Mechanisms of Desflurane-induced Preconditioning in Isolated Human Right Atria In Vitro

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Background: The authors examined the role of adenosine triphosphate–sensitive potassium (KATP) channels, adenosine A1 receptor, and α and β adrenoceptors in desflurane-induced preconditioning in human myocardium, in vitro.

Methods: The authors recorded isometric contraction of human right atrial trabeculae suspended in oxygenated Tyrode’s solution (34°C; stimulation frequency, 1 Hz). Before a 30-min anoxic period, 3, 6, and 9% desflurane was administered during 15 min. Desflurane, 6%, was also administered in the presence of 10 μM glibilidecamide, a KATP channel antagonist; 10 μM HMR 1098, a sarcolemmal KATP channel antagonist; 800 μM 5-hydroxy-decanate (5-HD), a mitochondrial KATP channel antagonist; 1 μM phenolamine, an α-adrenoceptor antagonist; 1 μM propranolol, a β-adrenoceptor antagonist; and 100 μM 8-cyclopentyl-1,3-dipropylxanthine (DPX), the adenosine A1 receptor antagonist. Developed force at the end of a 60-min reoxygenation period was compared (mean ± SD).

Results: Desflurane at 3% (95 ± 13% of baseline), 6% (86 ± 6% of baseline), and 9% (82 ± 6% of baseline) enhanced the recovery of force after 60 min of reoxygenation as compared with the control group (50 ± 11% of baseline). Glibilidecamide (60 ± 12% of baseline), 5-HD (57 ± 21% of baseline), DPX (63 ± 19% of baseline), phenolamine (56 ± 20% of baseline), and propranolol (63 ± 13% of baseline) abolished desflurane-induced preconditioning. In contrast, HMR 1098 (85 ± 12% of baseline) did not modify desflurane-induced preconditioning.

Conclusions: In vitro, desflurane preconditioning human myocardium against simulated ischemia through activation of mitochondrial KATP channels, adenosine A1 receptor, and α and β adrenoceptors.

Materials and Methods

Experimental Conditions

After approval of the local medical ethics committee (Caen, France), right atrial appendages were obtained during cannulation for cardiopulmonary bypass from patients scheduled for routine coronary artery bypass surgery or aortic valve replacement. All patients received midazolam or propofol, sufentanil, etomidate, pancuronium, and isoflurane. Patients with atrial arrhythmia and those who were taking oral hypoglycemic medications were excluded from the study.

Right atrial trabeculae (one to two per appendage) were dissected and suspended vertically between an isometric force transducer (UC3; Gould, Cleveland, OH) and a stationary stainless clip in a 200-ml jacketed reservoir filled with daily prepared Tyrode’s modified solution containing 120 mM NaCl, 3.5 mM KCl, 1.1 mM MgCl2, 1.8 mM NaH2PO4, 25.7 mM NaHCO3, 2.0 mM CaCl2, and 11 mM glucose. The reservoir was maintained at 34°C with use of a thermostatic water circulator (Polystat micropros; Bioblock, Illkirch, France). The bathing solution was bubbled with carbogen (95% O2–5% CO2), resulting in a pH of 7.40 and a partial pressure of oxygen of 600 mmHg. Isolated muscles were field-stimulated at 1 Hz by two platinum electrodes with rectangular wave...
pulses of 5 ms duration 20% above threshold (CMS 95107; Bionic Instrument, Paris, France).

Trabeculae were equilibrated for 60-90 min to allow stabilization of their optimal mechanical performance at the apex of the length-active isometric tension curve (Lmax). At the end of the stabilization period, trabeculae were randomized to experimental groups detailed below. The force developed was measured continuously, digitized at a sampling frequency of 400 Hz, and stored on a Writeable Compact Disc for analysis (MacLab; AD Instrument, Sydney, Australia).

At the end of each experiment, the length and the weight of the muscle were measured. The muscle cross-sectional area was calculated from its weight and length assuming a cylindrical shape and a density of 1. To avoid core hypoxia, trabeculae included in the study should have a cross-sectional area less than 1.0 mm², an active isometric force normalized per cross-sectional area (AF) greater than 5.0 mN/mm², and a ratio of resting force/total force (RF/TF) less than 0.45.

**Experimental Protocol**

In a time control group (n = 10), we measured the AF of isolated human atrial trabeculae every 10 min during 120 min.

