The protective effects of preconditioning decline in aged patients undergoing coronary artery bypass grafting

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**Objective:** We sought to investigate the effects of myocardial ischemic preconditioning in adult and aged patients undergoing coronary artery bypass grafting.

**Methods:** Eighty patients with 3-vessel disease undergoing coronary artery bypass grafting were randomized into one of the following groups: adult ischemic preconditioning, adult control, aged ischemic preconditioning, and aged control. Hemodynamic data and cardiac troponin I values were compared between the groups. The ischemic preconditioning groups received 2 periods of 2 minutes of ischemia, followed by 3 minutes of reperfusion. The Student $t$ test, $\chi^2$ test, and analysis of variance for repeated measures were used for the statistical analysis.

**Results:** The baseline for right ventricular ejection fraction and cardiac index was similar. Right ventricular ejection fraction was depressed after the operation in all groups. Ischemic preconditioning significantly improved the recovery of right ventricular ejection fraction and cardiac index after the operation in adult patients ($P = .013$ and .001, respectively), but in the aged group there was no difference in the changes of ejection fraction and cardiac index ($P = .232$ and .889, respectively). The cardiac troponin I value in the adult patients subjected to ischemic preconditioning was lower than that in the adult control subjects ($P = .046$), but in aged patients undergoing ischemic preconditioning, the value was similar to that in aged control subjects ($P = .897$). Ischemic preconditioning also resulted in a shorter postoperative mechanical ventilation time and in less inotropic use in the adult group.

**Conclusion:** Ischemic preconditioning protects the heart from ischemic reperfusion injury in adult patients undergoing coronary artery bypass grafting. The beneficial effects of ischemic preconditioning are manifested as a better recovery of right ventricular and global hemodynamic function, cellular viability, and surgical outcome. The protective effect of ischemic preconditioning is diminished in aged patients undergoing coronary bypass.

Myocardial ischemic preconditioning (IP) has been extensively studied in various kinds of experimental models, as well as in human subjects. It has proved a potent endogenous factor in preserving high-energy phosphates, suppressing arrhythmias, delaying myocardial infarction, and improving posts ischemic functional recovery\textsuperscript{1,2} Different experimental models and conditions simulating cardiac surgery have also been investigated\textsuperscript{2,3} Published reports on the IP protective effects in the aged heart are controversial. Animal model studies show that the effect is lost in the senescent\textsuperscript{4} or even middle-
aged rat heart,\textsuperscript{5} whereas in senescent sheep (5.7-8.0 years) the response is well conserved.\textsuperscript{6} Abete and colleagues\textsuperscript{7} found that the angina-induced IP protection against myocardial infarction was lost in aged patients. Warm-up phenomenon, a counterpart of IP, can be observed in adult, but not in older, patients.\textsuperscript{8} A recent report shows that exercise-induced IP protects the following ischemia, but it only occurs in adult, and not in elderly, patients; this reinforces the hypothesis of an age-related reduction of IP in the aged heart.\textsuperscript{9} However, Kloner and colleagues\textsuperscript{10} have shown that the protective effect of preinfarct angina to the subsequent myocardial infarction persists also in patients aged 60 years or older.

More recently, articles report the application of IP in cardiopulmonary bypass (CPB) surgery. The outcome, however, has been controversial. Differences in study protocols and patients might be the reason for the discrepancies (Table 1).\textsuperscript{11-21} No investigation of the age-related aspects of the IP phenomenon has been made in human cardiac surgery to our knowledge. The aim of the present study was to investigate the myocardial performance after IP in differently aged patients undergoing coronary artery bypass grafting (CABG).

### Materials and Methods

The study design was accepted by the Ethical Committee of Tampere University Hospital, Finland, and informed consent was obtained from all patients.

### Patients

Eighty patients with 3-vessel coronary artery disease undergoing CABG were randomized into an IP group (n = 40) and a control group (n = 40). According to the median age (68 years), patients were further divided into an adult IP group and an adult control group (<68 years) and an aged IP group and an aged control (≥68 years). Patients with low ejection fractions (<35\%), recent myocardial infarction (<3 months), additional cardiac diseases, severe noncardiac diseases, and calcified or severely dilated ascending aortas were excluded.

The preoperative data of the patients were similar between the IP and the control groups. There were no statistically significant differences in age, sex, New York Heart Association class, left ventricular ejection fraction, vessel diseases, previous myocardial infarction, left ventricular hypertrophy, diabetes, or preoperative medication use between the 2 groups. The numbers of vessels bypassed, cross-clamping time, and CPB time were also similar (Table 2).

