Relationship Between Interleukin 6 and Mortality in Patients With Unstable Coronary Artery Disease
Effects of an Early Invasive or Noninvasive Strategy

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Atherosclerosis is characterized by inflammation in the vessel wall. On the site of the atherosclerotic plaque, the intima is infiltrated with activated macrophages and T lymphocytes, which both produce and secrete cytokines to drive the inflammatory process on. In unstable coronary artery disease (CAD), increased levels of systemic markers of inflammation, such as the acute phase reactants C-reactive protein and fibrinogen, are common. Also, increased plasma levels of proinflammatory cytokines, such as interleukin (IL)-1β, IL-6, and IL-8, have been reported. Large cohort studies have shown an association between elevated levels of circulating C-reactive protein or fibrinogen and increased risk for cardiac events or death, both in patients who have experienced an episode of unstable CAD as well as in apparently healthy individuals.

Interleukin 6 is a cytokine with both proinflammatory and anti-inflammatory effects on many cell types, affecting both B-cell immunoglobulin production and T-cell cytotoxic activity. Interleukin 6 also affects platelet production and reactivity as well as endothelial function. It is the only substance known to induce synthesis of all of the acute phase proteins by the liver. Large prospective studies of healthy populations have shown that IL-6 plasma levels in the upper quartile of the considered normal range are inde-

Context Inflammatory activity is associated with high rates of long-term mortality in unstable coronary artery disease (CAD). Interleukin 6 (IL-6) induces C-reactive protein and fibrinogen, systemic markers of inflammation.

Objectives To determine whether plasma levels of IL-6 are predictive of mortality and to evaluate the interaction of IL-6 levels with the effects of invasive vs noninvasive treatment strategies in unstable CAD patients.

Design, Setting, and Patients The prospective, randomized Fragmin and Fast Revascularisation During Instability in Coronary Artery Disease II trial, conducted among 3489 patients, 3269 of whom had plasma samples analyzed for IL-6 levels, with diagnosed unstable CAD (67% male; median age, 67 years) at 58 Scandinavian hospitals between June 1996 and August 1998.

Interventions Patients were randomly assigned to receive either an early invasive (n = 1222) or a noninvasive treatment strategy (n = 1235). The latter group, as well as 666 patients with contraindications to invasive therapy, were further randomized to 90-day treatment with low-molecular-weight heparin (dalteparin, 5000-7500 IU twice per day; n = 1140) or placebo (n = 1127).

Main Outcome Measure Mortality at 6 and 12 months in the medically and interventionally randomized cohorts, respectively, in relation to IL-6 levels, measured at randomization.

Results Plasma levels of IL-6 that were at least 5 ng/L compared with levels lower than 5 ng/L were associated with greatly increased mortality in the noninvasive group (7.9% vs 2.3%; relative risk [RR], 3.47; 95% confidence interval [CI], 1.94-6.21) and in the placebo-treated group (7.9% vs 2.5%; RR, 3.19; 95% CI, 1.77-5.74). The association remained significant after adjustment for most established risk indicators. An early invasive treatment strategy strongly reduced 12-month mortality among those with elevated IL-6 levels (5.1% absolute reduction; P = .004) whereas mortality was not reduced among patients without elevated IL-6 concentrations. Those taking dalteparin with elevated IL-6 levels experienced lower 6-month mortality than those who did not take dalteparin (3.5% absolute reduction; P = .08).

Conclusions Circulating IL-6 is a strong independent marker of increased mortality in unstable CAD and identifies patients who benefit most from a strategy of early invasive management.

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INTERLEUKIN 6 AND MORTALITY

Three hundred sixty-six patients enrolled after the end of recruitment for the intervention trial.