In all other groups, ischemia-reperfusion was simulated by replacing 95% O2-5% CO2 with 95% N2-5% CO2 in the buffer for 30 min, followed by a 60-min oxygenated recovery period (I-R protocol).

In the control group (control; n = 11) muscles were exposed to the I-R protocol alone. Anoxic preconditioning (APC; n = 6) was induced by a 4-min anoxic period followed by a 7-min oxygenated period before the I-R protocol.9,11 The mechanisms involved in APC were studied by 15 min of pretreatment with 10 µm glibenclamide (n = 7), a nonselective KATP channel antagonist; 10 µm HMR 1098 (n = 6), a selective sarcolemmal KATP channel antagonist; 800 µm 5-hydroxydecanoate (5-HD; n = 7), a selective mitochondrial KATP channel antagonist; 0.1 µm 8-cyclopentyl-1,3-dipropylxanthine (DPX; n = 6), a selective adenosine A1 receptor antagonist; 1 µm propranolol (n = 6), a β-adrenoceptor antagonist; and 1 µm phentolamine (n = 6), an α-adrenoceptor antagonist. Concentrations used have been validated in previous experimental studies in human myocardium in vitro.2,11,15,18

In the desflurane treatment groups, desflurane was delivered to the organ bath by bubbling with 95% O2-5% CO2 passing through a specific calibrated vaporizer. Desflurane concentration in the carrier gas phase was measured with an infrared calibrated analyzer (Capnomac; Datex, Helsinki, Finland). After a 15-min exposure to 3% (n = 5), 6% (n = 6), and 9% (n = 5) desflurane, muscles underwent the I-R protocol. Mechanisms involved in desflurane-induced preconditioning were studied with 6% desflurane. Thus, 6% desflurane was administered after 15 min of pretreatment with 10 µm glibenclamide (n = 6), 10 µm HMR 1098 (n = 6), 800 µm 5-HD (n = 6), 10 nm DPX (n = 6), 1 µm propranolol (n = 6), and 1 µm phentolamine (n = 6).

In additional groups, the I-R protocol was performed after 15 min of pretreatment with 10 µm glibenclamide (n = 4), 10 µm HMR 1098 (n = 4), 800 µm 5-HD (n = 4), 0.1 µm DPX (n = 4), 1 µm propranolol (n = 4), and 1 µm phentolamine (n = 4).

Glibenclamide, 5-HD, DPX, and phentolamine were purchased from ICN Pharmaceuticals (Orsay, France), and desflurane was purchased from GlaxoWellcome (Marly-le-Roi, France). HMR 1098 was a gift from Aventis Pharma (Frankfurt am Main, Germany).

**Statistical Analysis**

Data are expressed as mean ± SD. Baseline values of main mechanical parameters and values of AF at 60 min of reperfusion were compared by a univariate analysis of variance (ANOVA). If an F value was less than 0.05, Newman-Keuls post hoc analysis was used. Within-group data were analyzed over time using univariate ANOVA for repeated-measures and Newman-Keuls post hoc analysis. All P values were two-tailed, and a P value of less than 0.05 was required to reject the null hypothesis. Statistical analysis was performed using Statview 5 software (Deltasoft, Meylan, France).

**Results**

One hundred forty-two human right atrial trabeculae were studied. There were no differences in baseline values for Lmax, cross-sectional area, RF/TF, and AF among all groups (table 1).

**Stability with Time of Isolated Human Atrial Trabeculae**

In the time control group, AF slightly decreased with time (fig. 1). This decrease became significant at 80 min (AF: 93 ± 7% of baseline; P < 0.05). At 120 min, AF was 90 ± 10% of baseline. Resting force was not significantly modified with time and was 97 ± 10% of baseline at 120 min.

**Effects of Simulated Ischemia and Reperfusion on Contractile Force of Human Right Atrial Trabeculae**

In the time control group, AF slightly decreased with time (fig. 1). This decrease became significant at 80 min (AF: 93 ± 7% of baseline; P < 0.05). At 120 min, AF was 90 ± 10% of baseline. Resting force was not significantly modified with time and was 97 ± 10% of baseline at 120 min.

