Release of Serum S-100β Protein and Neuron-Specific Enolase after Off-Pump Coronary Artery Bypass Grafting with and without Intracranial and Cervical Artery Stenosis

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Purpose: The aim of this study was to quantify the amount of brain damage suffered by patients who underwent off-pump coronary artery bypass grafting (OPCAB) in which S-100β protein and neuron-specific enolase were used.

Methods: Thirty-four patients undergoing scheduled OPCAB were enrolled in the study. The patients were divided into two groups according to the results of their magnetic resonance angiography (MRA) and cervical ultrasonography: 13 patients had cervical or intracranial arterial stenosis (Group A), and 21 patients did not (Group B). Blood samples were collected from the arterial catheters immediately before surgery, upon arrival to the intensive care unit, and 6 and 24 hours after surgery.

Results: In blood samples collected from patients upon arrival to the intensive care unit, the maximum concentration of serum s-100β protein in Group A was significantly higher than that of Group B (p = 0.029). Though patients in Group A tended to have higher maximum neuron-specific enolase (NSE) concentrations, there were no significant differences in NSE concentrations at any point between the two groups.

Conclusions: Our findings show a correlation between the stenosis detected by MRA or cervical ultrasonography and brain damage after OPCAB.

Key words: off-pump coronary artery bypass grafting, neurologic complications, s-100β protein, neuron-specific enolase

Introduction

Neurologic complications remain among the most significant problems after coronary artery bypass grafting (CABG). Patients with angina pectoris frequently display coexistent cervical and intracranial arterial stenosis. Some preoperative screening measures exist, such as magnetic resonance angiography (MRA), four-vessel angiography, cervical ultrasonography, and brain scintigram. All patients scheduled for coronary artery revascularization in our hospital underwent MRA and cervical ultrasonography because of their low invasion. However, cervical ultrasonography can detect only limited numbers of lesions, and MRA may produce false positives due to blood flow changes. The correlation between arterial stenosis detectable by MRA or cervical ultrasonography and brain damage has not been well defined to date.

Sequential sampling of serum S-100β protein and neuron-specific enolase (NSE) has been used to study brain damage resulting from cardiac surgery. Previous studies have revealed that off-pump coronary artery bypass...
(OPCAB) is better than conventional CABG\textsuperscript{5–7} at reducing the incidence of neurologic complications\textsuperscript{1–4} and release of s-100\textgreek{B} protein and NSE.

We hypothesized that MRA and cervical ultrasonography could be employed to measure s-100\textgreek{B} protein and NSE concentrations.

The aim of this study was: (1) to quantify the amount of brain damage suffered by patients who underwent OPCAB, and (2) to investigate whether the stenosis detectable by MRA or cervical ultrasonography is associated with higher serum s-100\textgreek{B} protein and NSE concentrations.

Materials and Methods

Between January 2008 and November 2008, 34 patients undergoing scheduled OPCAB were enrolled in the study. Patients who were directed from OPCAB to on-pump CABG were excluded from the study. All patients underwent MRA and cervical ultrasonography preoperatively. The arterial stenosis was defined as more than 75\% stricture for both MRA and cervical ultrasonography. Roentgenologists and cardiologists interpreted the images. The 34 patients were divided into two groups according to the results of their MRA and cervical ultrasonography: 13 patients had cervical or intracranial arterial stenosis (Group A), and 21 patients did not (Group B). The mean age was 72.6 ± 8.0 years in Group A, and 62.6 ± 11.7 years in Group B.

Hypertension was defined as a blood pressure greater than 130/90 mmHg and/or a history of hypertension and antihypertensive treatment. Diabetes mellitus was defined as a fasting blood sugar greater than 125 mg/dL and/or the presence of antidiabetic treatment. Renal insufficiency was defined as serum creatinine levels 2 mg/dL or greater. Hyperlipidemia was defined as total cholesterol in blood greater than 220 mg/dL or low density lipoprotein in blood greater than 140 mg/dL and/or triglyceride in blood greater than 150 mg/dL or the presence of lipid reduction treatment.

