Renal ischemia preconditions myocardium: role of adenosine receptors and ATP-sensitive potassium channels

THERESA J. PELL, GARY F. BAXTER, DEREK M. YELLON, AND G. MICHAEL DREW

Renal ischemia preconditions myocardium: role of adenosine receptors and ATP-sensitive potassium channels. Am. J. Physiol. 275 (Heart Circ. Physiol. 44): H1542–H1547, 1998.—Brief renal ischemia–reperfusion is reported to precondition the myocardium; however, the underlying mechanisms are unknown. This phenomenon was, therefore, investigated using an in vivo rabbit model of acute myocardial infarction. Characterization of the mechanisms involved was performed using the nonselective adenosine receptor antagonist 8-(p-sulfophenyl)theophylline (8-SPT) and the ATP-sensitive potassium (KATP) channel blocker sodium 5-hydroxydecanoate (5-HD). Pentobarbital-anesthetized rabbits underwent a left thoracotomy and pericardiotomy. A laparotomy was then performed to expose the left renal artery. Animals were either preconditioned with a 10-min occlusion of the renal artery followed by 10 min of reperfusion or underwent a 20-min sham period of anesthesia. Subsequently, the left coronary artery was then occluded for 30 min and reperfused for 2 h. Infarct-to-risk ratio was limited from 32.7 ± 4.0% (n = 12) in controls to 17.8 ± 3.0% (n = 9; P = 0.002) in preconditioned hearts. Protection was abolished by 7.5 mg/kg iv 8-SPT (36.7 ± 3.7%; n = 6) or 5 mg/kg iv 5-HD (33.1 ± 4.4%; n = 6) administered before preconditioning. 8-SPT (40.0 ± 4.4%; n = 6) or 5-HD (40.5 ± 4.2%; n = 6) did not affect infarct-to-risk ratio in sham controls. Thus activation of both adenosine receptors and KATP channels appears to be involved in acute renal preconditioning of the myocardium.

The phenomenon of ischemic preconditioning (brief periods of ischemia interspersed with reperfusion) is well established in the heart (16) and has also been observed in a number of other organs, including the brain (8), liver (11), and skeletal muscle (15). There is also limited evidence for increased tolerance to ischemia in the kidney; however, much of the data are contradictory, and the profile of protection appears to differ from preconditioning of the myocardium (20, 21).

Brief coronary artery occlusion has, furthermore, been demonstrated to precondition the myocardium not only within but also beyond the perfusion territory of the artery (17), termed “intraorgan” or “remote” preconditioning. The phenomenon of remote preconditioning is not, however, unique to the heart because Liauw et al. (9) have shown that one skeletal muscle can be protected against ischemia-reperfusion injury by prior ischemic preconditioning of the contralateral muscle. This evidence has lead to speculation that ischemic preconditioning of one organ might confer protection on a remote organ.

Interorgan or remote organ preconditioning of the myocardium was first described by McClanahan et al. (12) in an open-chest rabbit model of myocardial infarction. Brief renal artery occlusion followed by a short period of reperfusion was found to limit myocardial infarct size to a similar degree to ischemic preconditioning triggered by brief coronary artery occlusion. This phenomenon has also been observed in rats (4) in which transient ischemia and reperfusion of either the small intestine or the kidney protected the myocardium against prolonged ischemia. Although the evidence for remote organ preconditioning of the myocardium is mounting, there has been very little investigation of the mechanisms involved.

The aims of this study were to investigate this phenomenon of renal preconditioning of the myocardium and to characterize the mechanisms involved, using an in vivo rabbit model of myocardial infarction. The role of two mediators known to be involved in the mechanisms of myocardial preconditioning, in the rabbit, namely adenosine receptors (14) and ATP-sensitive potassium (KATP) channels (5), were studied in this novel form of protection.

