Hemoglobin Levels and Alzheimer Disease

An Epidemiologic Study in India

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Objective: *Anemia is common in developing countries, where populations are aging rapidly. The authors explored the cross-sectional relationship between hemoglobin concentration and Alzheimer disease (AD) in a rural elderly sample in Ballabgarh, India.* **Methods:** *A clinical diagnostic evaluation for dementia and a hemoglobin estimation were performed in 605 persons selected by screening a larger community-based sample age 55+ years. Twenty-six participants met criteria for AD.* **Results:** *Hemoglobin was inversely associated with AD after adjustment for age, sex, and literacy.* **Conclusion:** *Low hemoglobin is associated with AD and should be investigated further as a modifiable risk factor.* (Am J Geriatr Psychiatry 2004; 12:523–526)

The public health importance of Alzheimer disease (AD) in developed countries is undisputed. However, developing countries like India have the world's fastest-growing elderly populations. Thus, although prevalence of dementia in India is low, ranging from 0.8% to 3.5%, it represents a large number of affected individuals.[1] Identification of potential risk factors both sheds light on disease mechanisms and suggests preventive strategies. Different risk factors may operate in developed and developing nations, or a common risk factor may be more readily detected in certain populations than in others.

A potential risk factor of particular relevance to developing countries is anemia. Its estimated prevalence, based on blood hemoglobin concentration, is 45.2% among older adults in developing countries.[2] The few previous studies examining the association of anemia with AD were conducted in developed countries.[3–6] We tested the hypothesis that hemoglobin concentration was associated with AD in a rural, elderly, community-based sample in India.

METHODS

We performed our study in the rural Indian community of Ballabgarh from 1991 to 1999. We have previously described in detail the sampling and recruitment of the cohort of 5,126 Hindi-speaking, mostly illiterate, participants age 55+ years in this 28-village district.[1] Informed consent was obtained from all participants according to protocols approved by the Human Volunteers Protection Committee and the Institutional Review Board.

We identified dementia by a two-stage assessment. First, all 5,126 participants completed a standardized cognitive screening battery, while their next-of-kin answered a functional-impairment questionnaire. All participants were requested to provide finger-stick blood specimens for hemoglobin (Hb) estimation. On the basis of the screening, we selected participants for clinical evaluation if they were cognitively impaired, functionally impaired, or unable to complete cognitive testing, as reported previously.[1]

For the second stage, a clinical evaluation, we invited 536 "impaired" subjects who met the screening criteria and a randomly selected comparison group of 270 unimpaired subjects who did not meet the criteria. Of these 806 subjects, 659 completed the clinical diagnostic protocol, consisting of history taken and examination performed by an experienced neurolo-

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gist and a medical officer blind to the screening data. Consenting participants who met diagnostic criteria for dementia (described below) provided venous blood samples for chemistry screening and underwent brain MRI scans. The neurologist and medical officer interviewed the families of the 147 participants who were not directly examined and confirmed by history that these participants had no cognitive limitations; most of them refused examination because they were too busy, for example, out working in the fields. Fifty-four subjects who did not provide blood specimens for Hb estimation are excluded from the current analyses. We report data from 605 participants (340 women, 265 men) who completed both clinical diagnostic evaluation and Hb measurement.

We diagnosed dementia according to the *Diagnostic and Statistical Manual of Mental Disorders, 3rd Edition, Revised* (DSM-III-R) criteria.[7] We rated the stage (severity) of dementia by use of the Clinical Dementia Rating (CDR) scale,[8] on which participants rated ≥0.5 were classified as having dementia for the current analyses. We then diagnosed AD (Probable and Possible) according to the National Institute of Neurological and Communicative Disease and Stroke-Alzheimer's Disease and Related Disorders Association (NINCDS-ADRDA) Work Group clinical criteria.[9]

We measured Hb levels by the standard cyanmethemoglobin method. Because Hb norms and criteria for anemia are different for men and women (men: <13 g/dl; women: <12 g/dl),[2] we report our results for the overall sample and also separately by sex.

We used the Pearson chi-square test to test differences between groups on categorical data and the Mann-Whitney *U* test or Student's *t*-test, as appropriate, to test differences between groups on continuous variables. We fit multiple logistic-regression models to calculate odds ratios (ORs) with 95% confidence intervals (CIs) as estimates of the relative risk (RR) between Hb level (continuous variable) and AD, and all dementia, after adjusting for age, sex, and literacy, and including a sex × Hb interaction term. We also fit separate models in men and women, adjusting for age and literacy. We then fit similar models categorizing Hb levels according to the World Health Organization (WHO) criteria for anemia.[2] All models had adequate goodness of fit.

