

## Lipoprotein(a) and Coronary Heart Disease Meta-Analysis of Prospective Studies

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**Background**—Studies of the association between the plasma concentration of lipoprotein(a) [Lp(a)] and coronary heart disease (CHD) have reported apparently conflicting findings. We report a meta-analysis of the prospective studies with at least 1 year of follow-up published before 2000.

**Methods and Results**—The following information was abstracted for each study: geographical location of study, size, type of cohort (population-based or selected because of previous disease), mean age, follow-up duration, blood storage temperature and duration, assay methods, degree of adjustment for potential confounders, and relationship of baseline Lp(a) measurement with subsequent CHD risk. There were 5436 deaths from CHD or nonfatal myocardial infarctions during a weighted mean follow-up of 10 years in the 27 eligible studies. Comparison of individuals in the top third of baseline plasma Lp(a) measurements with those in the bottom third in each study yielded a combined risk ratio of 1.6 (95% CI 1.4 to 1.8,  $2P < 0.00001$ ), with similar findings when the analyses were restricted to the 18 studies of general populations (combined risk ratio 1.7, 95% CI 1.4 to 1.9;  $2P < 0.00001$ ). Despite differences among studies in blood storage techniques and assay methods, there was no significant heterogeneity among the results from the 18 population-based studies or among those from the 9 studies of patients with previous disease. Lp(a) was only weakly correlated with classical vascular risk factors, and adjustment for those that had been recorded made little difference to the reported risk ratios.

**Conclusions**—These prospective studies demonstrate a clear association between Lp(a) and CHD, but further studies are needed to determine the extent to which this is causal. (*Circulation*. 2000;102:1082-1085.)

**Key Words:** lipoproteins ■ coronary disease ■ meta-analysis

Epidemiological studies of coronary heart disease (CHD) and blood concentrations of lipoprotein(a) [Lp(a)], a large protein attached to an LDL particle (Table), have yielded apparently conflicting results, ranging from a strongly positive association to no association at all.<sup>1-3</sup> A previous meta-analysis<sup>4</sup> involved  $\approx 1600$  CHD cases in 12 prospective studies, but since the publication of that review, the evidence has increased to  $>5400$  cases of CHD death or nonfatal myocardial infarction in 27 prospective studies. This article provides an updated meta-analysis of the published studies in which CHD events were recorded for some years after “baseline” blood collection. Such prospective studies should be less prone to bias than retrospective studies because they limit any influence of preexisting disease itself on blood concentrations of Lp(a).

### Methods

#### Search Methods and Data Abstraction

Prospective studies with at least 1 year of follow-up that reported before 2000 on correlations between blood concentrations of Lp(a) and CHD death or nonfatal myocardial infarction were sought by Medline (<http://igm.nlm.nih.gov>) searches; by scanning relevant

reference lists; by hand searching cardiology, epidemiology, and other relevant journals; and by discussion and correspondence with authors of relevant reports.<sup>5-31</sup> Computer searches used combinations of key words relating to disease (CHD, myocardial infarction, atherosclerosis, and vascular disease) and Lp(a). Articles in languages other than English were to be translated. The following information was abstracted according to a fixed protocol (with discussion and checks to resolve any discrepancies): geographical location of study, size, type of cohort (population-based or selected because of previous disease), mean age, follow-up duration, blood storage temperature and duration, assay methods, and degree of adjustment for potential confounders.