### Preconditioning Protocol

After establishing CPB and running the pump to empty the heart, the ascending aorta was occluded by means of crossclamping for 2 minutes, followed by 3 minutes of reperfusion; the procedure was repeated once. In the control group the pump was also run for 10 minutes before routine operation. Normothermia was maintained during the procedure.

### Anesthesia, CPB, and Surgical Technique

A standardized anesthetic technique was used with sufentanil, midazolam, and pancuronium bromide. CPB with nonpulsatile perfusion flow (2.2-2.4 L · min\textsuperscript{-1} · m\textsuperscript{2}) was conducted by using membrane oxygenators with arterial line filtration. Mild hypothermia (32°C) was maintained without topical cooling. Blood from the pump reservoir was mixed with crystalloid in a ratio of 4:1, yielding a cardioplegia solution with a 0.21 hematocrit value and a 21 mmol/L potassium concentration in the initial dose and 9 mmol/L in subsequent doses. In antegrade delivery cardioplegia was administered at a pressure of 80 mm Hg, and in retrograde delivery it was administered at a pressure of 30 to 40 mm Hg, with a flow of at least 200 mL/min. The initial high-potassium cardioplegia was given for 1.5 minutes in an antegrade fashion and then for 2.5 minutes in a retrograde fashion at a temperature of 6°C to 9°C. Reinfusion of cardioplegia was administered for 1 minute.
with retrograde delivery and to vein grafts after completion of each distal vein graft anastomosis. Warm cardioplegia (37°C) was given in a retrograde fashion for 3 minutes before crossclamp release. Surgical techniques were the same in all cases. Aortic root and 2-stage single venous cannulas were used for CPB. Distal anastomoses were made in the order of right coronary artery–circumflex artery–left anterior descending artery. The proximal anastomoses were constructed in a reverse order and during aortic crossclamping. The left anterior descending artery was grafted with the left internal thoracic artery in all patients.

Hemodynamic Measurements and Treatment
Hemodynamic data were collected at 4 time points: (1) baseline (before induction of anesthesia); (2) 1 hour after declamping; (3) 6 hours after declamping; and (4) on the first postoperative day (POD). Heart rate (HR), mean arterial pressure (MAP), central venous pressure (CVP), mean pulmonary artery pressure (MPAP), pulmonary capillary wedge pressure (PCWP), cardiac output (CO), and right ventricular ejection fraction (RVEF) were monitored. Derived cardiovascular variables, including cardiac index (CI) and right ventricular end-diastolic volume index (RVEDVI), were calculated with standard formulas. All measurements based on the thermodilution technique were made at end expiration in triplicate with ice-cold saline solution. The mean value of 3 consecutive measurements at each time point was registered.

Perioperatively, volume infusion was designed to maintain filling pressures at least at the preoperative level and optimal for heart performance. Inotropes (dopexamine or adrenaline) were used to maintain the CI above 2.0 L·min⁻¹·m⁻². Amrinone with noradrenaline was used when dopexamine or adrenaline proved insufficient to maintain the CO. They were applied after release of crossclamping and continued for at least 6 hours. Inotropes were not discontinued at the time points when hemodynamic data were measured.

Cardiac Troponin I
Blood samples were collected from peripheral vessels (1) before CPB as baseline, (2) 5 minutes after declamping, (3) 6 hours after declamping, (4) at the first POD, and (5) at the second POD. Samples were collected in heparin-coated plastic tubes and centrifuged. Serum samples were measured with a Chiron ACS180 analyzer (Chiron Diagnostics Corp, East Walpole, Mass) by using a direct chemiluminescence method.

Statistics
The 2-sample Student t test (2-tailed) was used for continuous data. The χ² or Fisher exact tests were used for categoric data when comparing variables between the IP and control groups in the adult or aged patients. Analysis of variance (ANOVA) for repeated measures was used to test repeated observation variables after the operation. Data are presented as means ± SD. Statistical analyses were made with an SPSS/Windows (version 9.0) statistical package program.

Results
Hemodynamics in Adult Patients
The hemodynamic variables, namely HR, MAP, CVP, MPAP, PCWP, and RVEDVI, were similar between the IP and control groups (Table 3). There was no difference between the IP and control groups in the baseline level of RVEF (41.2% ± 6.2% vs 44.2% ± 7.0%, P = .167, t test). The baseline level of CI in the control group had a slight tendency to be higher than that in the IP patients (2.39 ± 0.37 vs 2.68 ± 0.52 L·min⁻¹·m⁻² in the IP and control groups, respectively; P = .058, t test). RVEF was depressed after the operation in both groups, with the depression being milder in the IP group (P = .013, ANOVA for repeated measures; Figure 1). IP resulted in better recovery of CI after the operation (P = .001, ANOVA for repeated measures; Figure 2).

