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# Interleukin-6 is associated with cognitive function: the Northern Manhattan Study

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## Abstract

**Background and purpose—**Inflammation has been linked to cognitive decline and dementia but the mechanism is not clear and few studies have included Hispanic and black subjects that may be at increased risk of these disorders.

**Methods—** We performed a cross-sectional analysis of the association between inflammatory marker levels and cognition in the stroke-free population-based cohort of the Northern Manhattan Study. Mini Mental State Exam (MMSE) scores were the continuous outcome and we adjusted for sociodemographic and vascular risk factors as well as subclinical atherosclerosis.

**Results—**Of the inflammatory markers, only interleukin (IL)-6 levels were associated with the MMSE. In univariate analysis age, hypertension, diabetes, smoking, moderate alcohol use, total homocysteine, carotid intima media thickness, and body mass index were positively associated with IL-6 levels. Hispanics compared to whites, those with less than a high school education, hypertension, cardiac disease, and total homocysteine were associated with lower MMSE scores. In a multivariate linear regression model, IL-6 was negatively associated with MMSE score adjusting for sociodemographic and vascular risk factors.

**Conclusions—**IL-6 levels were negatively associated with performance on the MMSE in this multiethnic cohort. Adjusting for vascular disease and subclinical atherosclerosis did not attenuate the association, suggesting a direct effect on the brain.

Cytokine dysregulation has been associated with cognitive disorders and dementia but the relative importance of vascular disease, neurodegeneration, aging, and genetic predisposition require further study.[1] Particular cytokines and their receptors have been linked to cognitive decline and dementia in population-based studies. In the Established Populations for the Epidemiologic Study of the Elderly, participants with interleukin (IL)-6 levels in the upper tertile had higher rates of cognitive decline after 2.5 and seven years of follow up. In the Honolulu-Asia Aging Study, high C-reactive protein (CRP) levels were predictive of dementia after 25 years.[2, 3] Other pro-inflammatory cytokines, such as tumor necrosis factor alpha (TNF- α), IL-1 beta, and IL-2, as well as $\alpha_1$-antichymotrypsin have been associated with aging, vascular disease, and Alzheimer disease but few studies have been population-based.[4–7] The Health ABC study found that the effects of inflammation on cognitive decline were modified by vascular risk factors. Subjects with CRP and/or IL-6 levels in the upper tertile were at greater

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risk of cognitive decline compared to those in the lowest only if they also had the metabolic syndrome.[8] If inflammation causes cognitive disorders through a vascular mechanism, it remains unclear whether large vessel atherosclerosis, small vessel disease, or both are involved. We have previously found associations between tumor necrosis factor alpha receptor (TNFR) levels and carotid atherosclerosis. Also, an IL-6 gene polymorphism is positively associated with carotid intima media thickness (IMT) in our cohort.[9] To our knowledge, no studies of inflammation and cognition have examined subclinical measures of atherosclerosis as a potential modifier. However, such surrogate measures are useful to help avoid residual confounding when considering vascular risk factors.

Most studies of the effects of inflammation on cognition have been limited to older cohorts made up of groups from different geographic locations and none have included Hispanics. Hispanics are the fastest growing segment of the US population, having increased by 58% in the 1990s and they may be at greater risk of dementia than whites as well as having a higher prevalence of vascular risk factors and stroke, with important public health implications.[10, 11] A follow-up by the Health ABC study found that black as well as white subjects with the metabolic syndrome and higher IL-6 and CRP levels developed cognitive impairment and decline more readily than those with the metabolic syndrome and low markers of inflammation.[8] We have previously found an independent association between elevated total homocysteine (tHcy) and lower cognitive performance as well as incident stroke in our cohort of Hispanic, black, and white subjects living in the same community.[12] We performed a cross-sectional study to investigate the relationship between inflammatory marker levels (including IL-1, IL-2, IL-2R, IL-6, TNF-α, TNFR-1, and TNFR-2) and cognition in a multi-ethnic sample from our cohort adjusting for carotid intima media thickness as well as important sociodemographic and vascular factors.