DEPENDENTLY PREDICTIVE OF AN INCREASED RISK OF PREMATURE DEATH OR FUTURE MYOCARDIAL INFARCTION (MI), EVEN AFTER ACCOUNTING FOR C-REACTIVE PROTEIN LEVEL. This indicates a possible role for IL-6 in the progression of CAD. We have previously shown a strong relationship between increased levels of C-reactive protein, fibrinogen or troponin T and increased risk for long-term mortality in patients with unstable CAD. Since IL-6 induces both C-reactive protein and fibrinogen, we sought to determine its predictive value for long-term risk of death or MI in patients included in the Fragmin and Fast Revascularization During Instability in Coronary Artery Disease II (FRISC II) trial. We also investigated the effects of an invasive vs. noninvasive treatment strategy and prolonged treatment with low-molecular-weight heparin (dalteparin) in relation to IL-6 levels at admission.

METHODS

Patients

In all, 3,489 patients were recruited at 58 Scandinavian hospitals between June 1996 and August 1998. Patients were eligible for study inclusion if they had symptoms of ischemia that were increasing or occurring at rest or that warranted the suspicion of acute MI, with the last episode within 48 hours before initiating dalteparin or heparin treatment. Myocardial ischemia had to be verified by electrocardiography (ST-segment depression ≥0.1 mV or T-wave inversion ≥0.1 mV) or by raised biochemical markers (creatine kinase (CK)-MB >6 µg/L, troponin T >0.10 µg/L, positive qualitative troponin-T test, or catalytic activity of CK, CK-B, or CK-MB higher than the diagnostic limit for MI). Exclusion criteria were increased risk for bleeding episodes, anemia, indication for treatment in the past 24 hours with thrombolysis, angio-plasty in the previous 6 months, being on a waiting list for a coronary revascularization procedure, other acute or severe cardiac disease, renal or hepatic insufficiency, known clinically relevant osteoporosis, other severe illness, hypersensitivity to randomized drugs, anticipated difficulties with cooperation, or previous participation in this or another clinical trial. Patients with previous open heart surgery, advanced age (>75 years), or other disorders that made randomization to early revascularization inappropriate were excluded from randomized intervention strategies but were still assigned to receive dalteparin or placebo.

Study Design

The FRISC II study was a prospective, randomized, multicenter trial with parallel groups. The details of the FRISC II medical and interventional studies have been described previously. A simplified overview of the study design and the subgroups evaluated in this article are shown in Figure 1. All patients were initially treated with subcutaneous dalteparin or standard heparin by intravenous infusion. Randomization into the different treatment strategies was completed within 72 hours of start of open-label dalteparin or standard heparin. All patients received dalteparin subcutaneously (120 IU/kg every 12 hours) for at least 5 days until they underwent an exercise test or revascularization. Thereafter, they entered the double-blind treatment with twice-daily subcutaneous injections of dalteparin (5000 IU per dose for men weighing <70 kg and women <80 kg; 7500 IU per dose for those who exceeded these weight limits) or placebo until 90 days after entry. The direct invasive strategy required coronary angiography within a few days of enrollment, aiming for revascularization within 7 days of the start of open-label treatment. Revascularization was recommended in all patients with an obstruction of at least 70% of the diameter of any artery supplying a substantial proportion of the myocardium. Percutaneous coronary intervention was recommended if there were 1 or 2 target lesions, and coronary artery bypass surgery was preferred in patients with 3-vessel or left main artery disease.

In the noninvasive strategy, coronary angiography and, if appropriate, revascularization were recommended in patients with refractory or recurrent symptoms despite maximum medical treatment or severe ischemia on an exercise electrocardiography test before discharge. During follow-up, invasive procedures were considered, irrespec-
tive of randomized strategy, for all patients with incapacitating symptoms, recurrence of instability, or MI.

Aspirin was given to all patients on admission (initial dose, 300-600 mg; maintenance dose, 75-320 mg once daily), and β-blockers were given unless contraindicated. Statins for lowering cholesterol, angiotensin-converting enzyme inhibitors, and aggressive antidiabetic treatment were given at the discretion of the treating physicians.

