Serum high sensitivity C-reactive protein and cognitive function in elderly women

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Abstract

Background: inflammation has been linked to cognitive impairment. However, limited data are available on the association between inflammatory markers and cognitive function.

Objectives: we tested the hypothesis that elevated serum concentration of high sensitivity C-reactive protein (hs-CRP), an established marker of low-grade inflammation, predicts cognitive impairment in elderly women.

Design: a 12-year population-based follow-up study.

Participants: a total of 97 women between 60 and 70 years of age at baseline.

Methods: serum hs-CRP concentration was measured by a high sensitivity assay. Global cognitive function was measured with the Mini-Mental State Examination (MMSE), and memory and cognitive speed were measured with a detailed cognitive test battery.

Results: higher baseline hs-CRP was associated with poorer memory at 12-year follow-up without adjustment and after adjustment for age, education and depression (standardised regression coefficient \( \beta = -0.842, 95\% \) confidence interval \(-1.602 \) to \(-0.083, P = 0.030 \)), and further adjustment for the use of hormone replacement therapy, smoking, serum LDL cholesterol and body mass index (standardised regression coefficient \( \beta = -0.817, 95\% \) confidence interval \(-1.630 \) to \(-0.004, P = 0.049 \)). Memory at 12-year follow-up worsened linearly with increasing hs-CRP at baseline (\( P = 0.048 \) for linear trend). There was no association between hs-CRP at baseline and cognitive speed or MMSE score at 12-year follow-up.

Conclusions: high serum hs-CRP concentration predicts poorer memory 12 years later in elderly women. Hs-CRP may be a useful biomarker to identify individuals at an increased risk for cognitive decline.

Keywords: cognitive function, high sensitivity CRP, elderly women

Introduction

Inflammatory mechanisms have been suggested to be involved in cognitive impairment and dementia [1, 2]. In addition, inflammation has been linked to the pathogenesis of cardiovascular disease [3, 4], obesity and insulin resistance [5], which in turn have been associated with the risk of cognitive impairment [6, 7].

C-reactive protein has been found in and around beta-amyloid plaques in the brains of patients with dementia.
Methods

Study population

The subjects of the present study were derived from a population-based, randomly selected sample of 299 women 50–60 years of age who were examined as a part of the large risk factor survey in Finland in 1982 [18]. The women were invited for re-examinations in 1991, and altogether 202 women aged 60–70 years completed the examinations between October 1991 and March 1992 (baseline of the present study). Because 32 women had died or could not be contacted, 170 women aged 70–80 years were eligible for the follow-up study in 2003. Of these 170 women, 57 did not participate. The reasons for not participating included musculoskeletal problems (n = 11), dementia (n = 2), stroke (n = 1), cancer (n = 1), death (n = 4), other health problems (n = 18), unwillingness to participate (n = 16), and unknown reasons (n = 4). Thus, 113 women completed all study visits in 2003. The final study sample consisted of 97 women with complete data in both examinations. None of the women had type 2 diabetes at baseline. The study protocol was approved by the Ethics Committee of the University of Kuopio, Finland. All participants provided a written informed consent.

The 57 non-participants were older (65.2 vs. 63.8 years, P = 0.003) and had less education (7.2 vs. 8.7 years, P = 0.003), a lower Mini-Mental State Examination (MMSE) score (27.5 vs. 28.7 points, P = 0.004), a higher body mass index (BMI) (29.3 vs. 27.3 kg/m², P = 0.005), a greater waist circumference (87.9 vs. 82.9 cm, P = 0.003), elevated diastolic blood pressure levels (92.0 vs. 88.8 mm Hg, P = 0.048), elevated serum triglyceride concentrations (128.1 vs. 103.5 mg/dl, P = 0.009), and reduced serum high-density lipoprotein (HDL) cholesterol concentrations (59.4 vs. 63.5 mg/dl, P = 0.047) at baseline than the participants. Smoking, alcohol consumption, the use of hormone replacement therapy, depression score, blood glucose concentrations, serum low-density lipoprotein (LDL) cholesterol concentrations, or the prevalence of cardiovascular disease did not differ between the participants and the non-participants.

