Lactate Dehydrogenase Isoenzyme-1 in Serum for Detection of Peri-Operative Myocardial Infarction after Cardiac Surgery

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We prospectively studied changes in serum lactate dehydrogenase isoenzyme-1 (LD-1, EC 1.1.1.27) in 99 consecutive patients after either coronary artery bypass grafting (CABG, n = 61), isolated cardiac-valve replacement (n = 24), or the two procedures combined (n = 14); 86 of these had no clinical evidence of peri-operative myocardial infarction (MI). Blood was sampled immediately after surgery and at 6-h intervals for up to 42 h thereafter. LD-1 was isolated by using the LD M-subunit antiserum. Samples from the non-MI patients were used to establish the reference intervals for LD-1. By 24 h after surgery, mean serum LD-1 values were higher (P < 0.001) in non-MI patients who underwent isolated valve replacement (222 ± 74 U/L) or combined CABG and valve replacement (266 ± 58 U/L) than in 50 non-MI patients who underwent CABG alone (134 ± 42 U/L). Separate reference intervals were determined for CABG and other patients at each sampling time. By 24 h after operation, LD-1 exceeded these reference intervals in the 10 CABG and two combined-procedure patients in whom other evidence of MI was present. Measurement of LD-1 24 to 42 h after cardiac surgery appears to be a useful test for the diagnosis of peri-operative MI.

The diagnosis of peri-operative myocardial infarction (MI) in cardiac surgery is difficult and problematic.6 Diagnostic modalities such as electrocardiography, serum enzyme measurements, and scintigraphy can all be misleading (1–8). Among the various biochemical approaches, measurements of creatine kinase (CK, EC 2.7.3.2) isoenzymes have become the most commonly used (1, 5, 7–16), despite conflicting results regarding their clinical specificity.

Fewer studies (1, 8, 17–19) have evaluated the use of electrophoretic methods for measuring lactate dehydrogenase (LD, EC 1.1.1.27) isoenzymes to identify MI associated with cardiac surgery. During the last few years, a quantitative immunochemical method for measuring LD-1 has become available (20). The method is simple, rapid, and precise, and it appears to be sensitive and specific for the diagnosis of acute MI in coronary-care units (20–23). Its utility in other settings has been little studied.

Quantitative assays are attractive for the study of peri-operative MI. Because cardiac surgical procedures involve cardiac trauma, one can expect cardiac enzymes to be released into the serum. By determining appropriate reference intervals for the concentrations of these enzymes at specific times after the procedures, one can assess results for patients with evidence of peri-operative MI.

Here we report a study in which we determined LD-1 concentrations in serum after common cardiac surgical procedures, specifically coronary artery bypass grafting (CABG), valve replacement, or both. We were especially careful to define surgical populations with no evidence of peri-operative MI.

Patients and Methods

The study sample comprised 99 consecutive patients (69 men, 30 women; mean age 53 y, SD 12, range 28–88) undergoing cardiac surgical procedures at the University of Virginia Hospital. Sixty-one patients underwent simple or sequential CABGs with autologous saphenous vein grafts. In patients with extensive atherosclerotic disease distal to a stenotic or occluded vessel, endarterectomy was performed before the graft insertion. Twenty-four patients underwent aortic or mitral valve replacement with either Starr–Edwards or porcine prostheses. Fourteen additional patients had combined CABGs and either valve replacement (n = 11) or aneurysmectomy (n = 3). Various anesthetic agents were used, halothane being the most common. Myocardial tissues were protected by lowering the core body temperature to 21–28 °C, placing iced saline in the pericardial well, and infusing cold cardioplegia solution every 15–30 min.