In post-hoc analyses limited to individuals with AD, we compared mean Hb levels between those with more and less severe dementia (CDR >1 versus CDR 0.5 or 1) to determine whether Hb was a function of dementia severity. We explored associations between Hb level and 1) serum albumin level (as a measure of protein malnutrition); and 2) serum creatinine (as a measure of renal function), using Spearman correlations, as these variables were not normally distributed.

RESULTS

The study sample (N = 605) had a mean (standard deviation [SD]) age of 69.6 (8.1) years; 55.4% were women, and 88.4% were illiterate. They were similar in literacy and sex distributions to, but significantly younger than, the 54 subjects who did not provide Hb samples and had a mean age of 73.4 (9.8) years ($t_{[657]} = 3.13$; $p = 0.0032$). Mean Hb level was significantly higher in men than women (13.1 [1.5] versus 11.8 [1.1] g/dl; $t_{[603]} = 13.4$; $p < 0.0001$).

Few participants reported comorbidities potentially relevant to anemia; 6.3% reported a history of chronic bronchitis; 8.1% reported gastrointestinal disease, including history of passing worms; 5.6% reported renal disease; and 0.3% reported cancer. History of surgery was reported by 37.7%, but these were predominantly for cataract extraction.

Dementia, as described earlier, was diagnosed in 32 participants (20 men, 12 women). Of these, 26 (15 men, 11 women) met criteria for AD. The frequency of AD was not significantly different between men (5.7%) and women (3.2%) within the clinically evaluated sample ($\chi^2_{[1]} = 2.3$; $p = 0.13$). Participants with AD had similar literacy and sex distributions to those without dementia but, at mean age 75.5 (10.5) years, were significantly older than those without dementia (mean age 67.2 [8.2]; $U = 11{,}353$; $z = 3.99$; $p < 0.0001$). Mean Hb was significantly lower among individuals with AD (11.5 [2.1] g/dl) than those without dementia (12.4 [1.4] g/dl; $t_{[597]} = 3.14$; $p = 0.0018$).

Unadjusted, as well as age-, sex-, and literacy-adjusted, ORs and 95% CIs were calculated for the associations of Hb (continuous) with AD. A significant inverse association between Hb and AD was seen in the overall sample (OR: 0.7; 95% CI: 0.6–0.9; Wald $\chi^2_{[1]} = 7.1$; $p = 0.0078$) and among men, but not among women (Table 1). However, the Hb × sex interaction

term was not statistically significant (Wald $\chi^2_{[1]} = 0.13$; $p = 0.72$), implying similar slopes for men and women.

When these analyses were repeated for "all dementias" (i.e., including six non-AD dementias), results were identical, possibly because of the small additional number of cases.

Those with more severe dementia (CDR >1) and less severe dementia (CDR 0.5 or 1) had mean Hb levels of 11.7 (1.5) g/dl and 11.3 (2.6) g/dl, respectively, which were not significantly different ($t_{[24]} = 0.55$; $p = 0.58$), although results were based on small numbers. In the 17 individuals with AD from whom we were able to obtain venous blood samples, post-hoc analyses showed no correlation between Hb level and serum albumin level (Spearman $r = 0.30$; $p = 0.24$) or serum creatinine (Spearman $r = -0.05$; $p = 0.84$).

The WHO categorical definition[2] classified 41.2% of our sample (30.0% of men, 49.9% of women) as anemic. Anemia thus defined was not significantly associated with AD (OR: 1.8; 95% CI: 0.8–4.1; Wald $\chi^2_{[1]} = 1.9$; $p = 0.17$) or all dementias (OR: 1.5; 95% CI: 0.7–3.2; Wald $\chi^2_{[1]} = 1.3$; $p = 0.26$) after adjusting for age, sex, and literacy, although the parameter estimates for anemia were in the same direction as those for Hb concentration.

