Effect of ATP-Sensitive Potassium Channel Inhibition on Resting Coronary Vascular Responses in Humans

H. M. Omar Farouque, Stephen G. Worthley, Ian T. Meredith, R. Andrew P. Skyrme-Jones, Michael J. Zhang

Abstract—Experimental data suggest that vascular ATP-sensitive potassium (K\textsubscript{ATP}) channels regulate coronary blood flow (CBF), but their role in regulating human CBF is unclear. We sought to determine the contribution of K\textsubscript{ATP} channels to resting conduit vessel and microvascular function in the human coronary circulation. Twenty-five patients (19 male/6 female, aged 56±12 years) were recruited. Systemic and coronary hemodynamics were assessed in 20 patients before and after K\textsubscript{ATP} channel inhibition with graded intracoronary glibenclamide infusions (4, 16, and 40 \textmu g/min), in an angiographically smooth or mildly stenosed coronary artery following successful elective percutaneous coronary intervention to another vessel. Coronary blood velocity was measured with a Doppler guidewire and CBF calculated. Adenosine-induced hyperemia was determined following bolus intracoronary adenosine injection (24 \textmu g). Time control studies were undertaken in 5 patients. Compared with vehicle infusion (0.9% saline), glibenclamide reduced resting conduit vessel diameter from 2.5±0.1 to 2.3±0.1 mm (P<0.01), resting CBF by 17% (P=0.05), and resting CBF corrected for rate pressure-product by 18% (P=0.01) in a dose-dependent manner. A corresponding 24% increase in coronary vascular resistance was noted at the highest dose (P<0.01). No alteration to resting CBF was noted in the time control studies. Glibenclamide reduced peak adenosine-induced hyperemia (P=0.01) but did not alter coronary flow reserve. Plasma insulin increased from 5.6±1.2 to 7.6±1.3 mU/L (P=0.02); however, plasma glucose was unchanged. Vascular K\textsubscript{ATP} channels are involved in the maintenance of basal coronary tone but may not be essential to adenosine-induced coronary hyperemia in humans. (Circ Res. 2002;90:1646–1655.)

Key Words: blood flow ■ potassium channels ■ sulfonylurea ■ vasoconstriction ■ coronary flow reserve

Membrane-bound adenosine triphosphate-sensitive potassium (K\textsubscript{ATP}) channels were first identified by Noma in cardiac myocytes. They have since been discovered in numerous cell types including pancreatic \ beta -cells, vascular smooth muscle cells, and arterial endothelial cells. These channels are regulated by the cellular metabolic state and are selectively inhibited by sulfonylurea derivatives such as glibenclamide, which are widely used in the treatment of type 2 diabetes mellitus.

K\textsubscript{ATP} channel activation results in membrane hyperpolarization, which in vascular smooth muscle cells leads to vasorelaxation. In mediating vascular smooth muscle cell membrane potential, K\textsubscript{ATP} channels provide a means by which cellular metabolism can be linked to vascular tone. In the coronary circulation, local myocardial metabolism exerts the most important influence in regulating coronary blood flow (CBF). K\textsubscript{ATP} channels in coronary resistance vessels appear to be an important intermediary in this process. Experimental studies in the coronary circulation of different animal species have indicated that K\textsubscript{ATP} channels are involved in mediating basal tone, reactive hyperemia, hypoxic vasodilation, and adenosine-induced vasodilation, as reflected by the fact that these processes are inhibited by pretreatment with glibenclamide. Relatively little data are available on the role of K\textsubscript{ATP} channels in human vasculature. Recent clinical studies suggest that K\textsubscript{ATP} channels may contribute to peripheral blood flow regulation. However, the role of K\textsubscript{ATP} channels in regulating human CBF is not known. Thus, we examined the contribution of K\textsubscript{ATP} channels to resting conduit vessel and microvascular function in the human coronary circulation.