In the operation room, typical monitoring equipment was used, and anesthetic techniques and medications used were similar in all patients. Anesthesia was induced with fentanyl, propofol, and neuromuscular paralytic drugs, and maintained with total intravenous anesthesia using the same drugs. All operations were performed without cardiopulmonary bypass (CPB). Conventional techniques with median sternotomy were used in all operations. Heparin 1.5 mg/kg was administered, and anticoagulation time (ACT) was maintained above 200 seconds. We prefer multiple and complete coronary revascularization with composite or sequential grafting, and in situ arterial grafts, in particular. When we used the saphenous vein, we adopted a pass-port system to avoid aortic side-biting clamps. We used a cell-saving device for all patients but did not use anti-fibrinolytic drugs during the operation. OPCAB was performed with one of several commercially available cardiac positioning and coronary artery stabilizing device. During the operation, blood pressure was maintained above 80 mmHg.

Blood samples were collected from the arterial catheters immediately before surgery, upon arrival to the intensive care unit, and 6 and 24 hours after surgery.

Baseline demographic and clinical data were available for all patients. We recorded the following preoperative variables: gender, age, body mass index (BMI), preoperative ejection fraction, and the presence of diabetes mellitus, hypertension, hyperlipidemia, and chronic renal failure. We recorded the following intraoperative variables: operative time, number of anastomoses, performance of blood transfusion, and the use of bilateral internal thoracic arteries.

Institutional approval for this study was obtained and each patient within the study gave informed consent to serve as a subject.

Statistical methods

All data were reviewed retrospectively. Values of all continuous variables are expressed as mean ± SD. Differences between the two patient groups were tested with univariate analysis (the \textit{\chi}\textsuperscript{2} test, the two-tailed \textit{t} test, and the Mann-Whitney U test, as appropriate). Values of \textit{p} <0.05 were considered significant.

Results

Of 34 patients, 13 (38.2\%) had cervical and intracranial arterial stenosis (Group A), and 21 did not (Group B). Clinical characteristics of the study population are shown in Table 1. Patients in Group A tended to be older (\( p = 0.01 \)). However, between these two groups, there were no significant differences in gender, preoperative ejection fraction, body surface area, history of hypertension, hyperlipidemia, diabetic mellitus, diabetic mellitus with insulin, chronic renal failure, chronic renal failure with hemodialysis, smoking history, and left main diseases.

Figure 1 shows the changes in serum NSE concentrations. In Group A, serum NSE concentrations averaged 7.5 ± 2.4 \( \mu \)g/l before surgery, 10.5 ± 4.9 \( \mu \)g/l upon arrival to the intensive care unit. They averaged 11.5 ± 2.9 and
10.5 ± 3.1 μg/l at 6 and 24 hours after surgery. In Group B, the corresponding concentrations were 6.4 ± 1.1 μg/l before surgery (p = 0.085), and 9.0 ± 2.8 μg/l upon arrival to the intensive care unit (p = 0.256). They averaged 12.3 ± 4.9 (p = 0.631), and 9.3 ± 3.6 μg/l (p = 0.309) at 6 and 24 hours after surgery.

**Figure 2** shows the changes in serum s-100β protein concentrations. In Group A, serum s-100β protein concentrations averaged 0.05 ± 0.01 μg/l before surgery, and 0.31 ± 0.18 μg/l upon arrival to the intensive care unit. They averaged 0.14 ± 0.06 μg/l, and 0.11 ± 0.14 μg/l at 6 and 24 hours after surgery. In Group B, the corresponding concentrations were 0.05 ± 0.02 μg/l before surgery (p = 0.774), and 0.19 ± 0.11 μg/l upon arrival to the intensive care unit (p = 0.029). They averaged 0.13 ± 0.08 (p = 0.764), and 0.07 ± 0.03 μg/l (p = 0.203) at 6 and 24 hours after surgery.

In both groups, NSE reached a maximum level, six hours after the operation and s-100β protein reached a maximum level upon arrival to the intensive care unit. Patients in Group A tended to have higher maximum NSE concentrations, although there was no significant difference in NSE concentrations at any point between the two groups. In contrast, the maximum level of s-100β protein in Group A was significantly higher than that of Group B (p = 0.029 though the difference between these has diminished by the time blood samples were collected at the time of arrival to the intensive care unit.

