Leukocyte-depleted Continuous Blood Cardioplegia for Coronary Artery Bypass Grafting

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SUMMARY
Many cardiac surgeries are performed with blood cardioplegia. However, some studies suggest that activated neutrophils form blood cardioplegia can cause reperfusion injury. In this study we assessed myocardial protection using a leukocyte-depleted cardioplegic solution.

Patients undergoing elective coronary artery bypass grafting (CABG) with continuous blood cardioplegia were divided into two groups: the LD group, which received leukocyte-depleted blood cardioplegia \( n = 11 \); and the control group, which received nonfiltered blood cardioplegia \( n = 11 \). IL-6, IL-8, CK-MB, and troponin T were measured in the coronary sinus blood immediately after the release of the aortic cross-clamp. Cytokine concentrations were also measured upon the patient’s return to the ICU. The total dopamine and dobutamine doses, hemodynamic measurements after surgery, and the leukocyte filtration rate were also measured.

During antegrade cardioplegia infusion, leukocytes were almost completely removed (filtration rate: 85.8 ± 4.0%). However, during terminal warm cardioplegia, leukocyte removal decreased (filtration rate: 39.9 ± 7.8%). Immediately after the release of the aortic cross-clamp, plasma CK-MB and troponin T concentrations were significantly lower in the LD group \( 17.7 ± 1.9 \) U/l and \( 0.017 ± 0.002 \) ng/ml, respectively) than in the control group \( 30.3 ± 3.6 \) U/l and \( 0.072 ± 0.029 \) ng/ml, respectively). The IL-6 and IL-8 concentrations were similar in the LD group and the control group. After the return to the ICU, the CK-MB and troponin T concentrations were similar in the two groups. No significant differences were found in the total doses of dopamine or dobutamine after surgery in the two groups (99 ± 77 vs 101 ± 128 \( \mu \)g/kg/min). No significant differences were found in the hemodynamic parameters after surgery in the two groups.

In patients undergoing CABG with continuous blood cardioplegia, leukocyte-depleted blood cardioplegic solution may attenuate reperfusion injury. (Jpn Heart J 2000; 41: 425-433)

Key words: Continuous blood cardioplegia, Leukocyte-depletion, CK-MB, Troponin T, Cytokine

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Despite significant advances in myocardial protection during cardiac surgery, myocardial injury can still occur during surgery performed on compromised hearts, such as those with severe left ventricular hypertrophy or preoperative severe ischemic injury. Blood cardioplegia was developed to attenuate ischemic myocardial damage during cardioplegic arrest and to accelerate myocardial metabolic recovery.1,2) However, studies have shown that activated neutrophils present in whole blood may cause reperfusion injury.3-5) Recent studies have shown that myocardial injury can occur when the hypoxic heart is suddenly reoxygenated (as occurs with cardiopulmonary bypass) resulting in myocardial functional depression, impaired oxygenation, and increased pulmonary vascular resistance.6) To achieve more effective myocardial protection, coronary bypass grafting (CABG) has been performed using continuous blood cardioplegia. In this study, we tested whether leukocyte-depleted cardioplegia solutions provide additional myocardial protection for CABG.

**Materials and Methods**

Twenty-two patients undergoing elective CABG were divided into two groups. The LD group consisted of 11 patients in whom leukocyte-depleted blood cardioplegia was used. The control group consisted of 11 patients in whom nonfiltered blood cardioplegia was used. There were no significant differences with respect to age, number of grafts, preoperative cardiac index, or aortic cross-clamp time between the two groups (Table I). All patients underwent complete revascularization. Informed consent for this study was obtained from all patients.

The aorta was cross-clamped and the heart arrested with antegrade infusion of blood cardioplegia (volume of cardioplegia: 700 ml), containing 20 mEq/l of potassium chloride. Continuous retrograde cardioplegia was then started. Retrograde cardioplegia delivery was adjusted to maintain a mean perfusion pressure of 40 to 45 mm Hg and a perfusion flow of 200 to 250 ml/min. The potassium chloride concentration of this solu-

<table>
<thead>
<tr>
<th>Table I.</th>
<th>There were no significant differences with respect to age, number of grafts, preoperative cardiac index, or aortic cross-clamp time between the two groups.</th>
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<tbody>
<tr>
<td>LD group</td>
<td>Control</td>
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<tr>
<td>Age (years)</td>
<td>60.5 ± 3.2</td>
</tr>
<tr>
<td>Number of grafts</td>
<td>3.56 ± 0.29</td>
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<tr>
<td>Preoperative cardiac index</td>
<td>3.26 ± 0.30</td>
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<tr>
<td>Aortic cross-clamp time</td>
<td>117.82 ± 12.17</td>
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Mean ± S.E.
tion was 8 mEq/l and its temperature 20°C. Before release of the aortic cross-clamp, retrograde terminal warm cardioplegia was infused. The terminal cardioplegia perfusion pressure was 40 to 60 mm Hg, the concentration of potassium chloride was 20 mEq/l, and the infusion volume was 700 ml.

