Relation Between Serum Albumin and Carotid Atherosclerosis
The NHLBI Family Heart Study

Luc Djoussé, MD, DSc; Kenneth J. Rothman, DrPH; L. Adrienne Cupples, PhD; Donna K. Arnett, PhD; R. Curtis Ellison, MD

Background and Purpose—Lower concentrations of albumin have been positively related to coronary disease. The aim of this project was to assess the association between serum albumin and carotid atherosclerosis.

Methods—B-mode ultrasound was used to assess carotid plaques and intima-media thickness (IMT) among white subjects from 592 randomly ascertained families in the National Heart, Lung, and Blood Institute Family Heart Study. Logistic regression was used to estimate prevalence odds ratios of any carotid plaques.

Results—Of the 2072 persons studied, 47% were men. Higher serum albumin was associated with younger age, lower body mass index, and lower prevalence of hypertension and coronary disease. Lower serum albumin levels were not associated with an increased odds of carotid plaques. From the lowest to the highest quartile of albumin, multivariate adjusted odds ratios for the prevalence of any carotid plaques were 1.05 (95% CI, 0.59 to 1.86), 1.34 (95% CI, 0.78 to 2.32), 1.03 (95% CI, 0.52 to 1.86), and 1.0 (reference), respectively, in men and 0.71 (95% CI, 0.40 to 1.26), 0.76 (95% CI, 0.46 to 1.36), and 1.0, respectively, in women. Similarly, in a linear regression model adjusted for demographic, lifestyle, and metabolic factors, serum albumin was not associated with carotid IMT. When the first 3 were compared with the highest quartile of serum albumin, regression coefficients for internal carotid IMT were 0.06458 (SE, 0.06408), 0.07205 (SE, 0.05469), and 0.000773 (SE, 0.05687), respectively, for men and −0.01795 (SE, 0.05080), −0.08501 (SE, 0.04800), and 0.009528 (SE, 0.04622), respectively, for women.

Conclusions—Our data suggest that lower serum albumin levels are not associated with an increased odds of prevalent carotid atherosclerosis in either men or women. (Stroke. 2003;34:53-57.)

Key Words: albumins ■ arteriosclerosis ■ carotid arteries

Stroke is the third-leading cause of death in the United States and is associated with a high burden of healthcare costs.1 Carotid artery atherosclerosis is a major risk factor for stroke.2–4 Early signs of atherosclerosis of the carotid arteries can be assessed through noninvasive ultrasound techniques. Epidemiological studies have suggested that lower concentrations of serum albumin are associated with an increased risk of cardiovascular disease (CVD).5–8 but the findings remain inconsistent. The association between albumin and early mortality suggests that serum albumin level is a predictor of subclinical disease. Subclinical atherosclerosis is frequently measured noninvasively by ultrasound in superficial large arteries such as the carotid. To date, limited data are available on the relation between concentrations of albumin and carotid artery disease, and results from those findings have been inconsistent.9–13

Albumin is synthesized in the liver, and circulating cytokines influence its production. Interleukin (IL)-6, a cytokine that has both proinflammatory and anti-inflammatory properties, is released in response to IL-1 and tumor necrosis factor-α (inflammatory response).14 This release leads to an acute-phase reaction in which the synthesis of albumin is depressed and the synthesis of acute reactant proteins (C-reactive protein, amyloid A, and α-1 acid glycoprotein) is increased.14 An association between inflammatory cytokines and serum albumin has been shown in experimental and clinical studies.14–16

We used data collected on participants from the National Heart, Lung, and Blood Institute (NHLBI) Family Heart Study to evaluate whether lower serum concentrations of albumin are associated with early signs of atherosclerosis of the carotid arteries as measured by the presence of plaques or increased thickness of the carotid artery walls.}

Materials and Methods

Study Population
The NHLBI Family Heart Study is a multicenter, population-based study designed to identify and evaluate genetic and
nongenetic determinants of coronary heart disease (CHD), preclinical atherosclerosis, and cardiovascular risk factors. A detailed description of the methods and design has been reported. Briefly, the subjects described in here represent members of families chosen from previously established population-based cohort studies (referred to as parent studies): the Framingham Heart Study in Framingham (Mass), the Atherosclerosis Risk in Communities’ (ARIC) cohorts in North Carolina and Minnesota, and the Utah Health Family Tree Study in Salt Lake City. From 1993 to 1995, individuals participating in each of the parent studies were selected at random and invited to furnish an updated family health history that contained information on their parents, children, and siblings. Of the 4679 individuals contacted, responses were obtained from 3150 (67%); their other family members were then contacted, and self-reported health data were obtained from a total of 22,908 individuals (86% of those contacted).