Biochemical analyses

Venous blood samples were taken at baseline and 12-year follow-up after a 12-h fast and were stored at −80°C until analysis. Serum hs-CRP concentrations were measured by a commercial immunoassay using the IMMULITE 2000 Analyzer (IMMULITE 2000 High-Sensitivity CRP, Diagnostic Products Corp., Los Angeles, CA, USA). In the 97 participants, the intra-class correlation between two hs-CRP assays during the same study visit was >0.999 at baseline and >0.997 at 12-year follow-up. The coefficient of variation was 1.7% at baseline and 3.6% at 12-year follow-up.

Serum cholesterol and triglyceride concentrations were measured by enzymatic colorimetric methods. Serum HDL cholesterol concentration was measured using an enzymatic colorimetric method in a supernatant after precipitation with dextran sulphate and MgCl₂. Serum LDL cholesterol was calculated by the Friedewald formula. Hexokinase method was used for blood glucose analyses.

Assessment of cognitive function

Global cognitive function at baseline and 12-year follow-up was assessed by the MMSE score [19]. At follow-up, cognitive function was assessed in more detail using measures of memory and cognitive speed. Memory was assessed using the Word Recall Test with three word lists, including 10 words matched for word frequency. The score of immediate recall was the total number of correct words in three word lists [20, 21]. In a prospective memory task, at the beginning of the test session, the subject was asked to remind the investigator that the subject must sign a paper at the end of the test session [22]. Cognitive speed was assessed with Stroop Test using time needed to read the normal text with colour names and naming of colour dots on a sheet of paper, higher scores indicating poorer cognitive speed [23]. Cognitive speed was also assessed with the Letter-digit Substitution Test, lower scores indicating poorer speed [24].

Other assessments

Diseases diagnosed by a doctor (no vs. yes), medications (no vs. yes), smoking (no vs. yes), alcohol consumption (drinks/week), and physical activity (sessions/week) were assessed by a self-administered questionnaire at baseline and follow-up. The use of hormone replacement therapy (no vs. yes) and education (years) were assessed at baseline. Depressive symptoms were assessed using the Zung self-report 20-item scale [25]. The item scores were summed up with a maximum score of 80 points, >50 points indicating depressive symptoms [25]. Body weight, height, waist circumference and blood pressure were measured according to the MONICA protocol. BMI was calculated as weight divided by height squared (kg/m²).
Statistical methods

The scores of memory and cognitive speed were converted to standardised $z$-scores. The $z$-values for the Letter-digit Substitution Test were reversed. A sum of $z$-scores from the Word Recall Test and the prospective memory test was computed to describe memory. Similarly, a sum of $z$-scores from the Letter-digit Substitution Test and the Stroop Test was computed to describe cognitive speed. The MMSE score of $\geq 25$ was defined as a good general cognition. This cut-off point was used in the analyses.

Serum hs-CRP concentration was categorised as low(<1.0 mg/l), average (1.0–3.0 mg/l), and high (>3.0 mg/l) according to the statement for healthcare professionals from the Centers for Disease Control and Prevention and the American Heart Association [3]. Differences in the characteristics of women with low, average or high hs-CRP levels were analyzed using analysis of variance or $\chi^2$ test as appropriate. The associations of hs-CRP with memory and cognitive speed as continuous variables were analyzed with linear regression analyses. The differences in the means of memory among the hs-CRP tertiles were tested using analysis of covariation, and the linear trend across the thirds was derived from linear regression analysis. Analyses were first adjusted for age, education, depression (model 1), and then additionally for the use of hormone replacement therapy, smoking and LDL cholesterol and BMI (model 2). These variables were chosen for the analyses based on their known association with inflammation and cognition as well as the association with hs-CRP in the present study ($P<0.10$). To avoid co-linearity, highly correlated variables were not allowed in the models. However, we re-run the analyses controlling also for fasting blood glucose (correlated significantly with LDL cholesterol) and systolic blood pressure (correlated significantly with hormone replacement therapy) at baseline.

Because the distribution of hs-CRP was skewed at baseline and 12-year follow-up, logarithmic transformation was used. Because, the distribution of MMSE was skewed, even after logarithmic transformation, logistic regression analysis was used. We used the level of statistical significance of $P<0.05$.

