reperfusion were analysed statistically as previously described (Vegh *et al.*, 1990).

Fifteen of the 24 dogs acted as controls and 9 were infused with a dose of bradykinin (25 ng kg^{-1} min^{-1}) which, from previous experience (Lochner & Parratt, 1964) was at the limit of that required to increase coronary blood flow. The protocol was to infuse saline for 20 min at a rate of 0.075 ml min^{-1} and then bradykinin (or saline) for 10 min preceding coronary artery occlusion and then throughout the 25 min occlusion period. The myocardium was reperfused at the end of this period.

Results Neither the intracoronary infusion of saline nor bradykinin elicited significant haemodynamic changes (e.g. arterial pressure 141 ± 6 mmHg systolic and 96 ± 5 mmHg diastolic during the saline infusion and 144 ± 5/98 ± 4 mmHg after 10 min of bradykinin infusion; heart rate 152 ± 5 to 149 ± 4 beats min^{-1}; LVEDP 6.7 ± 0.8 to 6.2 ± 0.7 mmHg; LV$dP/dt_{max}$ 2355 ± 111 to 2381 ± 119 mmHg s^{-1}). Coronary blood flow was hardly affected by this dose of bradykinin (change less than 5 ml min^{-1} from control values of 50–55 ml min^{-1} diastolic and 10–15 ml min^{-1} systolic). To increase flow, significantly higher dose of bradykinin (0.33 and 1.24 μg kg^{-1} min^{-1}) or single bolus injections of acetylcholine (0.05 μg kg^{-1}) were required.

Ventricular ectopic activity was pronounced in the control dogs (Figure 1) with a high incidence (47%) of ventricular

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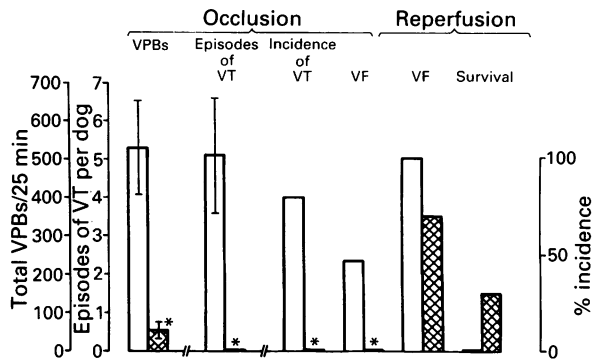

Figure 1 Ventricular arrhythmias during a 25 min occlusion of the left anterior descending coronary artery in anaesthetized dogs and following abrupt reperfusion at the end of that period. Bradykinin, infused into a small branch of the artery proximal to the occlusion in a dose of 25 ng kg^{-1} min^{-1} significantly reduced the incidence and the severity of these arrhythmias. Open columns are the controls ($n = 15$) and cross-hatched columns are from the bradykinin-treated animals ($n = 9$). * $P < 0.05$ (Mann-Whitney U test or Fisher's Exact Probability Test (for changes in the incidence of events)).

fibrillation (VF), a large number of ventricular premature beats (VPBs) in the survivors (528 ± 140) and several periods of ventricular tachycardia (VT; 5.1 ± 1.6 episodes per dog). In marked contrast, those dogs infused with bradykinin had little ventricular ectopic activity over the 25 min occlusion period (53 ± 19 VPBs, and there was no VT or VF in any of the dogs; Figure 1). Changes in the degree of inhomogeneity for the first 5 min of occlusion were similar (65 ± 5, 88 ± 10, 150 ± 17 and 154 ± 22 ms at 0, 1, 3 and 5 min of occlusion respectively versus 66 ± 3, 77 ± 5, 120 ± 13 and 154 ± 22 ms during the bradykinin infusion; NS). Changes in ST-segment elevation on occlusion were less marked during the bradykinin infusion (3.6 ± 0.9, 6.6 ± 1.2 and 7.9 ± 1.2 mV at 1, 3 and 5 min versus 1.7 ± 0.4 ($P < 0.05$), 3.9 ± 0.5 ($P < 0.05$) and 6.1 ± 0.9 (NS) at the same times during the bradykinin infusion).

Discussion A number of recent studies have shown that increased endogenous bradykinin levels are a major component of the beneficial effects of ACE inhibitors under conditions of experimental myocardial ischaemia since these drugs also inhibit kininase II. Perhaps the most conclusive evidence comes from a recent study (Martorana *et al.*, 1990) showing that cardioprotective effects of ACE inhibitors, such as an ability to reduce myocardial infarct size, are suppressed by bradykinin antagonists. Less attention has been given to the possibility that bradykinin, besides reducing ultrastructural damage resulting from ischaemia might also reduce the severity of ischaemia-induced arrhythmias. Tobe *et al.*, 1991 infused bradykinin, in a dose of 3.3 μg kg^{-1} min^{-1} by way of a Swan-Ganz catheter in the pulmonary artery, to anaesthetized pigs during balloon occlusion of a coronary artery (and reperfusion 45 min later). Two weeks later the dogs were re-anaesthetized and ventricular tachyarrhythmias were induced by use of programmed electrical stimulation. Electrical stability (assessed from the filtered QRS deflection using signal-averaging electrocardiography) was greater in those dogs that had been given bradykinin during the coronary artery occlusion-reperfusion period and it was less easy to induce ventricular tachycardia in these hearts. Myocardial creatine phosphokinase loss was also less in those animals administered bradykinin, signifying reduced myocardial damage.

Our present studies are concerned not with late ischaemic arrhythmias, but with those life-threatening arrhythmias that occur within minutes of coronary artery occlusion. These early ventricular arrhythmias were dramatically reduced by the local intra-arterial administration of bradykinin in a dose that hardly modified coronary blood flow. We have, as yet, no explanation for this profound modification of arrhythmia severity. Possibilities include prostanoid (prostacyclin?) and nitric oxide release, both of which contribute to the beneficial effects of preconditioning, (Vegh *et al.*, 1990; 1991), increased myocardial glucose uptake, which could decrease arrhythmia severity perhaps by reducing potassium loss (Bernier & Hearse, 1988) or a reduced severity of ischaemia (as indicated by the reduced ST-segment changes in the present study and by decreased myocardial lactate production in the studies of Linz *et al.*, 1990). Whatever the explanation, these results raise the possibility that bradykinin, released locally as a consequence of myocardial ischaemia, might participate in the protective effects of preconditioning.

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