End Points
The end points investigated in this study were all-cause death and the composite of death or nonfatal MI, which was the primary end point of the FRISC II trial. The medical study had a follow-up for 6 months. Concerning the interventional trial, longer follow-up was prep lanned and, hence, information on death or MI was also available at 12 months. Follow-up was unavailable for 14 patients in the dalteparin group and for 18 patients in the placebo group. In the interventional study, vital status was not available for 1 patient who requested withdrawal from the study. Definitions of the various predefined end points in FRISC II have been previously described.16,17 The study complied with the Declaration of Helsinki, and all local ethics committees approved the protocol. Informed consent was obtained from all included patients.

Analyses
Venous blood samples in tubes containing EDTA (Vacutainer, Becton-Dickinson, Plymouth, England) were taken from all patients at randomization. The plasma was separated by centrifugation (2000 g for 20 minutes) within 30 minutes of blood sampling, aliquoted, and stored at −70°C until analysis. Troponin T levels were measured with a third-generation assay kit on an Elecsys 2010 instrument (Roche-Boehringer Mannheim, Mannheim, Germany). Concentrations of C-reactive protein were analyzed using the Immulite Automated Analyzer and assay kit (DPC, Diagnostic Products Corp, Los Angeles, Calif). The IL-6 antigen levels were measured in plasma from 3269 patients by a sequential immunometric assay, also using the Immulite analyzer and kit. The lower detection limit of this system is 5 ng/L, which was therefore used as the cutoff for the statistical analyses. Levels higher than the cutoff are reported herein as higher than 5 ng/L, high, raised, or elevated.

Statistical Analyses
The levels of IL-6, below or above the cutoff level were used to test the association between IL-6 and outcome events. All statistical comparisons between randomized treatments were performed according to the intention-to-treat principle. In the medical part of the study, events were recorded from the initiation of the open-label dalteparin treatment until the 6-month follow-up. In the interventional study, recording of events terminated at the 12-month follow-up. The efficacy analyses of the 6-month and 12-month follow-ups were point estimates including only patients with an adjudicated event or with recorded absence of the specific event until at least 170 days and 335 days of the respective follow-up period. Pearson χ² analysis was used to test significance of the overall degree of association. Graphs of the Kaplan-Meier estimate of the survival function were used without statistical tests. Forward stepwise logistic regression analysis was used to adjust for established risk indicators regarding mortality and for evaluating factors that could contribute to elevated IL-6 levels. All P values are 2-tailed, and values lower than .05 were considered statistically significant. Relative risks (RRs) and odds ratios (ORs) are expressed with 95% confidence intervals (CIs). Data processing and statistical analyses were performed using SPSS version 10.0 software (SPSS, Chicago, Ill).

RESULTS
Patient Characteristics and Procedures
As shown in Figure 1, 2267 patients were included in the medical part of the FRISC II trial while 2457 were enrolled in the interventional trial. Within 7 days, 96% of patients in the invasive group and only 7% of the patients in the noninvasive group underwent coronary angiography. Within the first 10 days, 71% in the invasive group and 9% in the noninvasive group underwent revascularization procedures. Within 12 months, 78% in the invasive and 43% of the patients in the noninvasive groups had undergone revascularization. The details of treatments, follow-up, and outcome on these materials have previously been reported.16-18

The 3269 analyzed plasma samples were taken at a median of 39 hours after onset of the last episode of chest pain (interquartile range, 27-55 hours). Plasma IL-6 levels were distributed equally between the randomized patient groups (data not shown). There were no significant differences in other baseline characteristics between the groups.16,17 Baseline characteristics according to IL-6 levels are summarized in Table 1. Of note, patients with increased IL-6 levels also were more likely to have increased troponin T and C-reactive protein levels but were only slightly more likely to have ST-segment depression.

Outcome by Invasive and Noninvasive Cohorts
Because there was no influence of long-term dalteparin in the comparisons between the invasive and noninvasive groups, analyses of these cohorts were performed disregarding medical assignment (Figure 1).