The analyses were conducted using the SPSS for Windows, Release 11.5 (SPSS Inc., Chicago, IL).

Results

Characteristics of subjects

At baseline, the median serum hs-CRP was 1.12 mg/l, ranging from 0.01 to 70.14 mg/l (interquartile range 0.53 to 2.42). Women with high serum hs-CRP had more depressive symptoms, higher fasting blood glucose, higher systolic blood pressure levels, and larger BMI and waist circumference, and they were less likely to use hormone replacement therapy than those with low hs-CRP (Table 1). There was no difference in baseline MMSE across the baseline hs-CRP groups (Table 1).

At 12-year follow-up, the median serum hs-CRP was 1.02 mg/l, ranging from 0.01 to 22.11 mg/l (interquartile range 0.36 to 2.66). The median hs-CRP ($P = 0.506$) or number of women in the three hs-CRP groups ($P = 0.708$) did not differ at baseline and at 12-year follow-up.

At 12-year follow-up, hs-CRP was associated with BMI (26.3 ± 4.3 kg/m$^2$ in low, 29.5 ± 3.9 kg/m$^2$ in average, and 29.5 ± 5.6 kg/m$^2$ in high hs-CRP groups, $P = 0.010$ between groups) and waist circumference (87.9 ± 9.7 cm in low, 95.9 ± 10.6 cm in average, and 95.2 ± 12.3 cm in high hs-CRP groups, $P = 0.002$ between groups). The mean MMSE score decreased from 28.7 (SD ±1.8) at baseline to 26.0 (SD ±2.3) at 12-year follow-up ($P<0.001$).

Longitudinal association between hs-CRP and cognitive function

Higher baseline hs-CRP was associated with poorer memory (sum of $z$-scores from the Word Recall Test and the prospective memory test) at 12-year follow-up (Table 2). Additional adjustment for fasting blood glucose and systolic blood pressure slightly weakened the association ($\beta = -0.758$, 95% confidence interval $-1.585$ to 0.070, $P = 0.072$). Memory at 12-year follow-up worsened linearly with increasing hs-CRP at baseline (Figure 1). There was no association between hs-CRP at baseline and cognitive speed (Table 2) or MMSE (OR 0.56, 95% confidence interval 0.256 to 1.323, $P = 0.150$) at 12-year follow-up.

Cross-sectional association between hs-CRP and cognitive function

After 12 years, higher hs-CRP was associated with poorer memory without adjustment and after adjustment for age, education and depression (standardised regression coefficient $\beta = -0.757$, 95% confidence interval $-1.491$ to $-0.023$, $P = 0.043$) and further adjustment for the use of hormone replacement therapy, smoking, serum LDL cholesterol and BMI ($\beta = -0.709$, 95% confidence interval $-1.368$ to 0.006).
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Table 1. Characteristics of women with low, average and high serum hs-CRP concentrations at baseline