Initial blood samples were obtained on admission to the surgical intensive-care unit (i.e., immediately after the operation) and at 6-h intervals thereafter. Fifty-six of our 99 study patients remained in the intensive-care unit for at least 42 h, 76 for at least 36 h, and all but one for at least 18 h. From this study group of 99 patients, we obtained 681 (98%) of the 710 planned samples; 26 of these (3.8%) were grossly hemolyzed and discarded, including all eight samples from one CABG patient, who was excluded from further analysis. Faint hemolysis, which frequently was present in initial postoperative samples, was ignored. Only 25 of the remaining 655 specimens underwent less than the complete planned analyses, owing to laboratory error or an insufficient volume of sample.

LD and LD-1 were assayed at 30 °C with a centrifugal analyzer as previously described (21). Commercial immunochemical reagents were used (Roche Diagnostics, Nutley, NJ). In 81 patients we also determined CK-B subunit activity at 30 °C, using an immunoinhibition technique (24) (Smith Kline Instruments, Sunnyvale, CA 94086) and a RotoChem IIa/36 centrifugal analyzer (American Instruments, Silver Spring, MD 20910). For most of the 681 samples, CK isoenzymes were also viewed fluorometrically after electrophoresis in agarose (Corning ACI; Corning Medical, Medfield, MA 02052). Results graded by the laboratory as "trace" for CK-MB were ignored in this study. The results of the quantitative isoenzyme measurements were not available to the clinicians during the course of this study.

To define the expected range of LD-1 results after entirely uncomplicated cardiac surgery, we carefully selected, from
Table 1. Indicators of Acute Myocardial Injury in 12 Patients Excluded from the Two Control Groups

<table>
<thead>
<tr>
<th>Subject no.</th>
<th>Electrocardiographic findings</th>
<th>Electrophoretic CK-MB (pos/total)</th>
<th>Peak CK-B subunit (U/L)</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>New Q waves</td>
<td>4/8</td>
<td>26^b</td>
<td>—</td>
</tr>
<tr>
<td>12</td>
<td>New Q waves</td>
<td>5/8</td>
<td>60^b</td>
<td>Coronary angiogram: occluded graft</td>
</tr>
<tr>
<td>16</td>
<td>R/S &gt;1 in V1–V2 (Q wave equivalent)</td>
<td>0/4</td>
<td>13</td>
<td>Early ventricular arrhythmias</td>
</tr>
<tr>
<td>22</td>
<td>Left bundle branch block</td>
<td>4/6</td>
<td>N.D.</td>
<td>Defibrillation × 8</td>
</tr>
<tr>
<td>42^a</td>
<td>New inverted T waves V5–V6</td>
<td>5/8</td>
<td>N.D.</td>
<td>Early ventricular arrhythmias</td>
</tr>
<tr>
<td>45^b</td>
<td>New Q waves</td>
<td>4/4</td>
<td>14</td>
<td>Hemodynamic instability</td>
</tr>
<tr>
<td>52</td>
<td>NSSTTTWAd</td>
<td>2/7</td>
<td>N.D.</td>
<td>Autopsy: recent MI</td>
</tr>
<tr>
<td>59^c</td>
<td>NSSTTTWA</td>
<td>4/7</td>
<td>49^b</td>
<td>Coronary angiogram: proximal occlusion of saphenous graft</td>
</tr>
<tr>
<td>71</td>
<td>New Q waves</td>
<td>0/6</td>
<td>9</td>
<td>—</td>
</tr>
<tr>
<td>77</td>
<td>NSSTTTWA</td>
<td>2/4</td>
<td>28^b</td>
<td>—</td>
</tr>
<tr>
<td>83</td>
<td>New Q waves</td>
<td>N.D.</td>
<td>17</td>
<td>—</td>
</tr>
</tbody>
</table>

^a Number of samples with positive CK-MB results/total number of samples tested. Most patients with uncomplicated coronary artery bypass grafting (CABG) had no samples with MB bands; the mean number of samples with positive MB bands, per patient, was 0.3.

^b Increased values (see Figures 1–3).

^c These patients underwent combined CABG and aortic valve replacement; all others underwent CABG alone.