MATERIALS AND METHODS

Experimental materials. Sodium 5-hydroxydecanoate (5-HD) and 8-(p-sulfophenyl)theophylline (8-SPT) were obtained from Research Biochemicals (through Senat, St. Albans, UK), and both were dissolved in 0.9% wt/vol sodium chloride. Zinc-cadmium sulfide microspheres (1–10 µm) were from Duke Scientific (Palo Alto, CA), and 2,3,5-triphenyltetrazolium chloride was from Sigma (Poole, UK). All other reagents were of analytic quality. Male New Zealand White rabbits (2.2–3.5 kg body wt) were used for these studies and were cared for in accordance with UK Home Office guidelines set out in the Animals (Scientific Procedures) Act 1986.

Experimental procedures. Rabbits were anesthetized with a combination of 0.15 ml/kg im Hypnorm (contains: 0.315 mg/ml fentanyl citrate and 10 mg/ml fluanisone; Janssen Animal Health, Pettebridge, UK) and 40–50 mg/kg iv pentobarbital sodium. Surgical anesthesia was maintained by hourly administration of 0.075 ml/kg im Hypnorm and 5–10 mg/kg iv pentobarbital sodium when required. A tracheotomy was performed, and the animals were mechanically ventilated with oxygen-supplemented room air at a rate of 56 cycles/min. The right carotid artery was cannulated for measurement of hemodynamic, arterial blood gas, and pH parameters. Arterial blood pH was maintained within the range of 7.35–7.50 by adjustment of the tidal volume. The body core temperature,
measured with a rectal thermometer (T200; Digitron Instrumentation, Hertford, UK), was carefully maintained at 38.0–38.5°C by means of a heating pad (Harvard Instruments, Edenbridge, UK).

A left thoracotomy and pericardiotomy were performed, and a 3-0 silk suture (Mersilk W546; Ethicon) was placed around an anterolateral branch of the left coronary artery approximately midway between the left atrial insertion and the apex. A left laparotomy was then performed and the left renal artery exposed. A piece of polycotton silk was passed under the artery to facilitate later retrieval. To prevent spasm of the exposed renal artery, 0.25–0.5 ml of 2% wt/vol lidocaine solution was administered locally. Either 7.5 mg/kg 8-SPT (10) or 5 mg/kg 5-HD (6) was administered by intravenous bolus 10 or 15 min, respectively, before the preconditioning/sham period. Animals then underwent either a 20-min sham period of anesthesia or 10 min of left renal artery occlusion followed by 10 min of reperfusion. The renal artery was occluded 1–2 cm proximal to the kidney using anatraumatic clip; the polycotton silk was used to gently lift the artery to facilitate placement of the clip. Successful occlusion was confirmed visually by a change in the surface color of the left kidney from pinkish-red to cream; reperfusion was confirmed by blushing of the previously discolored kidney surface. Ten minutes later the coronary artery was occluded for 30 min by clamping the ligature with a polypropylene snare. Occlusion was verified by the appearance of epicardial cyanosis and ST-segment deviation in the surface electrocardiogram.

Reperfusion was instituted for 2 h by releasing the snare and was visually confirmed by epicardial blushing, gradual resolution of the electrocardiogram signal, and the occurrence of reperfusion-induced ventricular premature beats. At the end of reperfusion, 500IU heparin sodium was administered intravenously. The animal was administered an overdose of anesthesia, and the heart was then excised and perfused in the Langendorff mode with 0.9% wt/vol saline to wash out blood. The coronary artery ligature was securely tied, and a 5 mg/ml suspension of fluorescent zinc-cadmium sulfide microspheres, prepared in 0.9% wt/vol saline, was slowly infused through the aorta to delineate the myocardial risk zone under ultraviolet light. Hearts were trimmed of excess tissue, leaving only the left ventricle, frozen at −18°C for 2–18 h, and then sectioned into 2-mm transverse sections from apex to base. Slices were incubated in 1% wt/vol triphenyltetrazolium chloride in phosphate buffer (pH 7.4) at 37°C. Triphenyltetrazolium chloride reacts with dehydrogenases in viable tissue, producing a red formazan derivative, which is distinguished from the grey necrotic tissue on fixing with 4% vol/vol formaldehyde solution. Left ventricular infarct and risk volumes were determined in a blinded fashion using a computerized planimetric technique (Kurta, Phoenix, AZ), and the infarct size was expressed as the percentage of infarction of the risk zone.