Materials and Methods

Patient Selection

Patients undergoing elective percutaneous coronary intervention (PCI) were considered for the study. Suitable patients were required to have one angiographically smooth or mildly stenosed (<20% diameter stenosis) major epicardial coronary artery that had not previously been instrumented. All coronary flow studies were performed in a coronary artery fulfilling these criteria, after successful percutaneous single vessel intervention to an adjacent but
TABLE 1. Baseline Characteristics of the Study Population

<table>
<thead>
<tr>
<th>Variable</th>
<th>Gilbenclamide (n=20)</th>
<th>Time Control (n=5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>55±12</td>
<td>56±15</td>
</tr>
<tr>
<td>Gender (M/F)</td>
<td>16/4</td>
<td>3/2</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>28±3</td>
<td>27±4</td>
</tr>
<tr>
<td>Coronary risk factors (No. of patients)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>5 (25%)*</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Hypertension</td>
<td>9 (45%)</td>
<td>3 (60%)</td>
</tr>
<tr>
<td>Current smoker</td>
<td>1 (5%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>Hypercholesterolemia</td>
<td>19 (95%)</td>
<td>4 (80%)</td>
</tr>
<tr>
<td>Family history of IHD†</td>
<td>9 (45%)</td>
<td>3 (60%)</td>
</tr>
<tr>
<td>Number of risk factors per patient‡</td>
<td>2.9±1.1</td>
<td>2.8±0.4</td>
</tr>
<tr>
<td>Total cholesterol, mmol/L</td>
<td>4.5±1.0</td>
<td>3.5±0.8#</td>
</tr>
<tr>
<td>LDL cholesterol, mmol/L§</td>
<td>3.0±0.9</td>
<td>2.0±0.5#</td>
</tr>
<tr>
<td>Left ventricular ejection fraction, %¶</td>
<td>69±8</td>
<td>66±8</td>
</tr>
</tbody>
</table>

Data are expressed as mean±SD.

*These patients had type 2 diabetes mellitus, with an HbA1C of 7.2±1.3%.
†IHD indicates ischemic heart disease; ‡includes the 5 mentioned risk factors, obesity (body mass index >30 kg/m²), and age >60 years; §LDL indicates low-density lipoprotein; ¶left ventricular ejection fraction was calculated by quantitative left ventriculography; #P<0.05 vs the glibenclamide group.

Study Protocol

Vasoactive medication was discontinued at least 24 hours before the procedure. All patients received oral doses of aspirin (300 mg) and clopidogrel (300 mg) in preparation for PCI. Heparin (70 to 100 U/kg) was administered intravenously before coronary instrumentation. Following successful and uncomplicated PCI, a 0.014-inch Doppler guidewire (FloWire, Cardiometrics, EndoSonics) was advanced into the study artery and positioned in its proximal or mid-segment to obtain stable Doppler flow velocity signals. A 2.8F infusion catheter (Tracker, Target Therapeutics, Boston Scientific) for intracoronary drug administration was passed over the Doppler guidewire and positioned proximal to its tip. Resting blood flow velocity and coronary diameter were determined by quantitative coronary angiography. Images were analyzed off-line from end-diastolic frames using an automated edge-detection program (QCA-CMS version 4.1, MEDIS medical imaging systems) by an individual blinded to the study phase (S.G. Worthley). Mean coronary diameter was measured 5 mm distal to the tip of the Doppler guidewire over a 5-mm segment. Doppler indices, hemodynamic data (systemic blood pressure, heart rate), and the ECG were recorded continuously during the study and analyzed off-line as described previously. Heart rate, blood pressure, and average peak velocity (APV) were obtained from a minimum of 10 cardiac cycles. Coronary blood flow (ml/min) was calculated using the formula $I:\cdot \text{APV} \cdot 0.125 \cdot \text{coronary diameter}$. The rate-pressure product (RPP, bpm · mm Hg), an index of myocardial workload, was computed by multiplying heart rate and systolic blood pressure. Coronary blood flow was corrected for RPP by dividing CBF by the RPP. The calculation of peak adenosine-induced hyperemia was based on the assumption that conduit vessel diameter does not alter significantly in the brief period taken before maximum vasodilation is achieved. Coronary vascular resistance (CVR, mm Hg/ml/min) was calculated as the quotient of mean arterial blood pressure (MABP) and CBF.

Statistical Analysis

Baseline characteristics are presented as mean±SD; other values are reported as mean±SEM. Coronary vascular and hemodynamic data were tested for normality using the Kolmogorov-Smirnov test. Variables that were not normally distributed were transformed using the Box-Cox procedure. Changes in coronary vascular responses and systemic variables within each group were analyzed using repeated measures analysis of variance followed by post hoc testing as appropriate. Humoral parameters were compared using the paired
Student’s t test. Statistical significance was determined at a value of P<0.05.