^d NSSTTTWA = nonspecific ST and T-wave changes. N.D. = not done.

our final study cohort of 98, a subgroup of 86 patients for whom at least three postoperative blood samples were available. All 86 had uneventful postoperative courses and none showed objective evidence or any special grounds for clinical suspicion of peri-operative MI. The group of 86 patients included 50 patients with uncomplicated CABG, 24 with valve replacement, and 12 patients who underwent both CABG and valve replacement or aneurysmectomy. Reference intervals were calculated by use of non-parametric statistics (25). Values for this subset are presented in the text as means ± standard deviations. Among the 12 patients who were excluded (Table 1), eight were excluded because a diagnosis of peri-operative MI (n = 6) or possible MI (n = 2) was indicated by a physician’s note in the medical record, and four were excluded because their CK-B values for one or more samples exceeded the mean values by more than three standard deviations during the first 24 h after surgery. The LD data were not used in making the diagnosis of suspected MI. As shown in Table 1, all had independent evidence of myocardial damage. Ten of the 12 patients underwent CABG alone and two had undergone CABG and valve replacement. The statistical significance of differences between means was assessed by t-test.

Results

Uncomplicated Bypass Grafts

Figure 1 illustrates the LD-1, the percent LD-1, and the CK-B subunit activity in the 50 patients with uncomplicated CABGs. The mean LD-1 value observed in this group on admission to the intensive-care unit (Figure 1A, time 0) was 121 ± 53 U/L. This contrasts with the reference interval that we found for apparently healthy volunteers, 26–73 U/L (21). Mean LD-1 values increased during the subsequent 24 h after surgery, reaching a peak value of 134 ± 42 U/L at 24 h. Thereafter, LD-1 showed no statistically significant change between 24 and 42 h. Figure 1B demonstrates the percent LD-1 values ([LD-1/total LD] × 100%) in the same group of patients. Mean percent LD-1 values increased slightly but progressively during the initial 24 h after surgery (reference interval for volunteers: 21–35%). This was followed by a plateau throughout the remainder of the study period, and at 42 h the mean LD-1 was 33% (SD 7%). Figure 1C illustrates the results for CK-B subunit in the same CABG patients. On admission to the intensive-care

Fig. 1. Serum enzymes in 50 CABG patients without subsequent MI, measured at 6-h intervals after operation
unit, their mean CK-B subunit activity was 7.2 (SD 3.3) U/L. Subsequently, B-unit activity declined gradually, reaching 5.9 (SD 4.6) U/L at 18 h and 3.5 (SD 3.2) U/L at 42 h. CK electrophoresis was performed on samples from 37 of these 50 non-complicated CABG patients. Of these patients, four (11%) revealed a "positive" MB band on electrophoresis (in one to four samples).

The number of discrete aorto-saphenous vein grafts appeared to influence the peak LD-1 activity. When one graft was implanted, the mean peak LD-1 was 118 (SD 66) U/L; two grafts resulted in 155 (SD 77) U/L (P = 0.15 vs the one-graft group); three grafts, 148 ± 28 U/L (P = 0.1); and four grafts resulted in a mean peak LD-1 value of 186 (SD 61) U/L (P = 0.01). The effect of aortic cross-clamping interval on peak LD-1 activity was also evaluated: for those shorter and longer than 70 min, the mean peak LD-1 activities were respectively 139 (SD 41) U/L and 188 (SD 53) U/L (P = 0.008).