## METHODS

### Subjects

The Northern Manhattan Study (NOMAS) includes a community-based prospective cohort of 3,298 stroke-free Hispanic, black, and white subjects enrolled between 1993 and 2001 using random digit dialing. Data were collected using standardized instruments and review of medical records. Race-ethnicity, hypertension, diabetes, and cardiac disease were defined as described previously.[12] Bilingual trained research assistants assessed cognition using the 30-item mini-mental state exam (MMSE) in English or Spanish, depending on the language spoken by the subject at home.[13] Carotid IMT was assessed by high-resolution B-mode carotid ultrasound (Diasonics 2D-Gateway, 7.5-MHz probe) according to the standardized scanning and reading protocols as previously described.[9] All subjects signed informed consent and the Columbia University Medical Center Institutional Review Board approved the study.

### Laboratory Analyses

Serum samples for inflammatory markers were drawn into EDTA tubes, spun immediately at 3000g at 4ºC for 20 minutes, and then frozen at −70ºC for later analysis. Inflammatory marker levels were then measured in batched samples with the use of enzyme-linked immunosorbent assay utilizing monoclonal antibodies to IL-1, IL-2, IL-6, IL-2 receptor, TNFR-1 and TNFR-2 levels (Biosource International). Fasting tHcy samples were measured at the University of Colorado as previously described.[12]

### Statistical Analyses

Inflammatory marker levels were available in a subsample of our stroke-free cohort (sample size depended on marker, see table 1). We examined inflammatory marker levels as independent variables in relation to MMSE score as the dependent variable. The MMSE was

log transformed to create a normal distribution. We performed univariate analyses to assess the effect of potential confounders, including age, sex, race-ethnicity, education, hypertension, cardiovascular disease, current smoking status, tHcy, diabetes mellitus, body mass index (BMI), and moderate alcohol consumption. Those variables associated with the MMSE at $p < 0.1$ were included in multivariate linear regression models. The final significance level was $p < 0.05$ (two-sided). We also examined tertiles of inflammatory marker levels in relation to the MMSE. Analyses were performed using SAS software (version 8.2, Carey, NC).

## RESULTS

Cytokines IL-1, IL-2, IL-6, and cytokine receptors IL-2R, TNFR-1, TNFR-2, and CRP levels were available in a group of stroke-free participants. The same group of participants had cytokines processed, although not all cytokines were obtained for a given individual (see table 1). The prevalence of sociodemographic variables and vascular risk factors were similar to the overall cohort (mean age 66.8) but there were fewer blacks (15% vs. 20%).

Interleukin 6 levels ranged from 0.04 to 8.38 pg/mL in the sample (mean 1.4, SD 1.2; sensitivity <104 fg/mL; Inter-assay CV: <6.72%, Intra-assay CV: <6.17%). Performance on the MMSE was inversely associated with IL-6 levels ($p=0.02$) but there was no association between other cytokine or cytokine receptor levels and the MMSE (table 1). Lower performance on the MMSE (median MMSE = 27 overall, interquartile range 24-28) was seen for Hispanic subjects compared to whites and for those with less than a high school education. Hypertension, a history of cardiac disease, and total homocysteine levels were inversely associated with the MMSE. Inerleukin-6 levels were positively associated with age, hypertension, diabetes mellitus, current smoking, carotid IMT, tHcy, and BMI. Those with a history of moderate alcohol consumption had lower IL-6 levels than non-drinkers or those that drank intermediate and heavy amounts.

In a multivariate linear regression model, each unit increase in IL-6 was associated with lower performance on the MMSE ($p=0.003$) adjusting for age, gender, race-ethnicity, education, hypertension, diabetes, cardiac disease, tHcy, current smoking status, alcohol consumption, and carotid IMT. Hispanics compared to whites ($p=0.02$) and those with less than a high school education ($p<0.0001$) had lower MMSE scores in the adjusted model. A history of cardiac disease was also independently associated with lower MMSE scores ($p=0.006$). Carotid IMT was not associated with cognition in this sample.

We confirmed our findings using tertiles of IL-6 (upper tertile 2.9 pg/mL, middle tertile 1.3 pg/mL, lowest 0.7 pg/mL); those in the highest tertile were significantly more likely to have lower MMSE scores compared to those in the lowest tertile in the adjusted model ($p=0.004$).