Results

Safety and Systemic Effects of Glibenclamide
All coronary studies were completed without complication. The study vessel was the left anterior descending coronary artery in 10 patients, the circumflex coronary artery in 9 patients, and the right coronary artery in 1 patient. Intracoronary glibenclamide did not result in symptomatic or electrocardiographic evidence of myocardial ischemia. There was a small increase in MABP ($P=0.008$, Table 2) and an associated reduction in heart rate ($P=0.03$; Table 2) with increasing doses of glibenclamide. Compared with vehicle infusion, glibenclamide did not significantly alter RPP at any of the 3 doses used. Plasma insulin levels increased slightly (5.6±1.2 to 7.6±1.4 mU/L; $P=0.02$) during the study, but there was no change in plasma glucose (5.6±0.4 to 5.7±0.3 mmol/l; $P=NS$).

Effect of Glibenclamide on Conduit Vessel Diameter
Glibenclamide infusions at 4, 16, and 40 μg/min elicited a vasoconstrictor response in the study vessel compared with vehicle infusion. A dose-response relationship was noted, with the highest glibenclamide dose having the greatest vasoconstrictor effect (2.50±0.10; 2.43±0.10; 2.40±0.11; and 2.32±0.11 mm; $P=0.001$, Figure 1). Compared with baseline, the percentage change in diameter at each glibenclamide dose was 2.8%, 4%, and 7.2%. The difference in epicardial diameter was due to a significant vasoconstrictor effect at each dose of glibenclamide (Figure 1).

Effect of Glibenclamide on Coronary Microvascular Function
There was a strong trend to reduction in resting CBF with glibenclamide (32.9±5.3 versus 29.7±4.7 versus 28.4±3.8 versus 27.4±3.6 mL/min, $P=0.05$; Figure 2), which reached statistical significance when corrected for RPP ($P=0.01$; Figure 2). Glibenclamide also produced a graded increase in CVR ($P=0.006$, Figure 2), with the highest dose resulting in a 24% increase in CVR compared with vehicle infusion. Compared with vehicle, peak adenosine-induced CBF declined with glibenclamide infusion ($P=0.01$, Table 3). Baseline APV and maximal adenosine-induced APV were similar during vehicle infusion and after each of the glibenclamide doses used. Although peak adenosine-induced CBF was attenuated by glibenclamide, CFR did not decrease as basal CBF was proportionately reduced (Table 3). The duration of the vasodilator response was similar with vehicle or glibenclamide.

Time Control Study
The study vessel was the left anterior descending coronary artery in 1 patient and circumflex coronary artery in 4 patients. In this group, MABP and RPP were unchanged during the time course of the study ($P=NS$). There was no alteration in epicardial diameter (2.62±0.21 versus

![Figure 1](http://circres.ahajournals.org/)

Figure 1. Effect of glibenclamide on conduit vessel diameter. Glibenclamide decreased resting coronary diameter in a dose-dependent manner. Glib 1, 2, and 3 indicate intracoronary glibenclamide infusions at 4, 16, and 40 μg/min, respectively. *$P<0.05$ vs vehicle.

<table>
<thead>
<tr>
<th>TABLE 2. Systemic Hemodynamic Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parameter</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
</tr>
<tr>
<td>Blood pressure, mm Hg</td>
</tr>
<tr>
<td>SBP</td>
</tr>
<tr>
<td>DBP</td>
</tr>
<tr>
<td>MABP</td>
</tr>
<tr>
<td>RPP, bpm · mm Hg</td>
</tr>
</tbody>
</table>

Glib 1, 2, and 3 indicate intracoronary glibenclamide infusions at 4, 16, and 40 μg/min, respectively. bpm indicates beats per minute; SBP, systolic blood pressure; DBP, diastolic blood pressure; MABP, mean arterial blood pressure; RPP, rate-pressure product; CBF, coronary blood flow; and CVR, coronary vascular resistance.
2.64±0.21 versus 2.61±0.18 versus 2.61±0.20 mm; P=NS), CBF (30.5±4.3 versus 31.2±4.3 versus 31.3±4.2 versus 30.7±3.7 mL/min; P=NS), or CVR (3.7±0.7 versus 3.6±0.6 versus 3.6±0.6 versus 3.6±0.6 mm Hg/mL/min; P=NS).