Hemodynamics in Aged Patients
The hemodynamic variables, namely HR, MAP, CVP, MPAP, PCWP, and RVEDVI, were similar between the IP and control groups (Table 3). The baseline levels of RVEF and CI were similar between the IP and the control groups (41.2% ± 6.0% vs 41.0% ± 6.3%, P = .895 and 2.47 ± 0.31 vs 2.41 ± 0.28 L·min⁻¹·m⁻², P = .519, respectively, t test). In the IP group there was a slight tendency toward better recovery in RVEF immediately after the operation, but it was not statistically significant (P = .126, ANOVA for repeated measures; Figure 1). IP did not induce better recovery of CI after the operation (P = .889, ANOVA for repeated measures; Figure 2).
Cardiac Troponin I

The baselines of serum cardiac troponin I (CTnI) in the groups were similar (0.19 ± 0.04 vs 0.19 ± 0.08 µg/L in the adult IP and control groups, \( P = .966 \), and 0.23 ± 0.08 vs 0.15 ± 0.03 µg/L in the aged IP and control groups, \( P = .360 \), respectively, \( t \) test). There was a massive release of CTnI after the operation, and this did not recover until the second POD. The CTnI values in adult patients undergoing IP were lower than those in adult control subjects (\( P = .046 \), ANOVA for repeated measures), whereas there was no statistically significant difference in the CTnI values between aged patients undergoing IP and control patients (\( P = .897 \), ANOVA for repeated measures; Figure 3).

Surgical Outcome

The period of mechanical ventilation was significantly shorter in adult patients undergoing IP than in adult control subjects (\( P = .030 \), \( t \) test). There was no statistical significance in the period of mechanical ventilation between the aged IP and the aged control subjects (\( P = .187 \), \( t \) test). The length of stay in the intensive care unit was similar between the groups. More patients in the adult group undergoing IP were free of inotropes (\( P = .043 \), Fisher exact test), and the period of inotropic medication was also shorter than that in adult control subjects (\( P = .031 \), \( t \) test). The difference in inotropic support in the aged patients was similar in those undergoing IP and the control subjects (Table 4). There was no perioperative myocardial infarction. No intra-aortic balloon pump was used.

Discussion

Myocardial IP has been extensively studied in various kinds of experimental models, as well as in human subjects. However, reports on the IP effects in cardiac surgery are few in number. There are several studies showing that IP is effective in preserving high-energy phosphate,12-14 improving heart performance,14-16 reducing CTnI release,17,18 and increasing myocardial superoxide dismutase and decreasing malondialdehyde activity16 during cardiac surgery. There are also controversial reports as to the safety and effectiveness of IP during CPB surgery. Difference in the IP protocol and study subjects might be the reason for discrepancy. Age-
related factors may also play an important role. As far as we know, the possible relation of the controversial result of IP in cardiac surgery has not been studied.

Our present findings suggested that preconditioning with 2 periods of 2-minute ischemia, followed by 3 minutes of reperfusion, was cardioprotective during CABG operations with cold-blood cardioplegia in adult patients. These beneficial effects of IP included better recovery of right ventricular function, improved global hemodynamic performance after the operation, and preserved cellular viability. There are controversial reports as to whether IP is a phenomenon confined to the young myocardium. Our results suggested that the improved hemodynamic outcome and cellular viability were indeed closely associated with the age of the patients undergoing IP. It was clearly demonstrated that the protective IP effects were not present in aged patients undergoing CABG, which is in accord with previous observations of loss of IP in the aged heart.

The precise mechanism of IP remains unknown. Brief episodes of myocardial ischemia result in the production of adenosine, norepinephrine, free radicals, and bradykinin. These chemical factors act on one or more types of myocyte receptor (eg, adenosine A1, muscarinic receptor, and α sym pathetic receptor) and result in translocation and activation of tyrosine kinase and protein kinase C to the cellular membrane, working with inhibitory G-protein to subsequently