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>CRP &lt;1.0 mg/l</th>
<th>CRP 1.0–3.0 mg/l</th>
<th>CRP &gt;3.0 mg/l</th>
<th>P-value between groups</th>
</tr>
</thead>
<tbody>
<tr>
<td>63.6 ± 3.1</td>
<td>63.7 ± 2.7</td>
<td>64.2 ± 3.3</td>
<td>0.747</td>
<td></td>
</tr>
<tr>
<td>Education</td>
<td>8.7 ± 4.0</td>
<td>8.7 ± 3.1</td>
<td>8.7 ± 4.1</td>
<td>0.808</td>
</tr>
<tr>
<td>Depression</td>
<td>33.2 ± 4.9</td>
<td>34.6 ± 6.3</td>
<td>40.9 ± 4.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mini-Mental State Examination (points)a</td>
<td>28.7 ± 2.0</td>
<td>28.7 ± 1.7</td>
<td>28.7 ± 1.7</td>
<td>0.980</td>
</tr>
<tr>
<td>Alcohol</td>
<td>0.3 ± 0.9</td>
<td>0.3 ± 0.8</td>
<td>0.4 ± 1.3</td>
<td>0.895</td>
</tr>
<tr>
<td>Smoking</td>
<td>0</td>
<td>15</td>
<td>11</td>
<td>0.036</td>
</tr>
<tr>
<td>Physical activity (&gt;2 times/week) (%)</td>
<td>57</td>
<td>70</td>
<td>72</td>
<td>0.349</td>
</tr>
<tr>
<td>Hormone replacement therapy (%)</td>
<td>39</td>
<td>24</td>
<td>11</td>
<td>0.065</td>
</tr>
<tr>
<td>Cardiovascular disease (%)</td>
<td>20</td>
<td>18</td>
<td>17</td>
<td>0.962</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>26.5 ± 3.6</td>
<td>26.8 ± 3.3</td>
<td>30.2 ± 5.9</td>
<td>0.020</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>81.0 ± 8.4</td>
<td>82.5 ± 8.7</td>
<td>88.4 ± 14.2</td>
<td>0.073</td>
</tr>
<tr>
<td>Fasting blood glucose (mg/dl)</td>
<td>84.7 ± 7.8</td>
<td>84.2 ± 9.1</td>
<td>92.6 ± 15.1</td>
<td>0.040</td>
</tr>
<tr>
<td>Serum total cholesterol (mg/dl)</td>
<td>235.5 ± 34.0</td>
<td>245.8 ± 39.9</td>
<td>220.4 ± 46.1</td>
<td>0.083</td>
</tr>
<tr>
<td>Serum LDL cholesterol (mg/dl)</td>
<td>155.1±31.7</td>
<td>165.9±38.9</td>
<td>137.8±43.0</td>
<td>0.035</td>
</tr>
<tr>
<td>Serum HDL cholesterol (mg/dl)</td>
<td>63.6 ± 12.9</td>
<td>62.3 ± 9.8</td>
<td>65.1 ± 13.9</td>
<td>0.727</td>
</tr>
<tr>
<td>Serum triglycerides (mg/dl)</td>
<td>102.0 ± 31.9</td>
<td>104.3 ± 29.6</td>
<td>106.2 ± 48.3</td>
<td>0.920</td>
</tr>
<tr>
<td>Systolic blood pressure (mm Hg)</td>
<td>151.1 ± 21.6</td>
<td>156.8 ± 22.1</td>
<td>167.6 ± 21.1</td>
<td>0.027</td>
</tr>
<tr>
<td>Diastolic blood pressure (mm Hg)</td>
<td>87.3 ± 10.7</td>
<td>89.2 ± 9.5</td>
<td>91.8 ± 9.5</td>
<td>0.278</td>
</tr>
<tr>
<td>Drug treatment for hypertension (%)</td>
<td>28</td>
<td>15</td>
<td>56</td>
<td>0.010</td>
</tr>
<tr>
<td>Drug treatment for hypercholesterolaemia (%)</td>
<td>9</td>
<td>3</td>
<td>11</td>
<td>0.601</td>
</tr>
</tbody>
</table>

Values are means (± SD) from analysis of variance or percentages from χ² test.

a Data were available for 89 women.

SI conversion factors: To convert serum hs-CRP concentrations to nanomoles per litre (nmol/l), multiply by 8.45; to convert blood glucose to millimoles per litre, multiply by 0.055; to convert LDL, HDL, and total cholesterol to millimoles per litre, multiply by 0.0259; and to convert serum triglycerides to millimoles per litre, multiply by 0.0113.

Table 2. The association of baseline serum hs-CRP concentration with memory and cognitive speed at follow-up

<table>
<thead>
<tr>
<th>Model 1</th>
<th>Model 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Memory</td>
<td>β</td>
</tr>
<tr>
<td>-0.842</td>
<td>-1.602</td>
</tr>
<tr>
<td>Cognitive speed</td>
<td>-0.199</td>
</tr>
</tbody>
</table>

Data are from linear regression analyses. β denotes standardised regression coefficient.

Model 1: adjusted for age, education and depression. Model 2: adjusted for variables in model 1 and the use of hormone replacement therapy, smoking, serum LDL cholesterol and body mass index at the time of hs-CRP measurements.

Memory: sum of z-scores from the Word Recall Test and the prospective memory test.