Discussion
To our knowledge, this is the first report to examine the acute effect of glibenclamide, an inhibitor of K<sub>ATP</sub> channels, on human coronary vascular responses. We have demonstrated that under resting conditions, glibenclamide produces an incremental reduction in coronary conduit vessel diameter in a dose-dependent manner. Our findings also indicate that K<sub>ATP</sub> channel inhibition has a moderate effect on coronary microvascular function as evidenced by a reduction in resting CBF and an increase in CVR. Furthermore, glibenclamide reduced peak adenosine-induced vasodilation but not CFR. These changes occurred in the absence of significant alteration to myocardial workload or clinical evidence of ischemia. The results suggest that K<sub>ATP</sub> channels are active under resting conditions in the intact human coronary circulation and are involved in mediating basal coronary vascular responses but may not be essential to adenosine-induced coronary vasodilation.

K<sub>ATP</sub> Channels and Basal Coronary Tone
There is good evidence from experimental studies that K<sub>ATP</sub> channels contribute to basal coronary vascular tone. In the isolated perfused rabbit heart and anesthetized open-chest

### Table 3. Effect of Adenosine on Coronary Hemodynamics

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Vehicle</th>
<th>Glib 1</th>
<th>Glib 2</th>
<th>Glib 3</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline CBF, mL/min</td>
<td>32.9±5.3</td>
<td>29.7±4.7</td>
<td>28.4±3.8</td>
<td>27.4±3.6</td>
<td>0.05</td>
</tr>
<tr>
<td>Peak CBF, mL/min</td>
<td>81.1±9.6</td>
<td>77.1±9.7</td>
<td>72.3±7.7</td>
<td>66.8±7.8</td>
<td>0.01</td>
</tr>
<tr>
<td>CFR</td>
<td>2.9±0.2</td>
<td>3.0±0.2</td>
<td>2.9±0.2</td>
<td>2.8±0.2</td>
<td>NS</td>
</tr>
<tr>
<td>Duration of vasodilation, seconds</td>
<td>36.4±2.5</td>
<td>37.5±2.4</td>
<td>38.0±2.2</td>
<td>38.0±2.6</td>
<td>NS</td>
</tr>
</tbody>
</table>

*Glib 1, 2, and 3 indicate intracoronary glibenclamide infusions at 4, 16, and 40 μg/min, respectively.*
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not appear to play a prominent role in controlling resting blood flow. Most experimental studies suggest that its coronary vasodilator action may in part be mediated through K<sub>ATP</sub> channels. There is less evidence to support the alternate view that K<sub>ATP</sub> channels are not involved in adenosine-induced vasodilation, with the apparent inconsistency possibly due to methodological differences. Recent investigations have helped to clarify the basic mechanisms responsible for adenosine-induced coronary vasodilation. These studies suggest that adenosine may activate endothelial and smooth muscle K<sub>ATP</sub> channels by binding to the adenosine A<sub>2A</sub> receptor.

There is a paucity of data on the role of K<sub>ATP</sub> channels in adenosine-induced vasodilation in the human circulation. A study in the forearm circulation found that adenosine-induced vasodilation was not attenuated by the K<sub>ATP</sub> channel inhibitor, tolbutamide. In our study, coronary K<sub>ATP</sub> channel inhibition resulted in a diminution of the peak vasodilator response to adenosine. However, the observation that CFR did not change significantly with glibenclamide infusion suggests that K<sub>ATP</sub> channels are not critical to the vasodilator response to adenosine and that other mechanisms are involved. Taken together, these findings raise the possibility of species-related heterogeneity in the pathways responsible for adenosine-induced vasodilation.