### TABLE 3. Hemodynamic data

<table>
<thead>
<tr>
<th>Group</th>
<th>Baseline</th>
<th>1 h declamping</th>
<th>6 h declamping</th>
<th>First POD</th>
</tr>
</thead>
<tbody>
<tr>
<td>HR (beats/min)</td>
<td>Adult IP</td>
<td>56.0 ± 11.7</td>
<td>78.5 ± 14.6</td>
<td>86.7 ± 14.4</td>
</tr>
<tr>
<td></td>
<td>Adult control</td>
<td>60.9 ± 10.1</td>
<td>77.8 ± 17.8</td>
<td>87.0 ± 27.1</td>
</tr>
<tr>
<td></td>
<td>Aged IP</td>
<td>55.7 ± 6.9</td>
<td>78.3 ± 17.0</td>
<td>87.5 ± 13.6</td>
</tr>
<tr>
<td></td>
<td>Aged control</td>
<td>59.1 ± 10.7</td>
<td>76.7 ± 13.8</td>
<td>86.2 ± 14.5</td>
</tr>
<tr>
<td>MAP (mm Hg)</td>
<td>Adult IP</td>
<td>87.1 ± 14.5</td>
<td>76.8 ± 9.7</td>
<td>80.2 ± 12.8</td>
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<tr>
<td></td>
<td>Adult control</td>
<td>94.0 ± 13.5</td>
<td>78.9 ± 6.7</td>
<td>76.1 ± 11.3</td>
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<tr>
<td></td>
<td>Aged IP</td>
<td>88.0 ± 12.3</td>
<td>75.1 ± 9.1</td>
<td>75.9 ± 12.4</td>
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<tr>
<td></td>
<td>Aged control</td>
<td>89.4 ± 14.3</td>
<td>75.7 ± 14.3</td>
<td>79.4 ± 11.2</td>
</tr>
<tr>
<td>CVP (mm Hg)</td>
<td>Adult IP</td>
<td>6.6 ± 2.6</td>
<td>9.2 ± 1.9</td>
<td>10.0 ± 2.7</td>
</tr>
<tr>
<td></td>
<td>Adult control</td>
<td>8.1 ± 2.2</td>
<td>9.4 ± 2.9</td>
<td>10.3 ± 2.8</td>
</tr>
<tr>
<td></td>
<td>Aged IP</td>
<td>8.1 ± 2.7</td>
<td>10.2 ± 2.0</td>
<td>10.3 ± 2.7</td>
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<tr>
<td></td>
<td>Aged control</td>
<td>7.8 ± 2.9</td>
<td>9.9 ± 2.4</td>
<td>11.1 ± 3.3</td>
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<tr>
<td>MPAP (mm Hg)</td>
<td>Adult IP</td>
<td>19.0 ± 7.2</td>
<td>19.7 ± 8.2</td>
<td>23.1 ± 4.3</td>
</tr>
<tr>
<td></td>
<td>Adult control</td>
<td>20.3 ± 5.4</td>
<td>20.0 ± 6.0</td>
<td>23.6 ± 5.8</td>
</tr>
<tr>
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<td>Aged IP</td>
<td>20.3 ± 4.9</td>
<td>19.3 ± 3.6</td>
<td>23.1 ± 5.7</td>
</tr>
<tr>
<td></td>
<td>Aged control</td>
<td>20.6 ± 9.5</td>
<td>20.8 ± 4.4</td>
<td>23.6 ± 5.2</td>
</tr>
<tr>
<td>PCWP (mm Hg)</td>
<td>Adult IP</td>
<td>12.1 ± 3.9</td>
<td>10.4 ± 6.7</td>
<td>11.1 ± 3.1</td>
</tr>
<tr>
<td></td>
<td>Adult control</td>
<td>13.1 ± 3.5</td>
<td>11.6 ± 3.2</td>
<td>11.2 ± 3.4</td>
</tr>
<tr>
<td></td>
<td>Aged IP</td>
<td>11.4 ± 3.2</td>
<td>11.3 ± 2.2</td>
<td>10.5 ± 3.5</td>
</tr>
<tr>
<td></td>
<td>Aged control</td>
<td>12.7 ± 5.9</td>
<td>11.2 ± 2.8</td>
<td>11.4 ± 3.2</td>
</tr>
<tr>
<td>RVEDVI (mL/m²)</td>
<td>Adult IP</td>
<td>103.3 ± 14.8</td>
<td>86.9 ± 20.5</td>
<td>92.5 ± 15.6</td>
</tr>
<tr>
<td></td>
<td>Adult control</td>
<td>101.1 ± 21.8</td>
<td>82.3 ± 24.2</td>
<td>90.2 ± 24.5</td>
</tr>
<tr>
<td></td>
<td>Aged IP</td>
<td>106.1 ± 24.2</td>
<td>77.7 ± 11.1</td>
<td>92.1 ± 23.0</td>
</tr>
<tr>
<td></td>
<td>Aged control</td>
<td>100.6 ± 12.9</td>
<td>85.9 ± 20.6</td>
<td>93.9 ± 27.6</td>
</tr>
</tbody>
</table>

There were no statistically significant differences between the IP and control groups in the adult or in the aged patients. Data are presented as means ± SD.