DISCUSSION

We found an inverse relationship between Hb and AD, with every unit-1.0 g/dl increase in Hb concentration decreasing the probability of AD by 30%. Since Hb norms vary by sex,[2] men and women were examined separately; the inverse relationship between Hb and AD was statistically significant only in men, although an interaction term between Hb and sex was not statistically significant. We had relatively few cases of dementia overall, despite our large original cohort; we have previously discussed possible reasons for the low prevalence of dementia in our cohort and in developing countries, generally.[1] The low number of cases may also have been responsible for the absence of findings related to dementia severity and to non-AD dementias.

Because our epidemiologic study was nested within a large, representative community sample, its results are more generalizable to the population than earlier case-control studies. Previous studies, all performed in developed countries, defined anemia categorically by fixed Hb or hematocrit values, rather than examining continuously distributed Hb, as we did. To compare our results with those studies, we reexamined the association using the WHO definition of anemia,[2] but the results were not statistically significant. An Australian case-control study reported a non-significant elevated probability of AD in individuals with self- or informant-reported history of anemia.[5] An American case-control study at the Mayo Clinic found a twofold increase in odds for AD in persons meeting WHO anemia criteria, significant only in women; at the same site, a retrospective cohort study of anemia based on medical records showed no overall increase in the risk of incident AD.[3] An Australian community study found a significant association of anemia with vascular dementia but not AD.[4] A Greek community study found higher prevalence of cognitive impairment in anemic than in non-anemic participants, significant only in men.[6] Our results may differ because we treated Hb as a continuous variable or because Hb was distributed differently in the Indian than in the American and Australian study samples. The 40.5% of non-demented individuals meeting WHO criteria for anemia in our sample is typical of what is found in devel-

TABLE 1. Association of Hemoglobin (Hb) Concentration With Alzheimer Disease (AD)

	Men (N = 260)		Women (N = 339)		All Participants (N = 599)	
	Unadjusted	Adjusted for Age and Literacy	Unadjusted	Adjusted for Age[a]	Unadjusted	Adjusted for Age, Sex, and Literacy
Alzheimer disease	0.6 (0.5 – 0.8) Wald $\chi^2_{[1]} = 12.4$; $p = 0.0004$	0.7 (0.5 – 0.9) Wald $\chi^2_{[1]} = 7.0$; $p = 0.0083$	0.7 (0.4 – 1.1) Wald $\chi^2_{[1]} = 2.6$; $p = 0.11$	0.8 (0.5 – 1.4) Wald $\chi^2_{[1]} = 0.5$; $p = 0.47$	0.7 (0.5–0.9) Wald $\chi^2_{[1]} = 9.5$; $p = 0.0021$	0.7 (0.6–0.9) Wald $\chi^2_{[1]} = 7.1$; $p = 0.0078$

Note: Values are odds ratios (95% confidence intervals) based on multiple logistic-regression models.
[a] Adjusted for age only; all women with AD were illiterate.

oping countries,[2] but substantially higher than the 17% found among Australian control subjects.[4]

Our cross-sectional results could imply that low Hb leads to AD, that AD leads to low Hb, or that a third factor leads to both. Potential disease mechanisms linking anemia and dementia include exacerbation of focal cerebral ischemia,[4] oxidative stress,[10] and hypoxic injury.[11] Alternatively, anemia might simply be a non-specific marker for chronic conditions that decrease brain reserve, lowering the threshold for manifestation of dementia. Anemia in older adults can result from various causes, including chronic inflammatory disorders, renal diseases, malignancies, blood loss from surgery, gastrointestinal bleeding, and metabolic disorders. The self-reported prevalence of these was low in our sample. Iron deficiency from dietary deficiencies and parasitic infestations is a major factor contributing to anemia in developing countries.[2,12] Most of our study participants were vegetarians, known to have lower Hb, ferritin, and cobalamin levels than non-vegetarians;[12] we were only able to measure Hb. In this traditional, rural Indian community, all individuals with AD lived with their families and were adequately cared for at home; it is unlikely that their health and nutrition were neglected. Mean Hb was not different between those with greater and lesser dementia severity, as might be expected if progressive dementia had led to low Hb. Hb was not correlated with albumin, as might be expected if it was related to general malnutrition, or with creatinine, as expected if it was linked with chronic renal disease. Thus, in our study sample, low Hb may have increased susceptibility to AD. The potential role of low Hb in increasing risk of AD should be explored prospectively, in both developed and developing countries, considering its high prevalence in older adults and the intriguing possibility that low Hb could be a modifiable risk factor.

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