## DISCUSSION

Relative elevation of serum IL-6 levels was associated with a global measure of cognitive function in this multiethnic cohort, adjusting for relevant sociodemographic and vascular factors. Cross-sectional data do not allow determination of causality, but these findings support the concept that inflammation may have deleterious effects on cognition. The mechanism is poorly understood but may include interaction with vascular and/or neurodegenerative processes. For example, higher levels of IL-6 have been found in the spinal fluid of AD patients.[14] Data supporting the idea that peripheral IL-6 levels represent those in brain is lacking, but evidence suggests that IL-6 does cross the blood brain barrier and peripheral levels may represent spillover from the central nervous system and be a marker of inflammation.[15, 16]

Interleukin 6 may also affect cognition through an interaction with vascular disease as there is evidence that it increases the risk of diabetes mellitus and cardiovascular events.[17, 18] In addition, the finding that IL-6 and CRP levels above the median have predicted cognitive

decline in those with, but not without, the metabolic syndrome implicates a vascular mechanism.[8] Also, CRP levels have predicted dementia 25 years later and this was most dramatic for those with vascular dementia.[2] In the current study adjusting for a history of vascular risk factors had little effect on the association between IL-6 level and cognitive function. Thus, IL-6 may be an independent vascular risk factor or may be directly neurotoxic. We found a positive association between IL-6 and carotid IMT, an established measure of subclinical atherosclerosis known to predict myocardial infarction and stroke.[19] The Atherosclerosis Risk in Communities study found an association between carotid IMT and cognitive dysfunction.[20] However, we were not able to confirm such a relationship despite finding a history of cardiac disease to be independently associated with lower MMSE scores in this stroke-free sample. It may be that IL-6 affects cognition through a vascular mechanism such as small vessel damage that is independent of large vessel atherosclerosis. Future studies in a larger sample from our cohort may help answer this question.

Our finding of lower cognitive function in Hispanic compared to white subjects adjusting for age, education, and other factors may be due to differences in subclinical cerebrovascular disease such as silent infarcts, but was not mediated by IL-6 levels in this sample. Hispanics are a rapidly growing segment of the U.S. population and, together with blacks, may be at elevated risk of small vessel disease as well as cognitive disorders.[10, 21] In our cohort, Hispanics had twice the incidence of stroke compared to whites living in the same community and another study has shown a higher risk of dementia for Hispanics in Northern Manhattan as well.[10, 22] Factors such as literacy and activity level have been shown to affect cognitive reserve and may explain some of the variance in performance on the MMSE. Data with measures of literacy, activity level, and more sensitive measures of cognitive function are needed to clarify race-ethnic differences in cognitive function in our cohort.[23, 24]

Limitations of this study include a relatively small sample size. Also, use of the MMSE as our measure of cognition may have limited our ability to detect mild cognitive changes such as problems with executive control function since it lacks sensitivity. This may have been true especially in the more highly educated; about 25% of subjects with more than a high school education achieved the maximum score (30 points) whereas only 1% of those with less than an eighth grade education did so. Evidence suggests low education may increase the risk of cognitive disorders and vascular disease and it remained an independent predictor of cognitive function in our adjusted model.[25] There was no interaction between low education and IL-6 levels as correlates of MMSE score, but more sensitive measures of cognitive function would provide information about the effect of cytokine dysregulation in the more highly educated.

Our findings are cross-sectional but support the concept that IL-6 can be related to cognitive dysfunction independent of other vascular risk factors or the presence of subclinical atherosclerosis. However, evidence suggests vascular damage and neurodegenerative processes may both be operative. Brain imaging studies involving measures of brain atrophy, subclinical infarction, and white matter hyperintensities would be useful in understanding this further. A subsample of our cohort is currently undergoing brain MRI and neuropsychological testing; further studies in this group may help clarify the role of cytokine dysregulation in cognition and the relative importance of vascular disease.

## References

1. Wilson CJ, Finch CE, Cohen HJ. Cytokines and cognition--the case for a head-to-toe inflammatory paradigm. Journal of the American Geriatrics Society 2002;50:2041–2056. [PubMed: 12473019]
2. Schmidt R, Schmidt H, Curb JD, Masaki K, White LR, Launer LJ. Early inflammation and dementia: A 25-year follow-up of the honolulu-asia aging study. Annals of Neurology 2002;52:168–174. [PubMed: 12210786]