In the LD group, blood samples were obtained before and after filtration. Leukocytes were counted during antegrade cardioplegia and during terminal warm cardioplegia. IL-6, IL-8, CK-MB, and troponin T were measured in the coronary sinus blood immediately after the release of the aortic cross-clamp and sampled from the radial artery at the time that the patient was returned to the ICU. Total dopamine and dobutamine administered doses after surgeries were also recorded, and hemodynamic parameters were measured every 3 hours.

We initially used a single leukocyte filter for depletion. When continuous blood cardioplegia was used, a single filter was ineffective for leukocyte filtration. Double leukocyte filters for blood cardioplegia were placed in the cardioplegia line before blood was mixed with the cardioplegia solution.

Statistical methods: All data are expressed as the mean ± standard error (SE). Comparisons between groups were analyzed using Student’s t test for unpaired data.

### RESULTS

During antegrade cardioplegia, leukocytes were almost completely removed (filtration rate: 85.8 ± 4.0%). However, during terminal warm cardioplegia, leukocyte removal was not complete (filtration rate: 39.9 ± 7.8%) in spite of using a double filter (Figure 1). Blood samples were collected from the coronary sinus immediately after termination of the aortic cross-clamp. The plasma CK-MB concentration was significantly lower in the LD group (17.7 ± 1.9 U/l) than in the control group (30.3 ± 3.6 U/l). The plasma troponin T concentration was significantly lower in the LD group (0.017 ± 0.002 ng/ml) than in the control group (0.072 ± 0.029 ng/ml) (Figure 2).

No significant differences were noted in the IL-6 concentration (LD group: 80.8 ± 11.6 pg/ml, control group: 72.8 ± 21.3 pg/ml) or IL-8 concentration (LD group: 17.4 ± 1.45 pg/ml, control group: 21.0 ± 3.2 pg/ml) in the coronary sinus blood immediately after the release of the aortic cross-clamp (Figure 3). After the patients were returned to the ICU, the serum IL-6 and IL-8 concentrations were similar (101.9 ± 11.6 pg/ml vs
Figure 1. During antegrade cardioplegia, leukocytes were almost completely removed (filtration rate: 85.8 ± 4.0%). However, during terminal warm cardioplegia, leukocyte removal was not complete (filtration rate: 39.9 ± 7.8%) in spite of using a double filter.

Figure 2. The plasma CK-MB concentration was significantly lower in the LD group (17.7 ± 1.9 U/l) than in the control group (30.3 ± 3.6 U/l). The plasma troponin T concentration was significantly lower in the LD group (0.017 ± 0.002 ng/ml) than in the control group (0.072 ± 0.029 ng/ml).
91.7 ± 11.8 pg/ml, 22.3 ± 4.5 pg/ml vs 11.5 ± 8.6 pg/ml, respectively) (Figure 4). However, the cytokine concentrations tended to be higher in the LD group than in the control group. The CK-MB and troponin T concentration were not significantly different in the two groups after the

**Figure 3.** No significant differences were noted in the IL-6 concentration (LD group: 80.8 ± 11.6 pg/ml, control group: 72.8 ± 21.3 pg/ml) and the IL-8 concentration (LD group: 17.4 ± 1.45 pg/ml, control group: 21.0 ± 3.2 pg/ml) in the coronary sinus blood immediately after release of the aortic cross-clamp.

**Figure 4.** After the patients were returned to the ICU, the serum IL-6 and IL-8 concentrations were similar (101.9 ± 11.6 pg/ml vs 11.5 ± 8.6 pg/ml, respectively). However, the cytokine concentrations tended to be higher in the LD group than in the control group.
patients’ return to the ICU. No significant differences were noted in the total doses of dopamine and dobutamine after surgery (LD group: 99 ± 77 µg/kg/min, control group: 101 ± 128 µg/kg/min) (Figure 5). Heart rate, cardiac output, blood pressure, pulmonary artery pressure, pulmonary wedge pressure, left ventricular stroke work index and systemic vascular resistance index were not different between the two groups.