**Effects of Glibenclamide, 5-HD, HMR 1098, DPX, Propranolol, and Phentolamine on Simulated Ischemia-Reperfusion**

The decrease in AF induced by pretreatment with glibenclamide (AF: 94 ± 4% of baseline), 5-HD (AF: 94 ± 1%
of baseline), HMR 1098 (AF: 86 ± 12% of baseline), DPX (AF: 97 ± 3% of baseline), propranolol (AF: 89 ± 4% of baseline), and phenolamine (AF: 96 ± 2% of baseline) was not different among groups. As shown in figure 1 the time course of AF in the control group was not modified by 15 min of pretreatment with glibenclamide, 5-HD, HMR 1098, DPX, propranolol, and phenolamine. The recovery of AF at 60 min of reoxygenation measured in the control group (50 ± 11% of baseline) was not different from groups pretreated with glibenclamide, 5-HD, HMR 1098, DPX, propranolol, and phenolamine. The recovery of AF at 60 min of reoxygenation measured in the control group (50 ± 11% of baseline) was not different from groups pretreated with glibenclamide (61 ± 7% of baseline), 5-HD (56 ± 11% of baseline), HMR 1098 (62 ± 6% of baseline), DPX (47 ± 4% of baseline), propranolol (53 ± 15% of baseline), and phenolamine (58 ± 8% of baseline).

Mechanisms of Anoxic Preconditioning on Human Right Atrial Trabeculae

Figure 2 shows the time course of AF for APC group. The 4-min anoxic challenge induced a marked decrease in AF (37 ± 16% of baseline) followed by complete recovery after 7 min of reoxygenation (107 ± 5% of baseline). In the APC group, AF after 30 min of simulated ischemia was 22 ± 10% of baseline. At the end of the 60-min reoxygenated period, the recovery of AF in the APC group was significantly greater than that measured in the control group (91 ± 3 vs. 50 ± 11% of baseline; P < 0.05).

As shown in figure 3, the enhanced recovery of AF induced by APC was significantly decreased by pretreatment with glibenclamide (50 ± 11% of baseline), HMR 1098 (59 ± 15% of baseline), 5-HD (56 ± 19% of baseline), and DPX (63 ± 16% of baseline) and was no more different from the recovery of AF measured in the control group. In contrast, phenolamine (90 ± 19% of baseline) and propranolol (97 ± 6% of baseline) did not modify the enhanced recovery of AF induced by APC (fig. 5).

Direct Inotropic Effects of Desflurane

Desflurane at 3% (93 ± 2% of baseline; P < 0.05), 6% (87 ± 12% of baseline; P < 0.05), and 9% (76 ± 5% of baseline; P < 0.05) induced a dose-dependent decrease in AF. At a concentration of 6%, the desflurane-induced decrease in AF was not different among groups pretreated with glibenclamide (AF: 90 ± 7% of baseline), 5-HD (AF: 78 ± 6% of baseline), HMR 1098 (AF: 77 ± 10% of baseline), DPX (AF: 89 ± 4% of baseline), propranolol (AF: 72 ± 8% of baseline), and phenolamine (AF: 67 ± 15% of baseline). As shown in figure 4, the decrease in AF induced by 6% desflurane was significantly greater in the presence of propranolol and phenolamine.

Effects of Desflurane on Simulated Ischemia-Reperfusion

As depicted in figure 5, 15 min of exposure to desflurane at 3% (AF: 86 ± 6% of baseline), 6% (AF: 5 ± 13% of baseline), and 9% (AF: 82 ± 6% of baseline) prior to

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Table 1. Control Values of Main Mechanical Parameters of Human Right Atrial Trabeculae