### Glibenclamide and K<sub>ATP</sub> Channel Inhibition

It is well established that glibenclamide is a specific inhibitor of K<sub>ATP</sub> channels. This agent has been used extensively in animal studies to examine the role of K<sub>ATP</sub> channels in controlling vascular tone. Although the doses of glibenclamide used in animal studies have in general been higher than in our study, there is evidence that lower concentrations may also result in significant inhibition of vascular K<sub>ATP</sub> channels. Studies in the human forearm circulation have revealed that glibenclamide infused with the aim of attaining therapeutic regional concentrations, may attenuate the vasodilation induced by the K<sub>ATP</sub> channel opener, diazoxide. The doses of glibenclamide we used were chosen to achieve concentrations in the study vessel that were comparable with peak blood levels seen after oral administration of glibenclamide in type 2 diabetic subjects. However, the actual intracoronary concentration of glibenclamide at each dose would have been higher than initial estimates, which were based on a CBF of 80 mL/min. We found that the average CBF was approximately 30 mL/min. Recirculation of glibenclamide over the duration of infusion may also have resulted in higher intracoronary glibenclamide concentrations. Furthermore, no change in conduit vessel diameter or CBF was seen in the subgroup of patients undergoing the time control studies, providing evidence that glibenclamide was having a specific effect on the coronary circulation. We documented a small rise in plasma insulin concentrations consistent with pancreatic β-cell K<sub>ATP</sub> channel blockade. Of note, experimental evidence suggests that K<sub>ATP</sub> channels from pancreatic β-cells and coronary resistance vessels may have similar sensitivity to inhibition by glibenclamide. Although it is possible that using higher glibenclamide doses may have achieved a greater degree of vascular
K\textsubscript{ATP} channel inhibition, this would have resulted in large alterations to insulin and glucose, thereby confounding coronary hemodynamic parameters.

**Study Limitations**

Ethical considerations preclude the invasive study of patients without coronary disease, thus all patients had atherosclerotic risk factors. The effect of these processes on K\textsubscript{ATP} channel activity is unknown. It is conceivable that alteration of K\textsubscript{ATP} channel function may occur in certain disease states as has been demonstrated in animal studies.\(^{30,34}\) The coronary arteries studied were also likely to have been atherosclerotic with some degree of impaired endothelial vasodilator function. It is possible that a functional endothelium may have offset the vasoconstrictor response to K\textsubscript{ATP} channel inhibition. In this study, all subjects received aspirin prior to catheterization. Although this agent can inhibit the production of vasodilator prostanoids and affect coronary hemodynamics, these effects are prominent at aspirin doses higher than were administered in this study.\(^{23}\) Our study examined the effect of acute K\textsubscript{ATP} channel inhibition compared with chronic inhibition seen with prolonged oral therapy. The latter circumstance is more relevant to the clinical situation. There is evidence that chronic sulfonylurea therapy may lead to a reduction in the number of functional K\textsubscript{ATP} channels in pancreatic \(\beta\)-cells.\(^{35}\) These findings raise the possibility that chronic K\textsubscript{ATP} channel inhibition may also affect the function of vascular K\textsubscript{ATP} channels.

Glibenclamide is a nonselective antagonist of K\textsubscript{ATP} channels and in this sense not an ideal pharmacological modulator. This agent may interact with K\textsubscript{ATP} channels at various sites including the inner mitochondrial membrane and sarcolemma. In theory, a primary effect of glibenclamide on mitochondrial respiration, by inhibition of mitochondrial K\textsubscript{ATP} channels, could lead to secondary changes in coronary hemodynamics. However, a recent study has established that the reduction in CBF demonstrated with glibenclamide is due to primary vasoconstriction rather than a secondary effect related to a reduction in mitochondrial respiration.\(^{36}\)

**Clinical Implications**

Chronic K\textsubscript{ATP} channel inhibition with sulfonylureas for glycemic control represents the mainstay of pharmacological therapy in patients with type 2 diabetes mellitus. It has been suggested that this class of drug may be associated with increased cardiovascular events in diabetic patients with coronary disease.\(^{37–39}\) However, this remains a contentious issue with other studies refuting these claims.\(^{40,41}\) At the present time, it is unclear if this class of drug is free of clinically significant cardiovascular effects. The available data indicates that activation of K\textsubscript{ATP} channels leads to beneficial vascular effects and myocardial responses such as ischemic preconditioning. Inhibition of K\textsubscript{ATP} channels in this setting may therefore be potentially disadvantageous.\(^{42}\) Our results suggest that K\textsubscript{ATP} channel inhibition may affect coronary vascular responses in patients with underlying coronary disease. An understanding of the contribution of K\textsubscript{ATP} channels in modulating blood flow may have important ramifications for the management of patients with acute or chronic myocardial ischemia.

**Conclusion**

We provide evidence for the involvement of vascular K\textsubscript{ATP} channels in regulating resting coronary tone in humans. This was manifest as a reduction in conduit vessel diameter, CBF, and an increase in CVR. Our results also suggest that K\textsubscript{ATP} channels may not be essential to adenosine-induced coronary vasodilation. The role of these channels in human coronary metabolic vasodilation remains to be determined.

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**References**


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