#### Studies Identified

Twenty-seven published prospective studies involving a total of 5436 CHD cases were identified, of which 9 involved patients with preexisting CHD,<sup>23,26-28,30,31</sup> diabetes,<sup>29</sup> or renal disease.<sup>24,25</sup> The studies were conducted in Nordic countries,<sup>9,12,15,17,18,20,22,23,28,31</sup> Germany,<sup>14,19</sup> the United Kingdom,<sup>8,21,27,30</sup> the United States,<sup>5-7,10,11,13,24,26,29</sup> Canada,<sup>16</sup> and Japan<sup>25</sup>; most involved only white participants, and 4 were “nested” within randomized trials.<sup>6,7,12,23</sup> The weighted mean age at the time of the CHD event was 61 years, which represented a weighted mean age at baseline of 51 years and a weighted mean follow-up of 10 years. A few studies made measurements in fresh samples,<sup>5,10,13,19,22,31</sup> but most stored the blood for some years at

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**Characteristics of Lp(a)**

Description	Apo(a) is a very large protein molecule wrapped around, and linked by a disulfide bond to, an LDL-like particle. The molecular weight of apo(a) can vary widely between individuals, depending on the number of its repeats.
Vascular effects	Largely unknown. As a protein with regions homologous to the active sites of plasminogen, apo(a) might enhance or inhibit fibrinolysis.
Main site of synthesis	Liver.
Main modulators	Apo(a) gene variants, acute-phase reactions (≈2-fold rise during inflammatory response).
Main correlates	Weakly associated with blood lipids, fibrinogen, and smoking. Values increase substantially after menopause.
Plasma concentrations	Highly skewed and highly variable across studies (eg, top, middle, and bottom third mean values of ≈100, 20, and 5 mg/dL). <sup>11</sup>
Self-correlation*	≈0.90

Apolipoprotein(a) [apo(a)] refers to the large protein component of the Lp(a) particle.

\*Correlation between 2 measurements taken some years apart in the same individuals.<sup>33</sup>

low temperatures (range -20°C to -90°C) before thawing and analyzing samples from those who had subsequently suffered a CHD event and from age- and sex-matched controls for nested case-control comparisons. Different studies used different methods to measure Lp(a) concentrations, including electrophoresis,<sup>5,10,13,22</sup> ELISA,<sup>6-8,11,12,14,16,17,20,25-30</sup> radioimmunoassay,<sup>9,21,23,24</sup> electroimmunodiffusion,<sup>19</sup> electroimmunoassay,<sup>31</sup> and immunoradiometric assay.<sup>15,18</sup> Most studies made adjustments for smoking, blood lipid concentrations, and other classical vascular risk factors, such as blood pressure (see Figure legend).

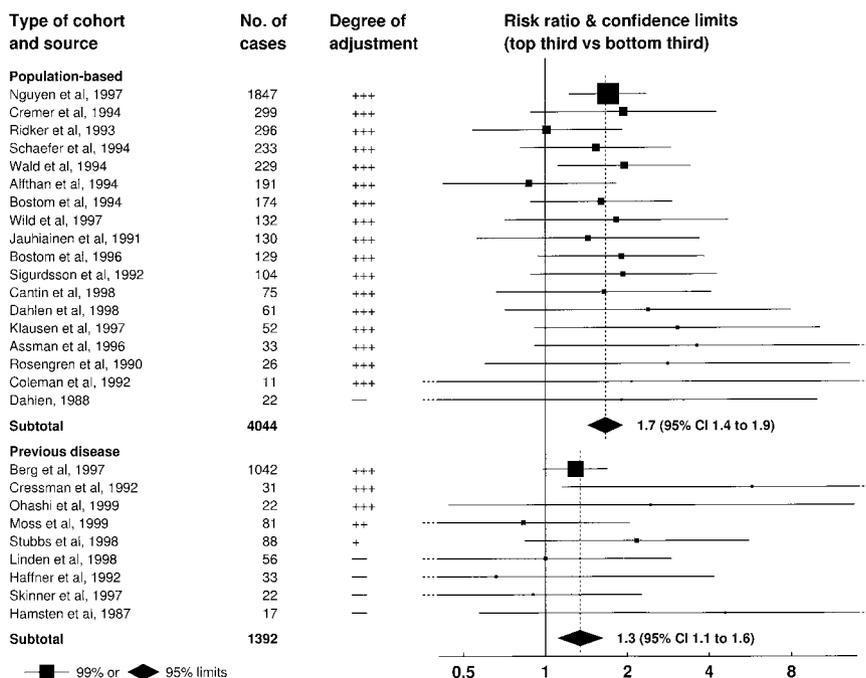
**Statistical Methods**

The present meta-analysis is based only on within-study comparisons, thereby avoiding any biases being caused by methodological