3. Weaver JD, Huang MH, Albert M, Harris T, Rowe JW, Seeman TE. Interleukin-6 and risk of cognitive decline: Macarthur studies of successful aging. Neurology 2002;59:371–378. [PubMed: 12177370]
4. Bruunsgaard H, Andersen-Ranberg K, Jeune B, Pedersen AN, Skinhoj P, Pedersen BK. A high plasma concentration of tnf-alpha is associated with dementia in centenarians. *Journals of Gerontology Series A Biological Sciences & Medical Sciences* 1999;54
5. van Exel E, de Craen AJM, Remarque EJ, Gussekloo J, Houx P, Bootsma-van der Wiel A, Frolich M, Macfarlane PW, Blauw GJ, Westendorp RGJ. Interaction of atherosclerosis and inflammation in elderly subjects with poor cognitive function. Neurology 2003;61:1695–1701. [PubMed: 14694032]
6. Holmes C, El-Okl M, Williams AL, Cunningham C, Wilcockson D, Perry VH. Systemic infection, interleukin 1{beta}, and cognitive decline in alzheimer's disease. J Neurol Neurosurg Psychiatry 2003;74:788–789. [PubMed: 12754353]
7. Dik MG, Jonker C, Hack CE, Smit JH, Comijs HC, Eikelenboom P. Serum inflammatory proteins and cognitive decline in older persons. Neurology 2005;64:1371–1377. [PubMed: 15851726]
8. Yaffe K, Kanaya A, Lindquist K, Simonsick EM, Harris T, Shorr RI, Tylavsky FA, Newman AB. The metabolic syndrome, inflammation, and risk of cognitive decline. JAMA 2004;292:2237–2242. [PubMed: 15536110]
9. Rundek T, Elkind MS, Pittman J, Boden-Albala B, Martin S, Humphries SE, Juo SH, Sacco RL. Carotid intima-media thickness is associated with allelic variants of stromelysin-1, interleukin-6, and hepatic lipase genes: The northern manhattan prospective cohort study. Stroke 2002;33:1420–1423. [PubMed: 11988625]
10. Gurland BJ, Wilder DE, Lantigua R, Stern Y, Chen J, Killeffer EH, Mayeux R. Rates of dementia in three ethnoracial groups. International Journal of Geriatric Psychiatry 1999;14:481–493. [PubMed: 10398359]
11. Sacco RL, Kargman DE, Gu Q, Zamanillo MC. Race-ethnicity and determinants of intracranial atherosclerotic cerebral infarction. The northern manhattan stroke study. Stroke 1995;26:14–20. [PubMed: 7839388]
12. Wright CB, Lee HS, Paik MC, Stabler SP, Allen RH, Sacco RL. Total homocysteine and cognition in a tri-ethnic cohort: The northern manhattan study. Neurology 2004;63:254–260. [PubMed: 15277617]
13. Folstein MF, Folstein SE, McHugh PR. "mini-mental state". A practical method for grading the cognitive state of patients for the clinician". Journal of Psychiatric Research 1975;12:189–198. [PubMed: 1202204]
14. Singh VK, Guthikonda P. Circulating cytokines in alzheimer's disease. Journal of Psychiatric Research 1997;31:657–660. [PubMed: 9447570]
15. Banks WA, Kastin AJ, Broadwell RD. Passage of cytokines across the blood-brain barrier. Neuroimmunomodulation 1995;2:241–248. [PubMed: 8963753]
16. Licastro F, Pedrini S, Caputo L, Annoni G, Davis LJ, Ferri C, Casadei V, Grimaldi LM. Increased plasma levels of interleukin-1, interleukin-6 and alpha-1-antichymotrypsin in patients with alzheimer's disease: Peripheral inflammation or signals from the brain? Journal of Neuroimmunology 2000;103:97–102. [PubMed: 10674995]
17. Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus.[see comment]. JAMA 2001;286:327–334. [PubMed: 11466099]
18. Ridker PM, Hennekens CH, Buring JE, Rifai N. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. New England Journal of Medicine 2000;342:836–843. [PubMed: 10733371]
19. O'Leary DH, Polak JF, Kronmal RA, Manolio TA, Burke GL, Wolfson SK. The Cardiovascular Health Study Collaborative Research Group. Carotid-artery intima and media thickness as a risk factor for myocardial infarction and stroke in older adults. N Engl J Med 1999;340:14–22. [PubMed: 9878640]
20. Cerhan JR, Folsom AR, Mortimer JA, Shahar E, Knopman DS, McGovern PG, Hays MA, Crum LD, Heiss G. Correlates of cognitive function in middle-aged adults. Atherosclerosis risk in communities (aric) study investigators. Gerontology 1998;44:95–105. [PubMed: 9523221]

21. Sacco RL, Boden-Albala B, Abel G, Lin IF, Elkind M, Hauser WA, Paik MC, Shea S. Race-ethnic disparities in the impact of stroke risk factors: The northern manhattan stroke study. Stroke 2001;32:1725–1731. [PubMed: 11486097]