An additional four rabbits were used to investigate the effects of increasing doses of adenosine on diastolic blood pressure and heart rate. Animals were anesthetized, as above, and a catheter was advanced from the left femoral vein into the vena cava to approximately the level of the renal veins. Boluses of saline followed by increasing doses of adenosine (10, 30, 100, and 300 µg/kg) were administered intravenously, and the diastolic blood pressure and heart rate were monitored. Blood pressure and heart rate were allowed to return to baseline between each administration. This dosing regime was performed twice in each animal.

Experimental treatment groups. Renal preconditioning protection of the mycardium was evaluated by randomly assigning rabbits to groups I and II. The role of adenosine receptors and KATP channels in preconditioning was then investigated by random assignment of additional rabbits to groups III to VI inclusive, with the addition of extra sham and preconditioned controls (Fig. 1). Group I was the control sham period corresponding to renal artery occlusion (sham) before 30 min of ischemia and 2 h of reperfusion. Group II was renal preconditioning (PC), which consisted of 10 min of renal ischemia followed by 10 min of reperfusion before ischemia-reperfusion. Group III consisted of sham + 8-SPT (7.5 mg/kg iv) 10 min before the sham period. Group IV was PC + 8-SPT (7.5 mg/kg iv) 10 min before renal PC. Group V was sham + 5-HD (5 mg/kg iv) 15 min before the sham period. Group VI was PC + 5-HD (5 mg/kg iv) 15 min before renal PC.

Statistical analysis. Differences in hemodynamic data, arterial blood pressure, core body temperature, and infarct size among groups were compared by one-factor ANOVA followed by Fishers protected least-significant difference test. All data are means ± SE. A P ≤ 0.05 was indicative of a statistically significant difference between groups.

Exclusion criteria. Animals that developed ischemia- or reperfusion-induced ventricular fibrillation (VF) that could not be restored to normal sinus rhythm within 2 min were excluded. Hearts were additionally excluded if the risk or infarct zones were not clearly defined or if the risk volume was <0.4 cm3 or >1.9 cm3.

RESULTS

Exclusions. A total of 62 rabbits were used in this study. Thirteen animals were excluded: one due to failure to occlude the renal artery, two due to failure to occlude the coronary artery, two due to failure to reperfuse the coronary artery, one due to severe hypo-
tension, one due to intractable ischemia-induced VF, two due to poor delineation of the risk zone, and four due to a risk zone <0.4 cm³ or >1.9 cm³. Final numbers in the study were therefore 12 sham, 9 renal PC, 6 sham + 8-SPT, 6 PC + 8-SPT, 6 sham + 5-HD, 6 PC + 5-HD, and 4 for the adenosine dose response.

Incidence of ventricular fibrillation. Nine hearts underwent VF during coronary artery occlusion or shortly after reperfusion: three sham, two sham + 8-SPT, two PC + 8-SPT, one sham + 5-HD, and one PC + 5-HD. There was a single incidence of transient reperfusion-induced VF in the sham + 8-SPT group. With the exception of one sham, which developed intractable VF and was excluded (see above), all were converted to normal sinus rhythm within 2 min and were included in the statistical analysis. None of the preconditioned hearts demonstrated VF.

