HHS Public Access

Author manuscript

Am J Clin Nutr. Author manuscript; available in PMC 2017 February 03.

Published in final edited form as:

Am J Clin Nutr. 2013 October ; 98(4): 1042–1047. doi:10.3945/ajcn.113.059683.

Near-elimination of folate-deficiency anemia by mandatory folic acid fortification in older US adults: Reasons for Geographic and Racial Differences in Stroke study 2003–2007[2,,3]

Oluwaseun A Odewole, **Rebecca S Williamson**, **Neil A Zakai**, **Robert J Berry**, **Suzanne E Judd**, **Yan Ping Qi**, **Demilade A Adedinsewo**, and **Godfrey P Oakley Jr**

Department of Epidemiology (OAO, RSW, DAA, and GPO) and Center for Spina Bifida Research, Prevention and Policy (GPO), Rollins School of Public Health, Emory University, Atlanta, GA; the Departments of Medicine and Oncology, Vermont College of Medicine, University of Vermont, Burlington, VT (NAZ); the Division of Birth Defects and Developmental Disabilities, National Center on Birth Defects and Developmental Disabilities, CDC, Atlanta, GA (RJB and YPQ); and the Department of Biostatistics, School of Public Health, University of Alabama at Birmingham, Birmingham, AL (SEJ)

Abstract

Background—The United States implemented mandatory folic acid fortification of enriched cereal grains in 1998. Although several studies have documented the resulting decrease in anemia and folate deficiency, to our knowledge, no one has determined the prevalence of folate-deficiency anemia after fortification.

Objective—We determined the prevalence of folate deficiency and folate-deficiency anemia within a sample of the Reasons for Geographic and Racial Differences in Stroke (REGARDS) cohort.

Design—The REGARDS cohort is a prospective cohort of 30,239 black and white participants living in the contiguous United States. We measured serum folate concentrations in a random sample of 1546 REGARDS participants aged ≥50 y with baseline hemoglobin and red blood cell mean corpuscular volume measurements. Folate deficiency was defined as a serum folate concentration <6.6 nmol/L (<3.0 ng/mL), and anemia was defined as a hemoglobin concentration <13 g/dL in men and <12 g/dL in nonpregnant women (WHO criteria). Folate-deficiency anemia was defined as the presence of both folate deficiency and anemia.

Results—The mean hemoglobin concentration was 13.6 g/dL, and 15.9% of subjects had anemia. The median serum folate concentration was 34.2 nmol/L (15.1 ng/mL), and only 2 of

---

[2]Findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the CDC.

[3]Supported by the CDC (Cooperative Agreement U48 DP001909O) and by unrestricted gifts from Gerry Davy and Sophie's Voice Foundation.

Address reprint requests and correspondence to GP Oakley Jr, Department of Epidemiology, Rollins School of Public Health, Emory University, 1518 Clifton Road, Atlanta, GA 30322. gpoakley@mindsping.com.

The authors' responsibilities were as follows—OAO, RSW, NAZ, YPQ, and GPO: designed the research; SEJ and NAZ: provided essential materials; OAO: analyzed data and had primary responsibility for final content of the manuscript; OAO, RSW, DAA, NAZ, RJB, and GPO: wrote the manuscript; and all authors: read and approved the final manuscript. None of the authors had a conflict of interest.

1546 participants 0.1%) were folate deficient. Both subjects were African American women with markedly elevated C-reactive protein concentrations, macrocytosis, and normal serum cobalamin concentrations; only one subject was anemic. Overall, the prevalence of folate-deficiency anemia was <0.1% (1 of 1546 subjects).

Conclusion—Our data suggest that, after mandatory folic acid fortification, the prevalence of folate-deficiency anemia is nearly nonexistent in a community-dwelling population in the United States.

INTRODUCTION

The global prevalence of anemia in the elderly (aged >65 y) has been estimated to be 23.9%, resulting in ~164 million elderly people being anemic worldwide (1). Anemia estimates in the elderly US population have ranged from 9% to 11% depending on the population being studied (2, 3). Some of the common causes of anemia in the elderly include nutritional deficiencies (deficiencies of folate, vitamin B-12, and iron), chronic inflammation, and renal insufficiency (3). Before the implementation of mandatory folic acid fortification, nutritional anemia accounted for approximately one-third of all anemia cause in the elderly US population (3). Folate deficiency alone accounted for 6.4% of anemia, the combination of folate and cobalamin deficiency accounted for 2% of anemia, and the combined deficiencies of iron with folate or vitamin B-12 or both accounted for an additional 3% of anemia as shown in an analysis of the NHANES III phase 2, 1991–1994 (3). Clinically, anemia of folate deficiency manifests as megaloblastic anemia (4, 5).