Cognitive speed: sum of z-scores from the Letter-digit Substitution Test and the Stroop Test.

Discussion

The main finding of the present study is that high serum hs-CRP concentration predicts poorer memory 12 years later in elderly women. Thus, elevated serum hs-CRP levels may be a useful biomarker to identify individuals with an increased risk for memory impairment and eventually dementia.

The present results are in line with some previous studies indicating that low-grade inflammation is deleterious for cognitive function. In population-based studies, increased serum hs-CRP concentrations have been associated with poor memory [9], poor global cognitive performance [10, 11, 13], as well as vascular dementia [12] and Alzheimer’s disease [14, 15]. However, in some other studies no association between hs-CRP and cognition has been found [16, 17]. In the previous studies, the age of the subjects has varied between 30 and 85 years, the follow-up time has generally been quite short (up to 6 years), and the most commonly used outcome has been dementia. Few studies have had a follow-up time of 10 years or more [13, 14] or used a detailed evaluation of cognitive functions [9, 16]. In the present study, we used detailed evaluation of cognitive functions at the follow-up. Because of lack of data of normative ageing-related rates in specific cognitive
The present data may broaden the aspects on the association between inflammatory biomarkers and mild cognitive impairment. Mild cognitive impairment typically occurs several years before clinical diagnosis of dementia [27] and individuals with mild cognitive impairment have a high risk for progression to dementia, especially Alzheimer’s disease [26]. Also, the current conceptualisations of Alzheimer’s disease presume that the neurodegenerative changes begin well before the clinical manifestations of the disease become apparent [28]. Aging is associated with increased inflammatory activity [29], and intrathecal proinflammatory cytokines such as tumour necrosis factor, alpha and tau proteins, a marker for neurodegeneration, are increased in patients with mild cognitive impairment [30]. Thus, propensity towards inflammation in those patients may indicate that inflammation in central nervous system may be an early hallmark in the pathogenesis of Alzheimer’s disease [30]. Inflammatory mediators are mostly highly expressed in the vicinity of beta-amyloid deposits and neurofibrillary tangles, classical hallmarks where neurodegeneration is known to occur [2]. However, there are limited number of studies investigating the role of inflammatory biomarkers in mild cognitive impairment [26].

Most cases of dementia seem to have a common vascular aetiology [31–33], and atherosclerosis may represent a chronic inflammatory process [34]. On the other hand, inflammation increases cardiovascular risk [3, 4], which in turn is associated with cognition. Interestingly, in the present study, adjustment for several vascular factors only slightly modified the association between hs-CRP and cognition, indicating that the association between high hs-CRP and poor memory is not totally mediated through vascular factors. Thus, inflammatory markers such as hs-CRP may provide an additional method for global assessment of cardiovascular risk and cognitive impairment. Early diagnosis and treatment of vascular disorders could modulate the onset of dementia. Further, lifestyle factors such as physical activity [35] and body weight control [6] may prevent cognitive impairment, possibly at least partly due to their anti-inflammatory effects [34, 36, 37].

The strength of the present study is the population-based sample of elderly women followed up for a total of 12 years. We used valid and standardised measures of serum hs-CRP concentrations [3] and cognitive function, performed in the research laboratory. Further, we had information concerning several potential confounding factors. However, because only MMSE of measures of cognitive function was available at baseline, we were not able to study the associations of hs-CRP with changes in memory or cognitive speed. Thus, we cannot exclude the possibility that some of the participants had a mild memory impairment already at baseline. However, global cognitive function at baseline was good among the women. It is known that people with cognitive decline are less likely to participate in clinical studies [38]. Because the study sample only included one 10-year age group and the participants were younger and more educated and had better vascular risk factor profile and better baseline global cognitive function than the non-participants, our results may underestimate the true association between hs-CRP and poor cognition.

The present study suggests that high serum hs-CRP concentration is a useful predictor for memory impairment, and may help in identifying persons at risk for cognitive decline. Suppression of inflammation in individuals with elevated hs-CRP levels may be a way to prevent or delay cognitive impairment. The associations of hs-CRP and other inflammatory markers with specific cognitive domains deserve further investigation in large prospective population studies.
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