Hemodynamic and temperature data. There were no differences in heart rate, mean arterial blood pressure, rate-pressure product (systolic blood pressure × heart rate), or core body temperature (Table 1) except at baseline for rate-pressure product in the preconditioning group. This resulted from a significantly higher systolic blood pressure; however, there were no differences during ischemia or reperfusion among the groups. The overall similarity in systemic hemodynamic and blood gas parameters, with careful maintenance of core body temperature (38.0–38.5°C) and pH (7.35–7.50), suggests that these factors did not contribute to the significant differences observed in infarct size.

Infarct data. Myocardial infarct volume was significantly smaller in animals pretreated with renal preconditioning (0.17 ± 0.03 cm³, n = 9) compared with sham controls (0.41 ± 0.07 cm³, n = 12; P < 0.05 by one-factor ANOVA). Neither 7.5 mg/kg 8-SPT (0.53 ± 0.07 cm³; n = 6) nor 5 mg/kg 5-HD (0.57 ± 0.08 cm³; n = 6) alone affected sham control infarct volume. These treatments, however, abolished the infarct volume limitation afforded by renal preconditioning (0.48 ± 0.06 cm³, n = 6, and 0.40 ± 0.07 cm³, n = 6, respectively). When infarct size was expressed as a percentage of the risk zone, renal preconditioning resulted in significantly smaller infarcts (17.8 ± 3.0%) compared with sham controls (32.7 ± 4.0%; P = 0.002 by one-factor ANOVA). This 46% limitation in infarct size is indicative of acute myocardial protection afforded by brief renal ischemia-reperfusion and was abolished by 7.5 mg/kg 8-SPT or 5 mg/kg 5-HD (36.7 ± 3.7 and 33.1 ± 4.4%, respectively). Neither compound alone had any effect on sham control infarct size (40.0 ± 4.4 and 40.5 ± 4.2%, respectively). Figure 2 is a graphical representation of percent infarction within the risk zone. Because mean myocardial risk volume was similar for all six groups (1.0–1.4 cm³),