Uncomplicated Valvular Procedures

Figure 2A illustrates the LD-1 values for the 36 patients who underwent uncomplicated valve replacement, alone or combined with CABG. Mean LD-1 values were higher in these 36 subjects than in those undergoing CABG alone at each time point after the operation. However, the pattern of change of LD-1 activity during the course of the study was similar to that observed in CABG patients, showing a slight but progressive increase during the first 24 h after operation. Figure 2B shows the percent LD-1 activity, and Figure 2C shows the CK-B subunit activity in this group of 36 patients. As can be seen, both were slightly higher than in patients who were undergoing only CABG. Table 2 compares mean LD-1, percent LD-1, and CK-B subunit activities at three time points for CABG, valve-replacement, and the combined procedures. Significant differences among these three groups are indicated in the table. Importantly, mean LD-1 values at 24 h and 42 h were significantly higher (P < 0.001) in the valve-replacement and combined-procedure groups than in the CABG group. Moreover, mean CK-B values at 0, 24, and 42 h were significantly higher in the valve-replacement group and in the combined-procedure group than in the CABG group. Because of these differences among groups, we constructed separate reference intervals for the three surgical groups.

Suspected Peri-Operative MI

Figure 3 demonstrates, as shaded areas, reference intervals for LD-1, percent LD-1, and CK-B subunit activities. These data were derived from our 50 patients with uncomplicated CABG procedures alone. These intervals were determined nonparametrically and include the lower 95% of the values at each time point in Figure 1. Superimposed on this range are the values for the 10 CABG patients with suspected peri-operative MI. Values for each patient are connected by a single line.

Figure 3A shows the LD-1 results for these patients. By 24 h after surgery, LD-1 in each of the 10 patients exceeded the interval previously determined for the uncomplicated CABG patients and remained increased throughout the study period. On the other hand, LD-1/total LD (percent LD-1) was above the range in only seven of the 10 patients with suspected peri-operative MI (Figure 3B). CK-B subunit activity was measured in seven of the 10 patients (Figure 3C). In four of them, the values were markedly above the

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**Table 2. Results Compared for Patients Who Underwent CABG, Valve Replacement, or Both**

<table>
<thead>
<tr>
<th>Time after operation, h</th>
<th>CABG (n = 50)</th>
<th>Valve replacement (n = 24)</th>
<th>CABG and valve replacement (n = 12)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>LD-1, U/L</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>121 ± 33</td>
<td>169 ± 79&lt;sup&gt;b&lt;/sup&gt;</td>
<td>147 ± 27&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>24</td>
<td>134 ± 42</td>
<td>222 ± 74&lt;sup&gt;c&lt;/sup&gt;</td>
<td>266 ± 58&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>42</td>
<td>137 ± 60</td>
<td>218 ± 69&lt;sup&gt;c&lt;/sup&gt;</td>
<td>258 ± 59&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>LD-1/total LD, %</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>26 ± 5</td>
<td>25 ± 6</td>
<td>26 ± 3</td>
</tr>
<tr>
<td>24</td>
<td>32 ± 6</td>
<td>34 ± 9</td>
<td>37 ± 4&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>42</td>
<td>33 ± 7</td>
<td>40 ± 9&lt;sup&gt;c&lt;/sup&gt;</td>
<td>39 ± 4&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>CK-B, U/L</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>7.2 ± 3.3</td>
<td>11 ± 4</td>
<td>13 ± 4&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>24</td>
<td>5.2 ± 3.7</td>
<td>10 ± 5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>14 ± 6&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>42</td>
<td>3.5 ± 3.2</td>
<td>5.5 ± 2&lt;sup&gt;c&lt;/sup&gt;</td>
<td>8 ± 4&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup> Mean ± SD.  
<sup>b,c</sup> Significantly different from CABG patients: <sup>b</sup>P < 0.05, <sup>c</sup>P < 0.001.
The suspected findings during the first 12 h after surgery, then declined toward the reference interval. In the remaining three patients, all of whom had new electrocardiographic findings consistent with peri-operative MI, values for CK-B did not exceed the reference interval anytime during the study.

Table 3 compares the mean isoenzyme values obtained during the first 24 h for CABG patients with and without suspected peri-operative MI group. Mean LD-1 values were significantly higher in the suspected MI group in comparison with the uncomplicated CABG group of patients, starting 6 h after operation, and remained higher. In contrast, CK-B values were significantly higher in the suspected MI group only 18 h after the operation.