differences between studies. Plasma concentrations of Lp(a) had been measured by various assay methods, with different studies reporting risk ratios on the basis of different cutoff levels (including comparisons of mean values, thirds, quarters, fifths, etc) or as increases in risk for a given increase in Lp(a). The risk ratios derived for the present review compare individuals in the top third of single baseline measurements versus those in the bottom third; see previous publication for statistical details.<sup>32</sup> Studies based on the interpretation of electrophoretic bands<sup>5,10,13,22</sup> involved only a few Lp(a) categories. For the present meta-analysis, the highest was taken to correspond with the top third of baseline values; the lowest, with the bottom third. The standard error of the log risk ratio in each study was estimated from the number of standard errors by which the reported relationship differed from zero.<sup>32</sup> Summary estimates of the risk ratios from all studies were obtained by combining the separate estimates of inverse-variance-weighted log risk ratios from each study. This is valid even though different studies used different assay methods, because cases were compared directly only with controls within the same studies. CIs were obtained by normal approximations to the log relative risk, with 99% CIs used for the individual study results to take account of the increased scope for the play of chance in multiple comparisons and 95% CIs used for the overall results. Heterogeneity was assessed by standard  $\chi^2$  tests.

**Results**

The Figure displays the results for each separate study, with separate subtotals for studies in general populations and in patients with preexisting disease. Overall, in the 18 population-based cohorts, the combined risk ratio for the comparison of CHD rates in the top third of baseline Lp(a) measurement versus those in the bottom third was 1.7 (95% CI 1.4 to 1.9,  $2P < 0.00001$ ). There was no significant heterogeneity between the risk ratios in these 18 studies ( $\chi^2_{17} = 19.6, P > 0.1$ ). One of those 18 studies<sup>5</sup> accounts for almost half the cases, but the risk ratio of 1.66 (95% CI 1.41 to 1.95) in that study is almost identical to the risk ratio of 1.69 (95% CI 1.45 to 1.96) in the aggregated results of the others, and in none of the 17 smaller studies was the risk ratio significantly different from the overall result. The risk ratios



Prospective studies<sup>5-31</sup> of Lp(a) and CHD. Risk ratios compare top and bottom thirds of baseline measurements. Black squares indicate risk ratio in each study, with square size proportional to number of cases and horizontal lines representing 99% CIs. The combined risk ratios and their 95% CI are indicated by shaded diamonds. Degree of adjustment for possible confounders is denoted as follows: +, adjustment for age and sex only; ++, adjustment for the preceding plus smoking; +++, adjustment for the preceding plus some other classical vascular risk factors.

were also similar when population-based studies were grouped by various characteristics, including studies that made measurements in fresh samples versus those that froze and thawed samples before measurement (1.72 versus 1.59,  $\chi^2_1=0.4$ ;  $P>0.1$ ), those that stored samples for  $<5$  years versus that stored samples for  $\geq 5$  years (1.58 versus 1.59,  $\chi^2_1=0.3$ ;  $P>0.1$ ), or those that used ELISAs versus those that used other assay methods (1.65 versus 1.67,  $\chi^2_1=0.0$ ;  $P>0.1$ ).

Overall, in the 9 studies of patients who already had some disease at baseline, the combined risk ratio was 1.3 (95% CI 1.1 to 1.6,  $2P<0.001$ ), which is significantly smaller than that for the population-based studies ( $\chi^2_1=3.8$ ,  $P=0.05$ ). This result for studies in patients with preexisting disease is dominated by just one large study of patients with a history of myocardial infarction,<sup>23</sup> which accounted for three fourths of the evidence. The other 8 studies were all small; hence, although their results are all statistically compatible with a relative risk of 1.3 (and there was no significant heterogeneity among the 9 studies;  $\chi^2_8=9.7$ ,  $P>0.1$ ), they add little information to the one large study in patients with preexisting disease.