Table 1. Hemodynamic and temperature data

<table>
<thead>
<tr>
<th>Group</th>
<th>Baseline</th>
<th>5 min</th>
<th>15 min</th>
<th>29 min</th>
<th>Reperfusion</th>
<th>60 min</th>
<th>120 min</th>
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<tr>
<td></td>
<td>Heart rate, beats/min</td>
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<tr>
<td>SH</td>
<td>227 ± 6.3</td>
<td>225 ± 6.9</td>
<td>220 ± 6.5</td>
<td>198 ± 6.1</td>
<td>199 ± 6.2</td>
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<tr>
<td>PC</td>
<td>246 ± 6.0</td>
<td>229 ± 7.1</td>
<td>231 ± 8.2</td>
<td>211 ± 5.6</td>
<td>208 ± 7.2</td>
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<tr>
<td>SH + 8-SPT</td>
<td>246 ± 15.6</td>
<td>247 ± 12.2</td>
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<td>254 ± 12.1</td>
<td>229 ± 17.7</td>
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<td>PC + 8-SPT</td>
<td>230 ± 11.8</td>
<td>246 ± 10.8</td>
<td>245 ± 10.0</td>
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<tr>
<td>SH + 5-HD</td>
<td>236 ± 8.7</td>
<td>228 ± 11.5</td>
<td>224 ± 16.2</td>
<td>207 ± 16.1</td>
<td>191 ± 16.0</td>
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<tr>
<td>PC + 5-HD</td>
<td>223 ± 8.4</td>
<td>218 ± 11.0</td>
<td>216 ± 12.9</td>
<td>184 ± 9.0</td>
<td>223 ± 8.8</td>
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<td>MAP, mmHg</td>
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<tr>
<td>SH</td>
<td>75 ± 3.8</td>
<td>72 ± 3.2</td>
<td>67 ± 2.7</td>
<td>61 ± 3.4</td>
<td>58 ± 3.6</td>
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<tr>
<td>PC</td>
<td>83 ± 3.0</td>
<td>68 ± 2.5</td>
<td>67 ± 1.9</td>
<td>64 ± 1.6</td>
<td>61 ± 2.4</td>
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<tr>
<td>SH + 8-SPT</td>
<td>70 ± 2.9</td>
<td>72 ± 5.5</td>
<td>73 ± 3.7</td>
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<td>55 ± 3.6</td>
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<tr>
<td>PC + 8-SPT</td>
<td>67 ± 1.3</td>
<td>73 ± 1.5</td>
<td>68 ± 2.0</td>
<td>57 ± 1.6</td>
<td>56 ± 1.5</td>
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<tr>
<td>SH + 5-HD</td>
<td>76 ± 3.3</td>
<td>70 ± 3.6</td>
<td>60 ± 3.2</td>
<td>57 ± 1.5</td>
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<tr>
<td>PC + 5-HD</td>
<td>74 ± 5.0</td>
<td>68 ± 4.8</td>
<td>66 ± 4.4</td>
<td>58 ± 3.7</td>
<td>58 ± 4.1</td>
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<td>RPP</td>
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<tr>
<td>SH</td>
<td>22,560 ± 970</td>
<td>21,811 ± 1,061</td>
<td>20,103 ± 675</td>
<td>16,836 ± 437</td>
<td>16,183 ± 664</td>
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<tr>
<td>PC</td>
<td>27,226 ± 1,338*</td>
<td>22,913 ± 1,140</td>
<td>22,444 ± 1,094</td>
<td>19,630 ± 690</td>
<td>18,854 ± 978</td>
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<tr>
<td>SH + 8-SPT</td>
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<td>24,739 ± 1,741</td>
<td>24,302 ± 1,764</td>
<td>20,282 ± 1,056</td>
<td>17,206 ± 826</td>
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<td>PC + 8-SPT</td>
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<td>22,795 ± 911</td>
<td>21,705 ± 1,440</td>
<td>17,591 ± 860</td>
<td>17,563 ± 1,182</td>
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<tr>
<td>SH + 5-HD</td>
<td>23,436 ± 1,189</td>
<td>21,589 ± 1,528</td>
<td>21,450 ± 2,248</td>
<td>18,162 ± 1,671</td>
<td>17,540 ± 1,459</td>
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<tr>
<td>PC + 5-HD</td>
<td>22,456 ± 1,121</td>
<td>19,793 ± 1,095</td>
<td>19,133 ± 1,069</td>
<td>15,126 ± 922</td>
<td>15,521 ± 1,502</td>
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<tr>
<td>SH</td>
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<td>38.2 ± 0.05</td>
<td>38.1 ± 0.04</td>
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<tr>
<td>PC</td>
<td>38.0 ± 0.1</td>
<td>38.3 ± 0.04</td>
<td>38.1 ± 0.01</td>
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<tr>
<td>SH + 8-SPT</td>
<td>38.1 ± 0.1</td>
<td>38.2 ± 0.1</td>
<td>38.1 ± 0.03</td>
<td>38.2 ± 0.03</td>
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<td>PC + 8-SPT</td>
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<td>38.1 ± 0.02</td>
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<tr>
<td>SH + 5-HD</td>
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</table>

Data are means ± SE. 5-HD, sodium 5-hydroxydecanoate; MAP, mean arterial pressure; PC, renal preconditioning; RPP, rate-pressure product (systolic blood pressure × HR); SH, sham; 8-SPT, 8-(p-sulphophenyl) theophylline. Group numbers were 12 SH, 9 PC, 6 SH + 8-SPT, 6 PC + 8-SPT, 6 SH + 5-HD and 6 PC + 5-HD. *P < 0.05 compared with all other groups.
the observed differences in infarct size were not likely to be due to variations in the risk zone.