The patients in Group A were divided into two groups: stenosis in patients detected by MRA versus cervical ultrasonography. **Figures 3** and **Figure 4** show the change of serum NSE and s-100β protein concentrations in

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**Table 1** Preoperative patient characteristics.

<table>
<thead>
<tr>
<th></th>
<th>Group A</th>
<th>Group B</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>13</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>72.6 ± 8.1</td>
<td>62.6 ± 11.7</td>
<td>0.01</td>
</tr>
<tr>
<td>Male (%)</td>
<td>9 (69.2%)</td>
<td>16 (76.2%)</td>
<td>0.96</td>
</tr>
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<td>Preoperative EF (%)</td>
<td>59.0 ± 9.3</td>
<td>55.0 ± 10.9</td>
<td>0.29</td>
</tr>
<tr>
<td>Body surface area (m²)</td>
<td>1.61 ± 0.17</td>
<td>1.77 ± 0.22</td>
<td>0.03</td>
</tr>
<tr>
<td>Hypertension (%)</td>
<td>12 (92.3%)</td>
<td>18 (85.7%)</td>
<td>0.97</td>
</tr>
<tr>
<td>Hyperlipidemia (%)</td>
<td>9 (69.2%)</td>
<td>18 (85.7%)</td>
<td>0.47</td>
</tr>
<tr>
<td>Diabetic Mellitus (%)</td>
<td>8 (61.5%)</td>
<td>11 (52.4%)</td>
<td>0.87</td>
</tr>
<tr>
<td>Diabetic Mellitus with insulin (%)</td>
<td>3 (23.1%)</td>
<td>5 (23.8%)</td>
<td>0.71</td>
</tr>
<tr>
<td>Chronic renal failure (%)</td>
<td>5 (38.5%)</td>
<td>5 (23.8%)</td>
<td>0.60</td>
</tr>
<tr>
<td>Chronic renal failure with hemodialysis (%)</td>
<td>2 (15.4%)</td>
<td>1 (4.8%)</td>
<td>0.66</td>
</tr>
<tr>
<td>Smoking History</td>
<td>8 (61.5%)</td>
<td>16 (76.2%)</td>
<td>0.60</td>
</tr>
<tr>
<td>Left main disease</td>
<td>2 (15.4%)</td>
<td>6 (28.6%)</td>
<td>0.64</td>
</tr>
</tbody>
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**Fig. 1** The changes in serum NSE concentrations.

**Fig. 2** The changes in serum s-100β protein concentrations.
these two groups. Though each group included only a small number of patients, there was no significant difference in s-100β protein concentrations at any point. Patients whose stenosis was detected by cervical ultrasonography showed higher NSE concentrations 24 hours after surgery. However, at the other time points, there was no significant difference in NSE concentrations.

Intraoperative and postoperative data are displayed in Table 2. The number of anastomoses, operative time, and the use of bilateral internal thoracic artery (ITA) did not differ between the groups. No patients suffered postoperative strokes in either group.

**Discussion**

S-100β protein, an acidic calcium-binding protein with a molecular weight of 21 kDa, is considered to be a beneficial marker for the prediction of cerebral infarction in patients who have suffered a stroke following cardiac surgery. S-100β is located in astrocytes primarily and can be released to the perivascular space and extravasate, immediately after opening of the blood-brain barrier. Ueno et al reported that serial measurement of serum s-100β protein can be used to predict early postoperative brain injury. Ramlawi et al reported that serum NSE, one of the five isozymes of the glycolytic enzyme, appears to be another marker of brain injury. NSE has a molecular weight of 78 kDa and is found in large amounts in neurons. Different studies have demonstrated a good correlation between the s-100β protein and NSE levels and the neurocognitive function, although other studies were unable to find evidence to support the hypothesis that early release of the s-100β protein may reflect short or long-term neurologic injury capable of producing cognitive impairment.