<table>
<thead>
<tr>
<th>Experimental Groups</th>
<th>L_{max} (mm)</th>
<th>CSA (mm²)</th>
<th>AF (mN/mm²)</th>
<th>RF/TF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time control</td>
<td>8.0 ± 1.8</td>
<td>0.62 ± 0.16</td>
<td>25 ± 13</td>
<td>0.35 ± 0.12</td>
</tr>
<tr>
<td>Control</td>
<td>7.0 ± 1.1</td>
<td>0.75 ± 0.28</td>
<td>15 ± 8</td>
<td>0.38 ± 0.14</td>
</tr>
<tr>
<td>I-R + glibenclamide</td>
<td>6.9 ± 0.7</td>
<td>0.50 ± 0.16</td>
<td>27 ± 18</td>
<td>0.31 ± 0.10</td>
</tr>
<tr>
<td>I-R + HMR 1098</td>
<td>7.6 ± 2.1</td>
<td>0.49 ± 0.11</td>
<td>28 ± 16</td>
<td>0.30 ± 0.14</td>
</tr>
<tr>
<td>I-R + 5-HD</td>
<td>6.8 ± 1.3</td>
<td>0.65 ± 0.21</td>
<td>21 ± 11</td>
<td>0.27 ± 0.10</td>
</tr>
<tr>
<td>I-R + DPX</td>
<td>7.5 ± 0.5</td>
<td>0.66 ± 0.29</td>
<td>15 ± 10</td>
<td>0.41 ± 0.21</td>
</tr>
<tr>
<td>I-R + propranolol</td>
<td>7.8 ± 0.7</td>
<td>0.60 ± 0.11</td>
<td>23 ± 12</td>
<td>0.25 ± 0.11</td>
</tr>
<tr>
<td>I-R + phenolamine</td>
<td>6.0 ± 1.2</td>
<td>0.80 ± 0.17</td>
<td>15 ± 7</td>
<td>0.30 ± 0.07</td>
</tr>
<tr>
<td>APC</td>
<td>7.0 ± 1.4</td>
<td>0.79 ± 0.30</td>
<td>18 ± 8</td>
<td>0.27 ± 0.12</td>
</tr>
<tr>
<td>APC + glibenclamide</td>
<td>8.1 ± 1.8</td>
<td>0.61 ± 0.14</td>
<td>15 ± 6</td>
<td>0.35 ± 0.17</td>
</tr>
<tr>
<td>APC + HMR 1098</td>
<td>6.6 ± 0.7</td>
<td>0.69 ± 0.23</td>
<td>21 ± 16</td>
<td>0.32 ± 0.14</td>
</tr>
<tr>
<td>APC + 5-HD</td>
<td>8.6 ± 1.0</td>
<td>0.51 ± 0.12</td>
<td>26 ± 10</td>
<td>0.25 ± 0.10</td>
</tr>
<tr>
<td>APC + DPX</td>
<td>6.3 ± 0.8</td>
<td>0.67 ± 0.11</td>
<td>15 ± 6</td>
<td>0.27 ± 0.11</td>
</tr>
<tr>
<td>APC + phenolamine</td>
<td>6.3 ± 1.4</td>
<td>0.55 ± 0.16</td>
<td>17 ± 9</td>
<td>0.40 ± 0.07</td>
</tr>
<tr>
<td>APC + propranolol</td>
<td>7.3 ± 1.8</td>
<td>0.60 ± 0.13</td>
<td>23 ± 11</td>
<td>0.30 ± 0.06</td>
</tr>
<tr>
<td>Desflurane 3%</td>
<td>7.0 ± 2.7</td>
<td>0.43 ± 0.10</td>
<td>24 ± 4</td>
<td>0.41 ± 0.05</td>
</tr>
<tr>
<td>Desflurane 6%</td>
<td>8.0 ± 1.0</td>
<td>0.58 ± 0.28</td>
<td>21 ± 5</td>
<td>0.31 ± 0.11</td>
</tr>
<tr>
<td>Desflurane 9%</td>
<td>7.8 ± 0.7</td>
<td>0.60 ± 0.18</td>
<td>28 ± 8</td>
<td>0.20 ± 0.05</td>
</tr>
<tr>
<td>Desflurane + glibenclamide</td>
<td>8.2 ± 1.5</td>
<td>0.58 ± 0.10</td>
<td>19 ± 7</td>
<td>0.29 ± 0.18</td>
</tr>
<tr>
<td>Desflurane + HMR 1098</td>
<td>7.9 ± 1.7</td>
<td>0.71 ± 0.30</td>
<td>23 ± 6</td>
<td>0.23 ± 0.11</td>
</tr>
<tr>
<td>Desflurane + 5-HD</td>
<td>8.5 ± 1.9</td>
<td>0.52 ± 0.17</td>
<td>22 ± 13</td>
<td>0.29 ± 0.16</td>
</tr>
<tr>
<td>Desflurane + DPX</td>
<td>6.8 ± 1.2</td>
<td>0.60 ± 0.20</td>
<td>20 ± 10</td>
<td>0.35 ± 0.14</td>
</tr>
<tr>
<td>Desflurane + phenolamine</td>
<td>7.4 ± 1.3</td>
<td>0.49 ± 0.21</td>
<td>23 ± 6</td>
<td>0.26 ± 0.07</td>
</tr>
<tr>
<td>Desflurane + propranolol</td>
<td>7.5 ± 1.8</td>
<td>0.76 ± 0.24</td>
<td>17 ± 6</td>
<td>0.28 ± 0.10</td>
</tr>
</tbody>
</table>

Values are mean ± SD.