The published reports of these 27 studies contained insufficient information to produce combined analyses within subgroups defined by other potentially relevant characteristics (eg, age, sex, and fatal versus nonfatal CHD).

### Discussion

This meta-analysis of 27 prospective studies with information on 5436 CHD cases observed during mean follow-up of 10 years provides the most reliable assessment so far of the association between plasma Lp(a) and CHD. It indicates that people in the general population with Lp(a) levels in the top third of baseline measurement are at  $\approx 70\%$  increased risk of CHD compared with those in the bottom third. The risk ratio appeared to be less extreme in the 9 studies of patients with preexisting disease, but those studies may be less directly relevant than the population-based studies in the assessment of cause-and-effect relationships. Concentrations of plasma Lp(a) and classical vascular risk factors have not been found to be strongly correlated (Table), and these prospective studies of CHD generally reported little or no change in risk ratios after adjustment for other risk factors. This suggests that any effects of Lp(a) are unlikely to be accounted for by effects on, or confounding with, classical vascular risk factors.

In the present meta-analysis, cases were compared directly only with controls within the same studies, thereby avoiding any bias due to the use of different assay methods, reagents, and blood storage conditions among the different studies. There was no evidence of significant heterogeneity of the CHD risk ratios among the 18 population-based cohorts (or among the 9 cohorts defined on the basis of previous disease). However, several potential limitations of the present meta-analysis warrant attention. First, assays for Lp(a) are now widely available, so other prospective studies of Lp(a) and CHD that have not yet been reported may well exist. But, any exaggeration of the CHD risk ratio due to the preferential publication of reports with more extreme associations is unlikely to be substantial, particularly because the combined

risk ratio for the 17 smaller population-based studies was so similar to that for the single largest study.<sup>5</sup> Second, certain chronic conditions (such as inflammatory diseases) might both alter Lp(a) concentrations and be associated with CHD, but this would be expected to have only a small effect on the association of Lp(a) with CHD. Moreover, attempts in certain studies to control for the possible effects of preexisting disease on Lp(a) by the exclusion of those known to have certain conditions or by omission of any CHD events during the first few years of follow-up did not appear to change the reported associations substantially. Conversely, it is possible that the published studies have somewhat underestimated the role of Lp(a) in CHD. Only slight underestimation is likely to be due to the lack of correction in most studies for regression dilution,<sup>33</sup> because within-individual fluctuations of measured Lp(a) concentrations over time are relatively small (self-correlation  $\approx 0.9$ , Table). However, more substantial underestimation is possible if Lp(a) variants in people with higher blood concentrations were selectively degraded in studies with suboptimal blood storage conditions.<sup>34</sup> Moreover, there are great differences between individuals in the nature of Lp(a), with different isoforms of the protein differing widely in molecular weight (Table) and, hence, perhaps also in biological activity. Some studies have suggested stronger associations between CHD and certain apolipoprotein(a) isoforms than with others,<sup>35–37</sup> but this question has not yet been studied with appropriately large numbers.

More detailed combined analysis of the prospective studies of Lp(a), perhaps with the use of individual participant data from each study, could help to characterize the shapes of any dose-response relationship, reduce any bias related to the selection of particular cutoff levels, allow more complete adjustment for other risk factors, and assess associations in particular subgroups. If practicable treatments become available that substantially lower Lp(a),<sup>38</sup> then randomized trials might eventually become relevant to assessing the causal nature of this association (and, hence, the potential for reducing CHD risk). Studies of CHD and genetic polymorphisms that are associated with Lp(a) concentrations may also help to elucidate causality. Thus, much further study is still needed to determine the relevance of Lp(a) in the causation of CHD, but at least the existence of a moderately strong association of Lp(a) with CHD, independent of the standard vascular risk factors, is now clearly established.

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