We used receiver–operator characteristic curves (Figure 4) to compare the abilities of the various enzyme tests to discriminate MI from non-MI. The LD-1 (U/L) was the best test. At a cutoff of 202 U/L, the test was 100% sensitive and 95% specific; at a cutoff of 230 U/L it was 100% specific and 90% (9/10) sensitive. By contrast, LD-1 (%) at 24 h achieved at best an 80% sensitivity at a specificity of 90% and CK-B (at 12 or 18 h) achieved only 62–71% sensitivities at specificities of 89–94%.

Two patients who underwent combined CABG and valve replacement had evidence of peri-operative MI (Table 1, patients no. 45, 59). In both, LD-1 greatly exceeded the values in comparable patients with uncomplicated courses, reaching values of 416 and 671 U/L, respectively, 12 h after surgery. CK-B values also were increased in these two patients 18–36 h after operation, as compared with the uncomplicated group.

**Discussion**

Peri-operative myocardial damage remains the major cause of early mortality associated with cardiovascular surgical procedures, especially CABG (1, 10, 19). Iatrogenic electrocardiographic changes and release of enzymes from extracardiac tissues complicate the evaluation of myocardial injury in the peri-operative period. The diagnostic value of new Q waves appearing in the postoperative period has been questioned, because they may result from hypothermia (26), severe metabolic acidosis, shock (27), cross clamping of the aorta (26), or the unmasking of an old MI (28). Use of data on total LD and CK has also been questioned, because these enzymes may not be specific enough to permit the diagnosis of MI after cardiac surgery (29, 30). Most attention has been focused on CK-MB determinations. Several reports (14, 31, 32) indicate a close correlation between electrocardiographic data and CK-MB detection in peri-operative patients, but the presence of CK-MB, although highly suggestive of myocardial injury, may not be specific. Baur et al. (7) reported that 77% of patients who were undergoing CABGs demonstrated the presence of this enzyme in the immediate postoperative period. The same study was unable to correlate the presence of CK-MB with postoperative arrhythmias or appearance of other complications such as
congestive heart failure or shock. The type of surgical procedure and the techniques used obviously affect postoperative serum CK-MB. Thus, the presence or absence of electroforethetically detectable CK-MB in serum is unlikely to be helpful.

Considering the above limitations of CK-MB measurements for diagnosing peri-operative MI, we tried in this study to assess the possible value of immunochemically measured LD-1 in the diagnosis of peri-operative MI. The ability of the test to identify patients with suspected perioperative MI within a population of CABG patients is identified in Figure 3. On the basis of LD-1 data (Figure 3A), eight of the 10 patients with suspected perioperative MI were separated from the reference interval within 18 h after operation. In the two additional patients, LD-1 was increased at 24–36 h. Increased LD-1 values persisted in all 10 patients up to 36–42 h after surgery (the end of the study). This may be of value in the late recognition of a perioperative MI one or two days—or even longer—after surgery, when CK-MB values have returned to the normal reference interval. Mean LD-1 values in the suspected MI group started to be significantly higher than in the noncomplicated CABG group as early as 6 h after operation (Table 3). In contrast, the LD-1/total LD (percent LD) provided a loss-clear discrimination of MI and non-MI patients. Only six of the 10 patients with other evidence of perioperative MI had values outside the previously determined reference interval. In the uncomplicated CABG patients, CK-B activity was greatest in the first postoperative specimen (Figure 1C), and this is similar to findings in several studies (2, 7, 10, 15) in which CK-MB reportedly peaked 8–12 h after surgery in non-complicated CABG patients. CK-MB was measured in eight of the CABG patients with suspected perioperative MI. Only in five of them did it clearly exceed the reference interval previously established for non-MI patients. Mean CK-B was significantly higher in the suspected MI group than in the uncomplicated group, but not earlier than 18 h after surgery. Delva et al. (33) also reported that CK-MB liberation was prolonged, with a delayed peak, when MI complicated CABG procedures.