L_{max} = maximal length at the apex of the length-active force curve; CSA = cross-sectional area; AF = acting isometric force normalized per cross-sectional area; RF/TF = ratio of resting force on total force; I-R = simulated ischemia-reoxygenation protocol; 5-HD = 5 hydroxydecanate; DPX = 8-cyclopentyl-1,3-dipropylxanthine.
the 30-min anoxic period resulted in a significant increase in the recovery of AF after 60 min of reoxygenation as compared with the control group (AF: 50 ± 11% of baseline). Recovery of AF at 60 min of reoxygenation measured in the 3, 6, and 9% desflurane groups was not different from that measured in APC group (fig. 5).

Pretreatment with glibenclamide (60 ± 12% of baseline), 5-HD (57 ± 21% of baseline), DPX (63 ± 19% of baseline), propranolol (56 ± 20% of baseline), and phentolamine (63 ± 13% of baseline) abolished desflurane-induced enhanced recovery of AF. In contrast, pretreatment with HMR 1098 (85 ± 12% of baseline) did not modify the enhanced recovery of AF induced by desflurane (fig. 6).

Discussion

The main results of our study are as follows: brief exposure to desflurane (3, 6, and 9%) preconditions isolated human right atrial myocardium against 30 min of simulated ischemia; mechanisms involved in desflurane-induced preconditioning are opening of mitochondrial K\textsubscript{ATP} channels and stimulation of adenosine A\textsubscript{1} receptor and \(\alpha\) and \(\beta\) adrenoceptors.

Strong evidence supports the cardioprotective effects of volatile anesthetics against prolonged ischemia. Isoflurane has been shown to decrease infarct size in canine myocardium \textit{in vivo}\textsuperscript{10} and to improve functional recovery of isolated human myocardium.\textsuperscript{11} Recently, sevoflurane and desflurane have been shown to exert similar cardioprotective effects in dogs.\textsuperscript{13,14} K\textsubscript{ATP} channels have been shown to play a pivotal role in mediating anesthetic-induced preconditioning.\textsuperscript{10–14} It has been suggested that activation of both sarcosomal K\textsubscript{ATP} (sarcK\textsubscript{ATP}) and mitoK\textsubscript{ATP} could be involved in the cardioprotection conferred by volatile anesthetics.\textsuperscript{14} Furthermore, the participation of adenosine A\textsubscript{1} receptor stimulation and mecha-nogated channel activation has recently been suggested in sevoflurane-induced preconditioning.\textsuperscript{11,12} The present results confirm and extend findings of Toller et al.,\textsuperscript{14} who showed that brief exposure to desflurane exerts cardioprotective effects against irreversible ischemia. Our study showed that 15 min of exposure to desflurane prior to 30 min of simulated ischemia enhanced contractile recovery of isolated human myocardium during the reoxygenation period. In addition, we showed that this effect was blocked by glibenclamide, indicating that opening of K\textsubscript{ATP} channels was implicated. Furthermore, specific blockade of mitoK\textsubscript{ATP} channels with 5-HD abolished desflurane-induced preconditioning, suggesting that opening of mitoK\textsubscript{ATP} channels is involved in desflurane-induced preconditioning. In contrast, specific blockade of sarcK\textsubscript{ATP} channels with HMR 1098 did not abolished desflurane-induced preconditioning but abol-

