*Br. J. Pharmacol.* (1990), **99**, 5–6 **SHORT COMMUNICATION** 

# Prevention of myocardial enzyme release by ranolazine in a primate model of ischaemia with reperfusion

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In control anaesthetized baboons subjected to 30 min occlusion of the left anterior descending coronary artery, followed by 5.5 h reperfusion, total plasma levels for creatine kinase (CK) and lactate dehydrogenase (LDH) were markedly elevated in a time-related manner. In a second group of baboons pretreated 10 min prior to ischaemia with ranolazine [(±)-N-(2,6-dimethyl-phenyl)-4[2-hydroxy-3-(2-methoxyphenoxy)propyl]-1-piperazine acetamide dihydrochloride; RS-43285-193] at 500 $\mu g\,kg^{-1}$ i.v., followed by continuous infusion of 50 $\mu g\,kg^{-1}\,min^{-1}$, neither enzyme was significantly elevated at any time point. Similarly, serum levels of the cardiospecific isoenzyme $CK_2$ were 8 fold greater in the controls than in the ranolazine-treated animals at 6 h. The results indicate that ranolazine pretreatment abolished cardiac enzyme release over a 5.5 h reperfusion period, indicating a potential protective effect.

**Introduction** The novel anti-anginal agent ranolazine [(±)-N-(2,6-dimethyl-phenyl)-4[2-hydroxy-3-(2-methoxyphenoxy)-propyl]-1-piperazine acetamide dihydrochloride] has been shown to be effective in inhibiting the biochemical and gross electrocardiographic consequences of transient myocardial ischaemia in the dog (Allely *et al.*, 1987; Allely & Alps, 1988) without causing direct haemodynamic effects. The present study was undertaken to investigate the ability of ranolazine to limit the biochemical consequences of a more severe sustained ischaemic insult in the subhuman primate.

**Methods** Fasted male baboons (*Papio hamadryas*, 6–11 kg) were anaesthetized with sodium pentobarbitone (8 $mg\,kg^{-1}$, i.v.) and respired with room air. Systemic blood pressure (BP) was monitored continuously from a femoral artery. The heart was exposed via a mid-sternal thoracotomy and a ligature was placed around the left anterior descending coronary artery (LAD) above the first diagonal branch. A 500 $\mu g\,kg^{-1}$ i.v. bolus of ranolazine (expressed in terms of free base) was administered in a saline vehicle to 4 animals and 10 min later the LAD was occluded. At this point a 6 h maintenance i.v. infusion of ranolazine (50 $\mu g\,kg^{-1}\,min^{-1}$) was begun. The LAD ligature was removed after 30 min to allow reperfusion. Femoral venous plasma and serum samples were taken to measure total creatine kinase (CK) and total lactate dehydrogenase (LDH) (Impact 400, Gilford reagents) and the cardiospecific isoenzyme $CK_2$ (Corning fluorometric electrophoresis). The enzyme levels were compared to those of 4 untreated control baboons by Fisher's randomisation test (Fisher, 1960).

**Results** In untreated control animals levels of total CK, LDH and $CK_2$ rose markedly after the first hour post-ischaemia (Figure 1a, b and c respectively). In the animals which were pretreated with ranolazine however, none of these enzymes was apparently elevated at any time point during the 6 h observation period. In terms of total CK and LDH all levels in the ranolazine group were significantly lower than in controls ($P < 0.05$). A marked overall fall in systolic (−45%) and diastolic (−50%) BP occurred in the control animals over 6 h, with an increase in heart rate (57%). In contrast, the BP reduction in the treated animals (−15% systolic, −35% diastolic BP) and accompanying tachycardia (12%) were much less.