22. Sacco RL, Boden-Albala B, Gan R, Chen X, Kargman DE, Shea S, Paik MC, Hauser WA. Stroke incidence among white, black, and hispanic residents of an urban community: The northern manhattan stroke study. American Journal of Epidemiology 1998;147:259–268. [PubMed: 9482500]

23. Manly JJ, Touradji P, Tang MX, Stern Y. Literacy and memory decline among ethnically diverse elders. Journal of Clinical & Experimental Neuropsychology 2003;25:680–690. [PubMed: 12815505]

24. Scarmeas N, Levy G, Tang MX, Manly J, Stern Y. Influence of leisure activity on the incidence of alzheimer's disease. Neurology 2001;57:2236–2242. [PubMed: 11756603]

25. Berkman LF. The association between educational attainment and mental status examinations: Of etiologic significance for senile dementias or not? Journal of Chronic Diseases 1986;39:171–175. [PubMed: 3949941]

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## TABLE 1

Associations between mean inflammatory marker levels and MMSE score.

| Inflammatory Marker | N | MMSE Regression coefficient |
|---|---|---|
| **IL-1** pg/mL | 152 | −0.01 |
| **IL-2** U/mL | 164 | −0.003 |
| **IL-2R** pg/mL | 251 | 0.001 |
| **IL-6** pg/mL | 269 | −0.03† |
| **TNF-**α pg/mL | 185 | 0.0004 |
| **TNF-**α **receptor 1** ng/mL | 268 | −0.02 |
| **TNF-**α **receptor 2** ng/mL | 268 | −0.02 |
| **CRP** mg/mL | 220 | −0.003 |

*All IL levels processed were in the same sample.

†P <0.1

MMSE=Mini Mental State Exam

IL=interleukin

TNF=tumor necrosis factor

CRP=C-reactive protein

## TABLE 2

Prevalence of sociodemographic and vascular factors in relation to Interleukin-6 levels and MMSE scores.

| | Mean (SD) | Prevalence N (%) | Interleukin-6 Mean (SD)* or regression coefficient (SE)† | MMSE Mean (SD)* or regression coefficient (SE)† |
|---|---|---|---|---|
| Age | 66.7 (8.4) | -- | 0.01 (0.01)* | −0.002 (0.001)* |
| Sex | Women | 145 (54) | 1.1 (1.9) | 25.2 (1.2)* |
| | Men | 124 (46) | 1.0 (2.2) | 26.1 (1.1) |
| Race-ethnicity | Hispanic | 170 (65) | 1.3 (1.2) | 25.1 (1.2)* |
| | Black | 44 (17) | 1.6 (1.5) | 26.1 (1.1) |
| | White (ref) | 48 (18) | 1.3 (1.4) | 27.4 (1.1) |
| Education | <HS | 146 (54) | 1.1 (2.0) | 24.9 (1.8)* |
| | ≥HS | 123 (46) | 1.0 (2.1) | 27.8 (1.1) |
| Hypertension | Yes | 193 (72) | 1.1 (2.0)* | 25.4 (1.2)* |
| | No | 76 (28) | 0.9 (2.1) | 26.3 (1.1) |
| Diabetes | Yes | 59 (22) | 1.3 (1.9)* | 25.6 (1.2) |
| | No | 210 (78) | 1.0 (2.1) | 25.7 (1.2) |
| Current smoker | Yes | 40 (15) | 1.3 (1.8)* | 25.2 (1.2) |
| | No | 229 (85) | 1.0 (2.1) | 25.7 (1.2) |
| Cardiac disease | Yes | 61 (23) | 1.1 (2.0) | 24.8 (1.2)* |
| | No | 208 (77) | 1.0 (2.0) | 25.9 (1.1) |
| Moderate alcohol use | Yes | 84 (31) | 0.9 (0.8)* | 26.2 (1.2) |
| | No | 183 (69) | 1.2 (0.7) | 24.9 (1.2) |
| tHcy umol/L | 9.2 (3.4) | -- | 0.42 (0.14)† | −0.05 (0.03) † |
| BMI | 28 (5) | -- | 0.03 (0.01)† | −0.002 (0.002) |
| Carotid IMT | 0.86 (0.15) | -- | 0.02 (0.01)† | 0.02 (0.07) |

*P<0.1

tHcy = total homocysteine, regression coefficient (SE)

MMSE = Mini Mental State Examination

BMI = Body mass index

Moderate alcohol use = >1 drink/month to 2 drinks/day