![Fig. 1. Time course of force of contraction of isolated human right atrial trabeculae in the time control group, during 30 min of simulated ischemia followed by 60 min of reoxygenation (control group), and in groups pretreated during 15 min prior to the simulated ischemia with 10 \(\mu\)M glibenclamide, 800 \(\mu\)M 5-hydroxydecanoic acid (5-HD), 10 \(\mu\)M HMR 1098, 0.1 \(\mu\)M 8-cyclopentyl-1,3-dipropylxanthine (DPX), 1 \(\mu\)M propranolol, and 1 \(\mu\)M phentolamine. \#P < 0.05 versus baseline value for control, glibenclamide, 5-HD, HMR 1098, DPX, propranolol, and phentolamine groups; §P < 0.05 versus baseline value for time control group. Data are mean ± SD.](anesthesiology.pubs.asahq.org)
ished APC. These results suggest that opening of sarcK<sub>ATP</sub> is involved in APC but not in desflurane-induced preconditioning. Although current opinion favors a predominant role for mitoK<sub>ATP</sub> channels in ischemic preconditioning, there is evidence demonstrating that sarcK<sub>ATP</sub> channels are important mediators of protection during the reoxygenation phase of injury. 19,20 At this time, the precise role and timing of activation of sarcK<sub>ATP</sub> and mitoK<sub>ATP</sub> channels during ischemic preconditioning remains unresolved. The results of Toller et al. 14 showing that both mitoK<sub>ATP</sub> and sarcK<sub>ATP</sub> channels were implicated in desflurane-induced preconditioning are not in accordance with our results showing that the specific inhibition of sarcK<sub>ATP</sub> channels failed to abolish desflurane-induced preconditioning. This discrepancy may be related to major differences in experimental models. First, it should be emphasized that HMR 1098 should be an effective blocker of sarcK<sub>ATP</sub> channels at 10 μM since its IC<sub>50</sub> value has been reported at 0.8 μM. 20,21 Thus, an HMR 1098 concentration of 10 μM has been used to block more than 90% of sarcK<sub>ATP</sub> channels in various experimental models. 18–21 Second, Toller et al. 14 measured myocardial infarct size after 60 min of coronary artery occlusion and 3 h of reperfusion, whereas we measured recovery of force of contraction in isolated myocardium after 30 min of anoxia and 60 min of reoxygenation. Recent findings suggest distinct roles of sarcK<sub>ATP</sub> and mitoK<sub>ATP</sub> channels in myocardial ischemic preconditioning benefits in infarct volume and contractile recovery. 19,20 Third, we administered K<sub>ATP</sub> channels blockers prior to the administration of desflurane, whereas Toller et al. 14 administered K<sub>ATP</sub> channels blockers prior to and during the administration of desflurane. The timing of pharmacologic agent administration has been suggested to be critical in the preconditioning phenomenon. 7,22 Fourth, interspecies differences may be a factor that helps to explain the different results. Thus, halothane has been shown to precondition rabbit 19 but not rat 17 and human 11 myocardium, and isoflurane has been shown to precondition rabbit 12 and human 11 but not rat 23 myocardium. Finally, differences between atrial and ventricular myocardium, influence of surgical stress, and barbiturate anesthesia in dogs cannot be ruled out.

The opening of K<sub>ATP</sub> channels has been shown to be an important mediator of ischemic and pharmacologic preconditioning. 5–6 Initially, it has been proposed that the decrease in action potential duration induced by opening of sarcK<sub>ATP</sub> resulted in better preservation of energy stores and suppression of deleterious downstream events, such as Ca<sup>2+</sup> overload. However, a lack of

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**Fig. 2.** Time course of force of contraction of isolated human right atrial trabeculae during 30 min of simulated ischemia followed by 60 min of reoxygenation (control group), and in anoxic preconditioned groups. Anoxic preconditioning (APC) was induced by a 4-min anoxic period followed by a 7-min reoxygenation period prior to the simulated ischemia. Pharmacologic groups were pretreated during 15 min prior to APC with 10 μM glibenclamide, 800 μM 5-hydroxydecanoic acid (5-HD), 10 μM HMR 1098, 0.1 μM 8-cyclopentyl-1,3-dipropylxanthine (DPX), 1 μM propranolol, and 1 μM phentolamine. #P < 0.05 versus baseline value for all groups; §P < 0.05 versus propranolol and phentolamine groups for glibenclamide group; *P < 0.05 versus APC, propranolol, and phentolamine groups for control group; ‡P < 0.05 versus APC, propranolol, and phentolamine groups for control, glibenclamide, 5-HD, HMR 1098, and DPX groups; $P < 0.05 versus APC, propranolol, and phentolamine groups for control, glibenclamide, 5-HD, HMR 1098, and DPX groups. Data are mean ± SD.