**Discussion** In this study pre-ischaemia treatment with ranolazine, plus maintenance dosing, clearly inhibited the peripheral appearance of clinically-used (Galen *et al.*, 1975) enzymatic markers of myocardial damage. In control animals

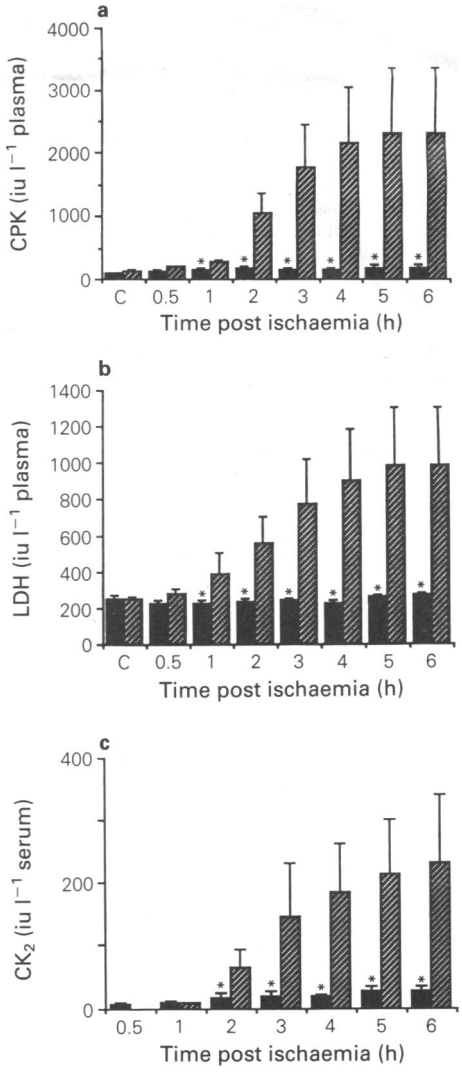


**Figure 1** Effect of ranolazine (solid columns) administered i.v. 10 min pre-ischaemia (500 $\mu g\,kg^{-1}$, followed by a continuous infusion of 50 $\mu g\,kg^{-1}\,min^{-1}$) in inhibiting cardiac enzyme release in 30 min ischaemic baboon hearts ($n = 4$) reperfused for 5.5 h. The results are compared to values for untreated control animals (hatched columns, $n = 4$). Panels (a), (b) and (c) show, respectively, plasma total levels of creatine phosphokinase (CPK), lactate dehydrogenase (LDH) and serum levels of the cardiospecific isoenzyme $CK_2$, initially detected at 0.5 h post-ischaemia. Columns represent group mean with s.e.mean shown by vertical bars. Values are significantly lower than control values at $^*P < 0.05$.

the appearance of these markers was rapid and pronounced. It has been suggested that the rapid appearance of such enzymes in plasma is not simply associated with reperfusion washout of previously unperfused regions of ischaemic myocardium but rather the result of accelerated damage resembling the reoxygenation-induced enzyme release from anoxic hearts (Hearse *et al.*, 1973; Ganote *et al.*, 1976).

With regard to the nature of the total CK composition it was evident from accompanying isoenzyme profiles that there was a much higher component of $CK_2$ (8 fold) than in ranolazine-treated animals at the 6 h time point. Care has to be taken in avoiding the potential inaccuracies involved in just relying on the total CK method which is invalidated in human patients subjected to recent surgery, especially if it involves the coronary arteries (Norris, 1979). Comparable correlations between estimated infarct size based on plasma CK activity and those related to myocardial $CK_2$ depletion have been observed in the baboon (Yasmineh *et al.*, 1977), in which species the organ distribution of $CK_2$ is similar to that in man. This enables improved estimates of infarct size to be made based on plasma $CK_2$ rather than on total CK activity, and this still holds true ($r > 0.9$) when the experimental protocol includes reperfusion of the myocardium (Manders *et al.*, 1975). Our other observations showed that the maintenance of general haemodynamic function during the time course of the study in the treated animals was superior to that of the controls.

To conclude, whilst at present we cannot rule out the possibility that we have not just delayed the appearance of these predictive markers of myocardial damage, the evidence is strongly suggestive of a potential beneficial effect for ranolazine, given prophylactically, in extending the time-window for therapeutic intervention where an acute myocardial infarction is judged to be imminent.

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