For patients randomized to a noninvasive strategy, IL-6 levels 5 ng/L or higher at inclusion were associated with a 3.5-fold increase in probability of death at 12 months; 7.9% compared with 2.3% in patients with IL-6 levels less than 5 ng/L (P < .001; Table 2). In patients with high IL-6 levels, an early invasive strategy led to a 5.1% absolute or 65% relative reduction in 12-month mortality (Table 2, Figure 2). At lower IL-6 levels, there was no significant difference in 12-month mortality between treatment strategies.

For the composite end point of death or MI, elevated plasma IL-6 levels were not associated with any significantly
larger event proportion in the noninvasive group (Table 2). Accordingly, an invasive strategy improved the end point outcome irrespective of IL-6 levels.

**Outcome in Noninvasive, Medically Randomized Cohorts**

In the noninvasive placebo-treated group, patients with IL-6 levels of 5 ng/L or higher had a 6-month mortality rate of 7.9% vs 2.5% in patients with levels lower than 5 ng/L (P = .001, Table 2). At elevated IL-6 levels, assignment to dalteparin tended to reduce the risk to 4.4% (P = .08; Table 2). In patients with IL-6 lower than 5 ng/L, dalteparin treatment did not influence 6-month mortality.

As for the combined end point, high plasma IL-6 levels were not significantly associated with risk (RR, 1.16; 95% CI, 0.83-1.62 in the placebo group; Table 2). Assignment to dalteparin treatment did not reduce the composite of mortality or occurrence of MI at 6 months regardless of IL-6 levels. However, it significantly lowered the incidence of these events during the first 60 days of treatment in patients with elevated plasma IL-6 (P = .01-.04 at 30, 45, and 60 days), but not in those with lower levels (Figure 3).

**Multivariable Analysis**

The independence of IL-6 levels as a predictor of mortality was assessed by forward stepwise logistic regression analysis, for which established risk indicators as well as randomized treatments and interaction terms were evaluated. Only 7 covariates remained in the interventional part of the trial and 5 covariates in the medical part as independent predictors (Table 3). Plasma IL-6 levels remained significantly associated with increased mortality in both the interventional study (adjusted odds ratio [OR], 2.08; 95% CI, 1.24-3.49; P = .006) and the medical trial (adjusted OR, 2.09; 95% CI, 1.31-3.33; P = .002).

**COMMENT**

In this study, we found that plasma IL-6 level is an independent marker for identifying patients with unstable CAD with increased risk of death over 6 to 12 months. Interleukin 6 was ‘predictive’ independent of other risk indicators, including the biochemical markers troponin T and C-reactive protein.

Two studies of healthy adults have shown an association between elevated IL-6 levels and total and cardiovascular mortality and future MI. In a study much smaller than ours, Biaucci et al observed that, in patients with CAD, increased levels of IL-6 were predictive of short-term coronary events. Based on these observations, as well as the documented prognostic values of C-reactive protein and fibrinogen, IL-6 seems to settle well as a predictor of long-term mortality of CAD patients having experienced 1 or more episodes of instability.

The FRISC II interventional trial was the first study to show that an early invasive treatment strategy reduced mortality and occurrence of MI in patients with unstable CAD. The greatest beneficial effect was seen in CAD patients presenting with indicators of higher risk at entry, such as elevated troponin T levels or ST-segment depression on entry.
electrocardiography. To predict which patients may benefit from an early invasive strategy, additional markers are needed that provide greater specificity. We now demonstrate that elevated IL-6 levels, independently of other well-known risk indicators, identify patients whose risk of death can be considerably reduced by an early invasive approach. It thus seems that an invasive strategy is of choice despite the increased inflammatory activity in these patients.

Assignment to prolonged treatment with subcutaneous dalteparin also tended to reduce the risk of death for patients with IL-6 levels of 5 ng/L or higher, thus identifying those who would benefit from this kind of treatment. This is noteworthy since not all patients at high risk are eligible for invasive treatment, so medical alternatives such as long-term anticoagulant therapy might be considered.