Of the families furnishing data, 592 were chosen at random (subjects from these families are referred to as the random sample), and 661 were chosen because of a higher-than-expected risk of CHD among family members. Individuals from these families are referred to as the high-risk sample. Of the total study participants, 94.8% were European American and 4.3% were African American. All members of both groups of families were invited to come to 1 of the 4 study clinics for an ~4-hour clinical evaluation. The evaluation included a detailed medical and lifestyle history obtained through personal interview and clinical tests, including an ultrasound of the carotid arteries. Each study participant provided informed consent, and the study protocol was reviewed and approved by the institutional review board at each of the participating institutions. The present study was limited to 2072 European Americans subjects from the 592 randomly ascertained families for whom complete data were available.

### Carotid Intima-Media Thickness Measurement

Using a high-resolution B-mode ultrasound, trained technicians measured carotid intima-media thickness (IMT) according to the ARIC protocol. Carotid artery assessment was completed bilaterally on 3 segments: the common carotid artery (1 cm proximal to the dilatation of the carotid bulb), bifurcation (the 1-cm segment proximal to the flow divider), and internal carotid artery (the 1-cm segment in the internal branch distal to the flow divider). B-mode images were recorded on high-resolution cassette. Recorded images were read at the central reading station by trained readers. The presence of an atherosclerotic plaque was determined from 3 criteria: irregularity of surface, increased overall thickness, and echogenicity. A plaque was ascertained at the visualized segment if at least 2 of these criteria were present. For each subject, the total number of plaques was recorded. For each of the 3 segments, the presence of a carotid atherosclerotic plaque was ascertained if at least 1 plaque was detected in any of the segments visualized. Reproducibility for the presence of any carotid plaques was very good (k statistic, 0.76). We also evaluated the relation of serum albumin to carotid IMT as a continuous outcome. We used the general linear model (Proc Mixed in SAS) to estimate adjusted means of IMT. From the lowest to highest quartile of serum albumin, the risk of carotid plaques increased odds of carotid plaques in either men or women (Table 2). From the highest to lowest quartile of serum albumin, the odds ratios for any carotid plaques within each sex: The full model controlled for age, field center, body mass index (BMI; quartiles), ratio of HDL to total cholesterol, creatinine, diabetes mellitus, hypertension, CHD, smoking (4 categories), and alcohol intake (3 categories). To account for familial correlation between sibships, we used generalized estimating equations for prevalent carotid plaques and a generalized linear model (Proc Mixed in SAS) to estimate adjusted means of IMT across quartiles of albumin. The multivariate model controlled for all factors mentioned above. All analyses were completed with PC SAS.

### Blood Collection and Assays

All participants were asked to fast for 12 hours before arrival at the study center. Evacuated tubes without additives were used to collect samples for lipids; blood samples were then spun at 3000g for 10 minutes at 4°C. Sera were stored at −70°C until shipped periodically to a central laboratory at the Fairview-University Medical Center in Minneapolis (Minn) for processing. Serum albumin was measured by a thin film adaptation of a bromcresol green colorimetric procedure. Serum total cholesterol was measured with a commercial cholesterol oxidase method. Serum creatinine was measured by a thin film adaptation of the amidoetchase enzymatic method with the Vitros analyzer (Johnson & Johnson Clinical Diagnostics Inc). Glucose was measured by a thin film adaptation of a glucose oxidase enzymatic, spectrophotometric procedure with the Vitros analyzer (Johnson & Johnson Clinical Diagnostics Inc). High density lipoprotein (HDL) cholesterol quantification was performed with the cholesterol method described above after precipitation of non–HDL cholesterol with magnesium/dextran. For samples with triglyceride levels <4.51 mmol/L, low-density lipoprotein (LDL) cholesterol was calculated with the Friedwald formula. For subjects with higher levels of triglycerides, LDL cholesterol quantitation was performed on EDTA plasma by ultracentrifugation (Beckman). Triglycerides were measured with Triglyceride G1 reagent on the Roche COBAS FARA centrifugal analyzer (Boehringer Mannheim Corp).