Only a few investigators, using electrophoretic methods, have studied LD isoenzymes in peri-operative cardiac surgical patients. Codd et al. (18) reported that all their patients with peri-operative MI showed peak values for LD-1 exceeding 160 U/L within the first 48 postoperative hours, whereas 18 of 22 patients (80%) with persistent ischemia showed peak LD-1 values >160 U/L. An additional six patients had peak values for LD-1 >160 U/L without diagnostic electrocardiographic changes. Of these six, four had associated increases in total enzyme, suggesting myocardial injury. Papadopoulos and Hufnagel (19) reported a characteristic reversal of the LD-1/LD-2 ratio in seven patients with perioperative MI out of 50 post-cardiac-surgery patients they studied. Mohiuddin et al. (17) reported that 18 of 23 postoperative patients (78%) with "flipped" LD-1/LD-2 patterns developed new Q waves, as compared with only one of 50 patients without LD-1/LD-2 reversal. Graeber et al. (8) reported that each of 19 patients who had valve replacement or CABGs complicated by MI had an LD-1/LD-2 ratio >1 from postoperative day 1 to day 4. These reports contrast with that of Keshgengan et al. (1), who reported a sensitivity of 64% and specificity of 88% for total LD activity (at the cutoff point of 600 U/L) for the diagnosis of peri-operative MI, in comparison with 45% and 97% for CK at the cutoff point of 300 U/L. These authors found total LD to be more efficient for the diagnosis of peri-operative MI than the LD-1/LD-2 ratio.

Postoperative increases in CK-MB in serum have been correlated positively with duration of extracorporeal circulation and length of aortic cross-clamp time (5). Strom et al. (10) noted that an aortic cross-clamp time >70 min led to an increased release of CK-MB. Hypothermic cardioplegic techniques combined with short aortic cross-clamp time have been shown to be associated with very low values for CK-MB in the postoperative period (34). In the present study, peak LD-1 was also increased in the non-complicated patients in whom the cross-clamp time exceeded 70 min. Our study showed also a relatively slight increase in peak LD-1 activity in correlation with increasing number of grafts.

We found significantly higher values for LD-1, percent LD-1, and CK-MB in the valve-replacement group and in the CABG-plus-valve-replacement group in comparison with the non-complicated CABG-only group. Similar results were reported by Jarvinen et al. (35) who found CK-MB to be higher 18 h after valve replacement in 76 patients than in 13 controls who were undergoing closure of atrial septal defect (59 ± 103 U/L vs 45 ± 39 U/L). We found both LD-1 and CK-B values to be higher in the two MI patients undergoing CABG and valve replacement in comparison with the uncomplicated group.

In conclusion, our study indicates that immunochemically determined LD-1 is a valuable tool for postoperative evaluation of patients undergoing either CABG procedures, valve replacement, or both. An important caveat is that separate reference intervals are required for each type of procedure and for specific times following the procedures. The immunochemical method provides a simple and accurate quantification of LD-1. The reproducibility of the assay (7, 14, 15) is excellent, and results can be made available quickly. An increased LD-1 can be used to confirm the results of CK-MB testing. Moreover, the duration of the increase in LD-1 considerably exceeds the few hours during which CK-MB remains increased after peri-operative MI. This allows myocardial damage to be detected at later times. Finally, CK-MB testing requires frequent blood drawing (every 4–8 h) if one is to detect and define reliably the brief rise and fall associated with MI. By contrast, a normal value for LD-1 in a single blood sample drawn 18–30 h postoperatively provides strong evidence against peri-operative MI. Fortuitously, the upper limit of the reference interval shows little variation during this convenient time window.

References