### TABLE 4. Postoperative care

<table>
<thead>
<tr>
<th></th>
<th>Adult IP</th>
<th>Adult control</th>
<th>Aged IP</th>
<th>Aged control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mechanical ventilation (h)</td>
<td>11.3 ± 3.5</td>
<td>16.3 ± 9.4*</td>
<td>13.4 ± 3.0</td>
<td>20.1 ± 22.4</td>
</tr>
<tr>
<td>Stay in ICU (h)</td>
<td>28.2 ± 17.2</td>
<td>37.4 ± 22.5</td>
<td>40.8 ± 31.5</td>
<td>46.2 ± 41.0</td>
</tr>
<tr>
<td>Free of inotropes (n)</td>
<td>11</td>
<td>3*</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>Duration of inotropes (h)</td>
<td>6.7 ± 10.2</td>
<td>13.9 ± 9.6*</td>
<td>13.7 ± 9.6</td>
<td>21.3 ± 31.9</td>
</tr>
<tr>
<td>Dopexamine or adrenaline (n)</td>
<td>5</td>
<td>10*</td>
<td>10</td>
<td>15</td>
</tr>
<tr>
<td>Aminronone-noradrenaline (n)</td>
<td>9</td>
<td>10</td>
<td>11</td>
<td>10</td>
</tr>
</tbody>
</table>

Data are presented as means ± SD. ICU, Intensive care unit. *P < .05, difference between IP and control group in the same age groups.
phosphorates the adenosine triphosphate–dependent potassium channel to trigger IP response. Increased release of one agonist or stimulation by more cycles of brief ischemic stimuli can compensate for the lack of another to achieve the protective effect. Protein kinase C translocates from cytosol to the cell membrane after activation and mediates phosphorylation of target proteins, ion channels, and myofilaments to achieve the effect. The delayed cardiac adaptation is consistent with induced gene transcription and the subsequent translocation of protective molecular proteins, including proto-oncogenes, stress proteins, and antioxidant enzyme systems.2,22

The aged myocardium evinces decreased adrenergic responsiveness, altered coronary microcirculation, impaired calcium transport, and impaired excitation-contraction coupling. Its maximal oxygen uptake decreases, and the rate of this decline accelerates with age.6,7,23,24 A reduction of norepinephrine release from cardiac adrenergic terminals has been shown after ischemic reperfusion in older animals.7 It is also reported that exogenous administration of norepinephrine is able to mimic IP in the senescent heart. The reduction in norepinephrine release after transient ischemic stimulus might thus be responsible for the age-related loss of IP.9 Exercise adaptation has been shown to increase the expression of the sarcolemmal Ca++ pump protein in the rat heart. It has been found that there is a reduction in the sarcolemmal Ca++ pump activity and abnormal sarcocellular Ca++ cycling in the aged heart.24 The age-related impairment of calcium transport and excitation-contraction coupling might also result in a loss of the protective effect of IP. The myocardial activity of superoxide dismutase and the production of heat-shock protein decline with age,5,7 and these agents are known to play an important role in the IP mechanism.2,22 Therefore in the aged heart the absence of IP may be due to the absence of a series of triggers, mediators, and effectors that are involved in the IP protective mechanism.

A review of studies of IP in cardiac surgery reveals that the effect of IP might be age related. In the reports that demonstrate the protective IP phenomenon in cardiac surgery, the patients have been relatively young. The report of Illes and Swoyer15 is one with a mean patient age of over 60 years (Table 1). However, the protocol used in that study to induce the protective effects was much different from that in the other 3 studies on elderly patients with a mean age of over 60 years. It has been suggested that the IP stimuli could have caused cumulative effects in the aged myocardium, which is more vulnerable to ischemia.7 The IP stimuli needed to induce a protective effect in adult and aged patients, respectively, might therefore be different. Further studies are called for to elucidate this hypothesis.

It is noteworthy that the age definition in the age-related IP effect is relative rather than absolute. Evidence of the IP phenomenon in patients at over 60 years of age suggests that the potential for IP could still in fact remain in an aged person.10 In the present study the right ventricular function, which is the most vulnerable in patients with 3-vessel disease undergoing CABG, tended to recover better in the aged patients undergoing IP than in control subjects.

Two periods of 2-minute ischemia, followed by 3-minute reperfusion preconditioning, protects the heart from ischemic reperfusion injury in adult patients with 3-vessel coronary artery disease undergoing CABG. The beneficial effects of IP are manifested in better recovery of right ventricular and global hemodynamic function, better preserved cellular viability, and better surgical outcome. The protective effect declines in aged patients undergoing CABG.

References