Elevated plasma IL-6 levels did not correlate with any increased risk of the composite end point of death or MI at 6 to 12 months. In the medical study, however, it identified patients for whom assignment to receive dalteparin had a beneficial effect during the first 60 days. This pattern was also seen in the main FRISC II medical study for patients with troponin T levels higher than 0.1 µg/L. Therefore, an elevated IL-6 level might be useful for identification of a high-risk subgroup of patients who are protected by low-molecular-weight heparin treatment while waiting for invasive treatment.

The fact that increased IL-6 levels were not predictive for the combined end point of death or MI is worthy of comment. Considering that the majority of deaths within a year from a severe unstable episode are likely due to cardiac causes, it seems that increased IL-6 levels indicate an inflammatory condition that may result in a higher risk of death from the index MI as well as from a subsequent MI. We found that patients presenting with elevated IL-6 levels were older and were characterized by a short angina history, no statin treatment on admission, low cholesterol levels at admission, myocardial

### Table 2. Outcome in Relation to Interleukin 6 (IL-6) Levels

<table>
<thead>
<tr>
<th>Strategy</th>
<th>No. Affected (%)/</th>
<th>Comparison of IL-6 ≥5 ng/L and IL-6 &lt; 5 ng/L</th>
<th>Comparison of IL-6 ≥5 ng/L and IL-6 &lt; 5 ng/L</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. in Group</td>
<td>RR (95% CI) P Value</td>
<td>RR (95% CI) P Value</td>
</tr>
<tr>
<td>Noninvasive</td>
<td>IL-6 ≥5 ng/L</td>
<td>25 (7.9)/318 19 (2.3)/838 3.47 (1.94-6.21) &lt;.001</td>
<td>53 (16.7)/318 112 (13.4)/838 1.25 (0.92-1.68) .15</td>
</tr>
<tr>
<td></td>
<td>IL-6 &lt; 5 ng/L</td>
<td>9 (2.8)/324 16 (1.9)/826 1.43 (0.64-3.21) .38</td>
<td>37 (11.4)/324 81 (9.8)/826 1.17 (0.81-1.68) .42</td>
</tr>
<tr>
<td>Invasive</td>
<td>IL-6 ≥5 ng/L</td>
<td>9 (2.8)/324 16 (1.9)/826 1.43 (0.64-3.21) .38</td>
<td>37 (11.4)/324 81 (9.8)/826 1.17 (0.81-1.68) .42</td>
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</tr>
</tbody>
</table>

Comparison of invasive vs noninvasive

<table>
<thead>
<tr>
<th>RR (95% CI)</th>
<th>P value</th>
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<tbody>
<tr>
<td>0.35 (0.17-0.74)</td>
<td>.004</td>
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<tr>
<td>0.85 (0.44-1.65)</td>
<td>.64</td>
</tr>
<tr>
<td>0.69 (0.46-1.01)</td>
<td>.06</td>
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<td>0.73 (0.56-0.96)</td>
<td>.02</td>
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</table>

Comparison placebo vs dalteparin

<table>
<thead>
<tr>
<th>RR (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.55 (0.28-1.09)</td>
<td>.08</td>
</tr>
<tr>
<td>1.27 (0.71-2.29)</td>
<td>.42</td>
</tr>
<tr>
<td>0.82 (0.54-1.26)</td>
<td>.37</td>
</tr>
<tr>
<td>1.08 (0.83-1.40)</td>
<td>.56</td>
</tr>
</tbody>
</table>

*RR indicates relative risk; CI, confidence interval.

**Figure 2. Mortality in the Interventional Study Related to Interleukin 6 (IL-6) levels**

Twelve-month probability of death in the invasive and noninvasive cohorts categorized by IL-6 plasma levels. Vital status was unavailable for 1 patient who requested to be withdrawn from the follow-up. In the patients with low IL-6 levels, there were 16 events among the 826 patients at baseline in the invasive group and 19 events among the 383 patients in the noninvasive group at 12 months. In the patients with high IL-6 levels, there were 9 events among the 324 patients in the invasive group and 25 events among the 318 patients in the noninvasive group at 12 months.