### Other Variables

Information on cigarette smoking was obtained by questionnaire. This information was used to classify each participant as a never smoker, former smoker, or current smoker. Alcohol information was self-reported. The average number of drinks (beer, spirits, and wine) consumed per week on average over the past 12 months was recorded. Anthropometric data were collected with subjects wearing scrub suits. A balance scale was used to measure body weight, and height was measured with a wall-mounted vertical ruler. Information on physical activity (minutes per day of various levels of leisure activity) was obtained by interview.

### Statistical Analysis

Within each sex, we created quartiles of albumin because of the following: (1) We did not assume a linear relation between serum albumin and carotid artery plaques or IMT, and (2) the risk of CVD is higher among men than women. We created 3 indicator variables using the highest category of albumin as reference.

We used logistic regression to estimate the prevalence odds ratio of any carotid plaques within each sex. The full model controlled for age, field center, body mass index (BMI; quartiles), ratio of HDL to total cholesterol, creatinine, diabetes mellitus, hypertension, CHD, smoking (4 categories), and alcohol intake (3 categories). To account for familial correlation between sibships, we used generalized estimating equations for prevalent carotid plaques and a generalized linear model (Proc Mixed in SAS) to estimate adjusted means of IMT across quartiles of albumin. The multivariate model controlled for all factors mentioned above. All analyses were completed with PC SAS.

### Results

Of the 2072 subjects included in the analyses, the average age was 52.2 years (range, 25 to 93 years), and 47% were men. Table 1 presents the baseline characteristics of the study subjects. Higher serum albumin was associated with younger age, lower BMI, and lower prevalence of diabetes mellitus, hypertension, and coronary disease.

Low serum albumin was not associated with an increased odds of carotid plaques in either men or women (Table 2). From the highest to lowest quartile of serum albumin, the odds ratios for any carotid plaques were 1.0 (reference), 1.03 (95% CI, 0.97 to 1.09), and 1.05 (95% CI, 0.78 to 1.39), respectively, for men with adjustment for age, center, BMI, ratio of HDL to total cholesterol, creatinine, diabetes mellitus, hypertension, coronary disease, smoking, and alcohol consumption. Corresponding odds ratios for women were 1.0, 0.89 (95% CI, 0.65 to 1.21), 1.07 (95% CI, 0.79 to 1.46), and 0.96 (95% CI, 0.72 to 1.31), respectively. For both men and women, CIs were wide and included 1. When IMT was analyzed as a continuous variable, there was little evidence for an association between serum albumin and IMT. From the lowest to highest quartile of serum albu-
TABLE 1. Characteristics of the Participants According to Quartiles of Serum Albumin Among Subjects in the NHLBI Family Heart Study

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Men</th>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean, g/L</td>
<td>37.8</td>
<td>37.0</td>
</tr>
<tr>
<td>Range</td>
<td>33–39.0</td>
<td>31.0–38.9</td>
</tr>
<tr>
<td>Number</td>
<td>210</td>
<td>248</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Quartiles of Albumin, g/L, Mean (range)</th>
<th>Cases/n</th>
<th>Odds Ratio (95% CI)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low (32.9–39.0)</td>
<td>80/210</td>
<td>0.98 (0.58–1.66) 1.05 (0.59–1.86)</td>
</tr>
<tr>
<td>39.1–41.0</td>
<td>90/277</td>
<td>1.23 (0.74–2.05) 1.34 (0.78–2.32)</td>
</tr>
<tr>
<td>41.1–43.0</td>
<td>51/258</td>
<td>0.84 (0.48–1.47) 1.03 (0.52–1.86)</td>
</tr>
<tr>
<td>High (43.1–51.0)</td>
<td>39/235</td>
<td>1.0</td>
</tr>
</tbody>
</table>