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The correlation between the extent of action potential shortening and the reduction of infarct size has been shown.24 Furthermore, cardioprotection conferred by KATP channel openers has been shown to occur on quiescent myocardium.18 The participation of mitoKATP in ischemic preconditioning has been supported by numerous studies using specific antagonists or openers.4,5,18 The mechanism of mitoKATP-induced cardioprotection involves the activation of mitoKATP channels, leading to a decrease in intracellular calcium levels and a reduction in mitochondrial dysfunction, which ultimately leads to a decrease in infarct size.
tection may involve alterations in mitochondrial Ca$^{2+}$ handling, the optimization of energy production, and modulation of reactive oxygen species during ischemia or reperfusion. The precise role and importance of sarcK$_{ATP}$ and mitoK$_{ATP}$ during ischemic preconditioning remain unresolved. At the present time, the influence of volatile anesthetics on K$_{ATP}$ channel function has been poorly studied. Recently, Roscoe et al.\textsuperscript{11} showed that isoflurane does not modify sarcolemmal K$_{ATP}$ activation, whereas halothane partially blocked it in isolated human myocytes. These findings suggest that cardioprotective effects of isoflurane do not implicate a direct effect on sarcK$_{ATP}$ channel function but rather an effect on mitoK$_{ATP}$ and upstream intermediates, such as G protein-coupled receptors and PKC. Thus, it has been shown that volatile anesthetic-induced cardioprotection was attenuated by adenosine A$_1$ receptor antagonist\textsuperscript{11,16} and PKC inhibitors.\textsuperscript{25} In addition, it has recently been shown that activation of G$_i$ proteins was implicated in isoflurane-induced preconditioning.\textsuperscript{26} Further studies are needed to determine the precise effects of volatile anesthetics on K$_{ATP}$ channels and signaling pathways leading to the preconditioned state.

This is the first study showing that specific blockade of adenosine A$_1$ receptors with DPX abolishes the desflurane-enhanced postischemic recovery of force. Previous results showed that cardioprotection conferred by isoflurane was mediated through activation of adenosine A$_1$
receptors, suggesting a role of adenosine in volatile anesthetic-induced preconditioning. However, further studies are required to elucidate the mechanisms through which volatile anesthetics interact with adenosine receptors.

Our findings show that specific blockade of α and β adrenoceptors abolishes the desflurane-enhanced post-ischemic contractile function recovery. These results strongly suggest that stimulation of α and β adrenoceptors plays a role in desflurane-induced preconditioning. In contrast to other volatile anesthetics, desflurane has been reported to induce sympathetic activation in healthy volunteers but also to release intrinsic store of catecholamines in isolated rat heart and human myocardium. A growing body of evidence suggests that stimulation of α1 adrenoceptors could mediate ischemic preconditioning in human myocardium. However, Loubani et al. showed that activation of α1 adrenoceptors before ischemia is protective but is detrimental during ischemia. Recently, involvement of the β-adrenergic signal transduction pathway in ischemic preconditioning has been suggested, and isoproterenol has been shown to precondition isolated rat heart through activation of PKC.

The main advantage of isolated human preparations in studying myocardial preconditioning is that the effect of variable myocardial collateral flow, which may occur in in vivo models, could be eliminated. However, our results must be interpreted within the constraints of several possible limitations. First, the effects of anesthetics drugs, diseases, or treatments received by the patients cannot be eliminated. Therefore, patients taking oral hypoglycemic medications were excluded from the study. Furthermore, we have previously reported that preoperative treatment, such as β-adrenergic blocking drugs, do not mask desflurane-induced adrenoceptor stimulation. The use of isoflurane and opioids during anesthesia of patients included in this study could have theoretically precondition the appendage. However, in vitro studies were initiated at least 90 min after removal of the atrial appendage. Most importantly, comparisons have been made with control experiments. Nevertheless, a superimposed effect of opioids or isoflurane used during the surgical procedure cannot be ruled out. Second, rather than the true ischemia obtained by coronary occlusion, we used 30 min of anoxic superfusion to simulate ischemia. However, it has been shown in various experimental models that anoxia is as effective as ischemia in inducing preconditioning. Third, we measured posts ischemic contractile function recovery but not infarct size. However, it has been shown that the improved recovery of contractile function produced by preconditioning was proportional to reduced infarct size. In addition, our results, as well as previous ones, showed that this model provides a useful tool to study the mechanisms involved in ischemic preconditioning in human myocardium.

Fourth, our experiments were performed at 34°C, which may have decreased KATP channel sensitivity and the effect of preconditioning. However, during surgical procedures, moderate hypothermia may occur in patients.

In conclusion, desflurane exerts a cardioprotective effect in anoxia-challenged isolated human right atrial myocardium. This effect involves, at least in part, mitochondrial KATP channels, and stimulation of adenosine A1 receptor and α and β adrenoceptors.

References
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