Some previous studies have disclosed the equivalent quality of OPCAB to conventional coronary artery bypass grafting, which is performed with CPB. One of the most significant advantages of OPCAB is its reduction of neurologic complications. Bonacchi M et al demonstrated that coronary artery bypass surgery without cardiopulmonary bypass causes a significantly greater decrease in s-100β protein and NSE levels than conventional coronary artery bypass surgery. Anderson RE et al. demonstrated that
CABG with CPB caused a 10-fold increase in s-100β protein than OPCAB.\textsuperscript{5} It is highly unlikely that the effect of heparin, cross-clamping, side-biting clamps, and aortic cannulation can account for the difference in s-100β protein release.

At our hospital, we prefer multiple and complete coronary revascularization with composite or sequential grafting, with a preference for in situ arterial grafts in particular. Ahonen et al. reported that partial clamping may induce damage to the diseased vessel wall that can lead to brain damage even without CPB.\textsuperscript{15} Some previous studies have revealed that the aortic no-touch technique reduced the incidence of stroke.\textsuperscript{16–18} Therefore, we have adapted the aortic no-touch technique, and when using the saphenous vein, we have adopted a pass-port system to avoid aortic side-biting clamps.

Patients of angina pectoris frequently have coexistent cervical and intracranial artery stenosis. Such stenosis may be associated with neurologic complications. In this study, we examined differences between the groups with and without stenosis detected by MRA and cervical ultrasonography. S-100β protein is known to have a half-life of about two hours. In the present study, s-100β protein concentrations increased to a maximum level at the point of arrival in the intensive care unit in almost all patients, and decreased to half its maximum value after about six hours postoperatively. NSE concentrations increased to a maximum level six hours after operations in almost all patients. Moreover, patients who have cervical and intracranial artery stenosis showed significant higher maximum s-100β protein concentrations compared to patients without stenosis. This indicates that even OPCAB can potentially damage the central nerve system, especially in patients with arterial stenosis. However, the maximum level of NSE concentration did not show a significant difference between patients with and without arterial stenosis. This may indicate that the damage to the central nerve system resulting from OPCAB is relatively small.

Some previous studies have revealed that OPCAB reduces the release of the serum s-100β protein and NSE concentrations compared with CABG with CPB. Bonacci M et al. reported that the maximum value of the s-100 protein was 1.4 ± 0.4 μg/l and NSE was 177 ± 6.5 μg/l for patients with CPB.\textsuperscript{7} Similarly, Anderson RE et al. reported that the maximum value of the s-100β protein was 2.4 ± 1.5 μg/l for patients with CPB after operation.\textsuperscript{5} Compared to these reports, the patients in the present study displayed relatively slight increases in s-100β protein and NSE, even those patients with arterial stenosis. We were able confirm the hypothesis that OPCAB has less of an effect on the central nerve system, particularly for patients who have arterial stenosis, than was previously thought.

Patients with postoperative neurologic complications tend to display prolonged release of s-100β protein for 24 to 48 hours after operation.\textsuperscript{19} In the present study, s-100β protein levels normalized for almost all patients in both groups 24 hours after operation, and no patients appeared to display any clinical symptoms of neurologic damage. No patients suffered low blood pressure (under 80 mmHg) during operation; keeping blood pressure over 80 mmHg seems to be essential to avoid neurologic complications.

The main limitation of this study is its small number of patients at a single institution. A larger population is required to detect clinical differences in neurologic complications. Another limitation is that we did not perform other preoperative screening measures. In addition, we did not investigate neurocognitive dysfunction after operation in detail. Moreover, whether these slight increasing of s-100β protein and NSE have any clinical meaning or not is not uncertain. Thus, repairing the intracranial and cervical artery stenosis before OPCAB is controversial.

In conclusion, our findings show a correlation between the stenosis detected by MRA or cervical ultrasonography and brain damage after OPCAB. We found that it is necessary to perform MRA and cervical ultrasonography, preoperatively, to evaluate neurologic complication risk factors. Even OPCAB has the potential damage to the central nerve system. During the operation, especially during distal anastomosis, maintaining blood pressure above 80mmHg is critical for avoiding neurologic complications.

References