Blood pressure. An interesting observation arising from this study was the appearance of a transient drop in diastolic blood pressure (mean drop of 22 ± 0.9 mmHg lasting ~30 s; n = 7) when the renal artery clip was removed to allow reperfusion (Fig. 3). This fall in blood pressure was associated, in the majority of instances, with a small tachycardia (10–20 beats/min). The drop in diastolic blood pressure was abolished by pretreatment with 8-SPT but only attenuated by 5-HD (mean drop of 14 ± 1.5 mmHg; n = 6). The duration of the response, however, was unchanged at ~30 s. The associated tachycardia was also abolished by 8-SPT but was unchanged by 5-HD (Fig. 3).

The effects of increasing doses of adenosine, administered at the level of the renal veins, were subsequently investigated on the diastolic blood pressure and heart rate. Tachyphylaxis did not occur with increasing doses of adenosine; therefore, blood pressure and heart rate responses were averaged for the two dose-response curves performed in each animal. Saline was found not to alter the blood pressure or heart rate. The effect on diastolic blood pressure produced by the lowest dose of adenosine (10 µg/kg) was indistinguishable from the saline controls, although it did result in a small increase in heart rate. Doses of 30, 100, and 300 µg/kg adenosine produced a dose-dependent decrease in diastolic blood pressure (Fig. 4). Tachycardia, probably reflex in origin, was observed to coincide with the drop in blood pressure, and only at the highest dose of adenosine was this tachycardia preceded by an appreciable bradycardia.

DISCUSSION

In this in vivo rabbit model of myocardial infarction, renal preconditioning, instigated by a brief period of left renal artery occlusion followed by reperfusion, limited myocardial infarct size by 46%. This protection was abolished by pretreatment with the nonselective adenosine receptor antagonist 8-SPT or the K<sub>ATP</sub> channel blocker 5-HD before renal preconditioning.
minimized in the following ways: 1) by excluding risk zones <0.4 cm³ and >1.9 cm³; 2) by expressing infarct size as a percentage of the risk zone to allow for differences in vascular anatomy or occlusion site; 3) by maintaining core body temperature at 38.0–38.5°C, physiological for the rabbit, since differences of 1°C during ischemia can markedly affect infarct size (2); and 4) by choosing the rabbit because it is a noncollateralized species.

Renal preconditioning of the myocardium was first reported by McClanahan et al. (12) in a similar in vivo rabbit model of myocardial infarction. In their study, however, control infarct size (43 ± 3%) was limited to the same degree as coronary preconditioning (renal PC: 11 ± 2%; coronary PC: 8 ± 2%). The reason for the difference in the magnitude of protection between the two studies is unclear, although contemporaneous studies on myocardial preconditioning were not carried out in the present series of experiments. Coronary, mesenteric, and renal ischemic preconditioning of the myocardium in the rat has been shown to be enhanced (coronary and mesenteric) or become apparent (renal) by whole body hypothermia (4). Because core body temperature was not reported by McClanahan et al. (12), perhaps the difference in the magnitude of protection arises from differing temperature conditions. McClanahan et al. (12) and Gho et al. (4) have, moreover, demonstrated the importance of brief intervening reperfusion between the remote organ preconditioning and sustained myocardial ischemia. Protection was lost if the renal or mesenteric artery, respectively, was permanently occluded before myocardial ischemia, suggesting that infarct size limitation required the washout of an endogenous protective factor from the previously ischemic organ.

Potential mechanisms for renal preconditioning of myocardium. The exact mechanisms by which adenosine and K_{ATP} channels mediate renal preconditioning of the myocardium are unclear. Several possibilities warrant consideration including the following. First, adenosine, generated within the ischemic kidney, might be the sole mediator of cardioprotection. Experiments with bolus doses of adenosine showed that ~50 µg/kg iv elicited a fall in blood pressure comparable to that seen immediately on renal artery reperfusion. This implies that the kidney generated at least 100–150 µg of adenosine during the 10 min of ischemia (assuming complete washout on reperfusion). It seems unlikely, however, that this would be sufficient to confer cardioprotection directly, since doses of adenosine as high as 5 mg iv (infused over 5 min) (11) and 25 mg/kg iv (4a) failed to protect rabbit myocardium. Second, adenosine, acting via adenosine receptors within the kidney, might evoke local release of another substance more stable in blood, such as kinins, endothelins, prostaglandins, or renal medullary lipids, which may confer cardioprotection perhaps via a K_{ATP} channel-linked mechanism. Third, adenosine, or some substance released by it locally within the kidney, might stimulate renal afferents. A neurogenic mode of action has been alluded to by Gho et al. (4), who prevented mesenteric ischemic preconditioning of the myocardium with the ganglion blocker hexamethonium.