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damage as indicated by increased troponin T levels, and inflammatory activity reflected by elevated levels of C-reactive protein. These observations suggest that IL-6, which is present in the atheroma and secreted by endothelial cells, smooth muscle cells, macrophages, and T cells, may reflect a greater atherosclerotic burden as well as increased inflammatory activity in the plaques. These would subsequently be more vulnerable and prone to deeper thrombotic episodes and myocardial damage. The ischemic or necrotic myocardium could also be a source of cytokines. We saw a moderate but significant correlation between levels of troponin T and IL-6 (Spearman correlation coefficient = 0.38, P < .001), which may be explained by this reasoning. However, IL-6 levels as a predictor of death was additive to but independent of troponin T. Thus, our results further the understanding that unstable CAD is an inflammatory disorder.

One may also regard the issue from the opposite point of view: IL-6 reflects an ongoing low-grade inflammation other than the atherosclerotic disease and from there contributes to the progression of CAD. In 2 recent review articles, collected evidence and indices for a central role of IL-6 in the development of coronary heart disease are presented, taking into account the pleiotropy of this cytokine and its wide range of actions, including effects on platelets, endothelium, factors of metabolism, and coagulation.

The method of analysis that we used to measure plasma levels of IL-6 can be considered as rather insensitive. ELISA-based methods can detect levels of as little as 0.1 ng/L of IL-6, whereas we had a limit of detection already at 5 ng/L. Possibly, a more sensitive IL-6 assay method would have revealed more detailed prognostic information.

We also relied on a single blood sample per patient, taken at varying times after the last episode of chest pain. This could act as a confounder given the relatively short half-life of IL-6 in plasma (4 hours). Still, our results very clearly showed a pronounced difference in mortality using 5 ng/L as a cutoff level and the effect of sample time in predicting high IL-6 levels was moderate (data not shown). Only the latest sample time quartile had a slightly lower frequency of elevated levels.

In conclusion, we showed that circulating IL-6 is a strong independent marker of increased risk for mortality in patients with unstable CAD and that while patients with high plasma levels of IL-6 have the highest mortality rates, they also benefit most from a strategy of early revascularization. Prolonged treatment with subcutaneous dalteparin reduces the risk of death or MI during the first 60 days of treatment in patients with elevated levels of IL-6 and could be used while these patients await invasive treatment. Thus, circulating IL-6 provides important additional information when added to established indicators for risk stratification in patients with unstable CAD.

Author Contributions: Dr Siegbahn, as the principal investigator of this project, had full access to all of the data and attests to the validity of the analysis. All coauthors had access to the data.

Study concept and design: Diderholm, Wallentin, Siegbahn.
Acquisition of data: Lindmark, Diderholm, Wallentin, Siegbahn. Analysis and interpretation of data: Lindmark, Wallentin, Siegbahn. Drafting of the manuscript: Lindmark, Siegbahn. Critical revision of the manuscript for important intellectual content: Lindmark, Diderholm, Wallentin, Siegbahn. Statistical expertise: Wallentin, Siegbahn. Obtained funding: Wallentin, Siegbahn. Administrative, technical, or material support: Lindmark, Diderholm, Wallentin, Siegbahn. Study supervision: Wallentin, Siegbahn. Funding/Support: The FRISC II study was supported by and performed in collaboration with the Pharmacia and Upjohn Co. The project organization within the research group was also supported by the Swedish Heart-Lung Foundation (Drs Wallentin and Siegbahn), by grant k2001-32GX-11568-06 from the Swedish Medical Research Council (Dr Siegbahn), and by the Ake Wiberg Foundation (Drs Lindmark and Siegbahn).

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