| Low (31.0–38.9)                        | 58/248  | 0.74 (0.43–1.28) 0.71 (0.40–1.26) |
| 39.0–40.0                               | 63/308  | 0.77 (0.44–1.32) 0.76 (0.42–1.36) |
| 40.1–42.0                               | 62/294  | 0.86 (0.52–1.42) 0.79 (0.46–1.36) |
| High (42.1–49.0)                        | 44/242  | 1.0                 |

*Odds ratios obtained through generalized estimating equations.
†The full model controlled for age (5-year categories), field center, body mass index, ratio of total to HDL cholesterol, creatinine, diabetes mellitus, hypertension, coronary disease, cigarette smoking (never, former smokers, current smokers of 1–10 cigarettes/d, and 11+ cigarettes/d), and alcohol consumption (nondrinkers, current drinkers 1–7 drinks/d, 8+ drinks/d).

Discussion

Previous epidemiological studies have suggested that lower levels of serum albumin are associated with an increased risk of CVD and mortality. It is not known whether low serum albumin is associated with early signs of atherosclerosis, as would be reflected in the carotid arteries, as assessed by high-resolution ultrasound. In the present study, we demonstrated that serum albumin showed little association with prevalent carotid plaques or IMT in either men or women. Our report is consistent with the findings of Folsom et al., who found no association between serum albumin and carotid IMT in a community-based population. In addition, Stenvinkel and colleagues did not find an association between serum albumin and carotid atherosclerosis among subjects with renal disease. Specifically, albumin was inversely related to carotid plaques among dialysis patients and in subjects with end-stage renal disease. Thus, although an inverse relation has been observed between serum albumin and carotid atherosclerosis among subjects with impaired renal function, no such relation has been reported in a community-based cohort.

Because the association between serum albumin and the risk of CVD has been inconsistent, it has been suggested that low serum albumin might not be an independent risk factor of CVD and mortality but rather a reflection of cytokine activation and ongoing IL-6–mediated subclinical inflammation. Atherosclerosis can be viewed as an inflammatory disease. It is thus possible that low albumin merely reflects the inflammatory process and is not causally related to the development of carotid atherosclerosis.
related to atherosclerosis, CVD, and mortality. On the other hand, several studies have reported an association between serum albumin and mortality.25–27,29 If low albumin were only an indication of an existing inflammation, one would not expect to observe an association between low albumin and mortality in the absence of inflammation. Reuben et al27 found that among subjects without evidence of IL-6–mediated inflammation, having lower albumin was associated with a 2-fold increase in 4-year mortality compared with having higher albumin. It is reasonable to postulate that other biologic mechanisms may be involved in the association of albumin and atherosclerosis, CVD, and mortality and that the lack of an association in our study does not preclude a causal relation between low albumin and atherosclerosis.

Our study had some limitations. First, because of its cross-sectional nature, we had only a single evaluation of the carotid arteries. Thus, it is difficult to distinguish changes of the carotid artery walls that may have preceded changes in serum albumin from those that may be consequences of low albumin. Second, despite the use of highly trained ultrasound technicians, a potential source of bias is nondifferential measurement error of the IMT, which could partially explain our findings. Third, selection bias and residual confounding by unmeasured factors might have influenced our findings. Fourth, the wide CIs make it difficult to exclude a possible association between low albumin and carotid atherosclerosis. Finally, the generalizability of our findings is limited given the restriction to European Americans subjects. Nevertheless, the large sample size, availability of data on chronic conditions that affect serum albumin, wide age range, and multicenter nature of the study population are strengths of this study.

In conclusion, our data suggest that serum albumin may not be associated with carotid plaques or carotid IMT. However, because the odds ratios for carotid plaques in men were above unity and had wide CIs, we cannot exclude an association between serum albumin and carotid atherosclerosis in men, and replication in larger studies is warranted.

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