5-HD has been demonstrated to abolish cardioprotection without affecting the action potential duration shortening or vasodilator effects of potassium channel openers (3, 13), which are mediated via sarcolemmal K_{ATP} channels. In addition, K^+ flux through reconstituted rat heart mitochondrial K_{ATP} channels was inhibited by 5-HD (K_{1/2} = 83 µM, where K_{1/2} is half-maximal K^+ flux) (3). These data suggest that 5-HD is a specific blocker of mitochondrial K_{ATP} channels and, therefore, implicates a possible role for these channels in renal preconditioning of the myocardium.

Recent evidence for remote organ preconditioning. As this study was in progress, remote organ preconditioning of the myocardium was further reported. Birnbaum et al. (1) demonstrated that skeletal muscle ischemic preconditioning in the rabbit, elicited by reduction of blood flow with concomitant electrical stimulation of the gastrocnemius muscle, limited myocardial infarct
size arising from sustained ischemia. In contrast to all the other studies discussed here, there was no interven-
ing reperfusion between muscle preconditioning and sustained ischemia. Because preconditioning was in-
duced by partial stenosis of the femoral artery, there
would, however, have been residual blood flow through
the muscle, allowing the potential release of protective
mediators into the circulation. Takaoka et al. (18),
using a similar rabbit model to ours, reported a com-
parable limitation of myocardial infarct size by renal
preconditioning (controls: 34%, renal PC: 20%, coronary
PC: 16%). Coronary or renal preconditioning also dimin-
dished the detrimental effects of sustained ischemia on
myocardial energy metabolism, determined by intracel-
lar pH, and the concentration of ATP and phosphocreas-
tine. This limitation of infarct size and improvement
in myocardial energy metabolism were both abolished
by 8-SPT, thus providing the first indication for a role of
adenosine receptors in remote organ preconditioning of
the myocardium. It was noted in those studies that
8-SPT was administered during the intervening period of
reperfusion between preconditioning and sustained
ischemia, suggesting that occupation of adenosine recep-
tors by adenosine is necessary during the prolonged
myocardial ischemia. In our study, 8-SPT was given
before the preconditioning ischemia, implying that
adenosine receptors are important for both triggering
and mediating remote organ preconditioning of the
myocardium, akin to acute coronary preconditioning
(19). However, the half-life of 8-SPT, ~45 min, (10)
is long enough to block adenosine receptors during both
the preconditioning and sustained ischemia.

In summary, renal ischemic preconditioning pro-
vided significant protection against sustained myocar-
dial ischemia. The nonselective adenosine receptor
antagonist 8-SPT or the K<sub>ATP</sub> channel blocker 5-HD
abolished this protection. This study provides the first
evidence for the activation of K<sub>ATP</sub> channels, perhaps in
the inner mitochondrial membrane, in the mechanism
of remote organ preconditioning and supports a role for
adenosine receptors in this phenomenon. Further inves-
tigation, however, is required to elucidate the precise
nature of the involvement of adenosine receptor and K<sub>ATP</sub>
channel activation in cardioprotection afforded by re-

dote organ preconditioning.

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Address for reprint requests: G. F. Baxter, The Hatter Institute
for Cardiovascular Studies, Univ. College London Hospitals &
Medical School, Grafton Way, London WC1E 6DB, UK.

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