**DISCUSSION**

When myocardial injury occurs during cardiac surgery, it often results in low cardiac output states. Blood cardioplegia has been used to attenuate ischemic myocardial damage during cardioplegic arrest by accelerating myocardial metabolic recovery and preserving high energy phosphates. However, studies have shown that activated neutrophils can cause reperfusion injury and “no-reflow phenomenon”. It is therefore likely that neutrophils cause myocardial injury. Furthermore, polymorphonuclear leukocytes have been implicated as a major effector of reperfusion injury.

It has been reported that the removal of neutrophils at the time of reperfusion improves functional recovery and myocardial function. In addition, leukocyte depletion of the perfusate attenuates reperfusion injury and prevents ultrastructural damage. Conversely, other studies have shown that the neutrophil content of the stunned myocardium is not increased and that interventions that remove neutrophils do not improve
function.\textsuperscript{12,13) Previous studies have demonstrated that the use of a leukocyte removal filter in the extracorporeal circuit does not affect the clinical outcome.\textsuperscript{14) Therefore, we tested whether the use of a leukocyte-depleted cardioplegic solution improves myocardial protection. Removal of leukocytes from the perfusate has been shown to limit neutrophil accumulation,\textsuperscript{5) improve ventricular performance,\textsuperscript{15) and reduce infarct size.\textsuperscript{15,16) Sawa, et al.\textsuperscript{17) demonstrated a decreased need for inotropic support and a decreased serum creatine kinase (CK)-MB concentration in patients undergoing emergent revascularization when a leukocyte-depleted cardioplegic solution was used. In our study, the CK-MB and troponin T concentration were significantly lower in the LD group than in the control group. These results indicate that leukocyte depletion decreases myocardial damage.

We studied the postoperative hemodynamic effect of leukocyte depletion, but found no significant differences between the two groups. One report found leukocyte depletion had no effect on global myocardial function.\textsuperscript{18) The findings suggest that leukocyte depletion alone has little myocardial protective effect compared with the use of conventional cardioplegia. The lack of a significant difference in indices of global function may be due to compensatory increases in regional function in the remote, nonischemic myocardium.\textsuperscript{19) Previous studies have shown that microvessels exhibit detectable dysfunction earlier than larger epicardial vessels after ischemic injury.\textsuperscript{20) Leukocyte depletion during reperfusion has been shown to attenuate endothelial reperfusion injury.\textsuperscript{5,18} Endothelial function is mediated via nitric oxide. Nitric oxide synthesis by the endothelium inhibits platelet adhesion, neutrophil adhesion, homotypic aggregation, and superoxide formation.\textsuperscript{21} Absence of this potent vasodilator may play a more important role in the no-reflow phenomenon than extrinsic capillary compression\textsuperscript{22} or mechanical plugging.\textsuperscript{23}

The neutrophil-related cascade amplifies inflammatory activation through the release of cytokines\textsuperscript{24} and free radicals.\textsuperscript{25) Atrial IL-8 mRNA expression increases during cardiopulmonary bypass following ischemic arrest.\textsuperscript{26) In addition, increases in the IL-8 concentration correlate with the duration of cardiopulmonary bypass and coronary sinus myoglobin concentration, a marker of myocyte injury.\textsuperscript{27,28) Cytokines play an important role in both neutrophil activation and chemotaxis.\textsuperscript{29) Leukocyte-depleted blood cardioplegia may have beneficial effects by reducing the ischemia-reperfusion injury caused by cytokines and free radicals.

In this study, leukocyte depletion decreased myocardial damage. How-
ever, the cytokine concentrations tended to be higher in the LD group. During complete leukocyte removal, leukocyte-depleted blood cardioplegia was effective in providing myocardial protection. However, when the leukocytes were not removed, the leukocyte filter increased cytokine activity. Therefore, the use of the filter may be harmful when the volume of blood cardioplegia is large or when the aortic cross clamp time is long. This study used the Pall BC1B leukocyte filter (Pall Biomedical Products Corp., East Hills, NY). The leukocyte filtration capacity of a single Pall BC1B filter for whole blood is 800 ml.30) During terminal warm cardioplegia, the leukocyte filtration rate decreased to 40%. The use of a higher capacity leukocyte filter may decrease the cytokine concentration and result in more effective myocardial protection.

**Conclusion:** Leukocyte-depleted blood cardioplegic solution may attenuate reperfusion injury in patients with ischemic heart disease undergoing CABG. However, the use of a leukocyte removal filter may be associated with increased cytokine activity. Future studies should focus on enhancing myocardial protection through the use a high capacity filter for leukocytes to decrease cytokine activity.

**REFERENCES**