In March 1996, the US Food and Drug Administration required mandatory fortification of enriched cereal grains with folic acid to be implemented by January 1998 (6). This requirement has resulted in Americans consuming an average of 138 μg folic acid/d from mandatory fortification (more than one-third of the recommended daily intake of 400 μg/d) (7). According to the estimates from the Flour Fortification Initiative, worldwide, 69 countries have policies requiring the fortification of wheat or maize flour with folic acid (8).

Mandatory folic acid fortification and the resulting decrease in the prevalence of neural tube defects (NTDs) were recently called one of the 10 greatest public health achievements of the decade by the CDC (9). Although the effects of folic acid fortification on NTD prevention have been well recognized (10–13), the additional benefits of fortification on the anemia of folate deficiency have not been well explored. Several studies from the United States and Canada after mandatory folic acid fortification have shown a decline in the prevalence of both folate deficiency and anemia in general (14–19); however, to our knowledge, the effect of folic acid fortification on the prevalence of folate-deficiency anemia has yet to be determined. This study aimed to determine the prevalence of folate deficiency and folate-deficiency anemia in a sample of the Reasons for Geographic and Racial Differences in Stroke (REGARDS) cohort after fortification.

SUBJECTS AND METHODS

Study population and participants

The REGARDS trial is a prospective, longitudinal study of 30,239 subjects aged ≥45 y and recruited between January 2003 and October 2007 with the goal to better understand geographic and racial differences in stroke incidence in the United States. Participants were selected from a commercial, nationwide list of >250 million individuals in the United States (Genesys Incorporated) (20, 21). Approximately 50% of the cohort was selected from the stroke belt (ie, North Carolina, South Carolina, Georgia, Tennessee, Alabama, Mississippi, Arkansas, and Louisiana), ~50% of the cohort was black, and 50% of the cohort was women (20, 21).

Participants were contacted via telephone, and one resident aged ≥45 y per household was randomly screened for eligibility; exclusion criteria included race other than black or white, active treatment of cancer, the presence of medical conditions that prevented long-term participation, cognitive impairment as judged by the telephone interviewer, residence in or being on a waiting list for a nursing home, and inability to communicate in English. Verbal informed consent and a medical history were also obtained (20, 21). The overall response rate (defined as the percentage of subjects agreeing to be interviewed among known eligible candidates contacted plus an adjustment for an estimate of likely eligible participants among unknown eligible participants) of the cohort was ~40% (22).

Physical measurements and a medication inventory, phlebotomy, and urine collection were carried out during an in-home visit at baseline. Participants also provided written informed consent and completed a self-administered questionnaire, including a food-frequency questionnaire. Participants were followed up and contacted by telephone every 6 mo about stroke symptoms, hospitalizations, and general health status (21).

From this larger cohort, we selected a random sample of 1547 REGARDS participants aged ≥50 y with a baseline complete blood count and measured their serum folate and cobalamin concentrations for our study. With the use of statistical software (SAS 9.3; SAS Institute Inc), this sampling scheme involved a simple random-assignment procedure of eligible subjects from the cohort. One participant had an insufficient stored sample for serum folate testing and was excluded from the analysis. Our final analytic sample size was 1546 participants.

This study was approved by both the Institutional Review Board of Emory University and the REGARDS study executive committee, and all research procedures were conducted in accordance to their ethical standards.

Measurements

A complete blood count was performed the day after sample collection by using an automated cell counting on a Beckman Coulter LH 755 Hematology Workcell analyzer (Beckman Coulter Inc); measurement details have been described elsewhere (21). Serum folate concentrations were measured on stored serum samples collected at baseline by using the Cobas Folate II assay, which is a binding assay for the in vitro quantitative determination

of folate in human serum intended for use on Elecsys and Cobas e immunoassay analyzers (Roche Diagnostics) in the laboratory for clinical biochemistry research at the University of Vermont. The concentration range of the folate assay was 1.4—45.3 nmol/L (0.6–20 ng/mL). Serum vitamin B-12 concentrations were measured retrospectively by using the Elecsys cobalamin assay (Roche Diagnostics) at the clinical laboratory. The range of the vitamin B-12 assay was 22–1476 pmol/L.

Outcome definitions

Folate deficiency was defined as serum folate concentrations <6.8 nmol/L (<3 ng/mL) (23). Anemia was defined by using both WHO criteria (hemoglobin concentration <12 g/dL for women and <13 g/dL for men) (23) and age-, sex-, and race-specific criteria defined by Beutler and Waalen (ie, Beutler's definition) (24). For the purpose of our analysis, folate-deficiency anemia was defined as the presence of both folate deficiency and anemia by using either anemia definition. We provide a descriptive overview of individual participants with folate deficiency.

Statistical analysis

The statistical software (SAS 9.3; SAS Institute Inc) was used to determine the descriptive statistics and the prevalence of anemia, folate deficiency, and folate-deficiency anemia. Chi-square and 2-sample *t* tests were used to determine differences in variables of interest across white and black races (level of significance was set at $P < 0.05$), and 95% CIs for prevalence estimates were calculated by using Poisson's approximation to the binomial distribution. For the purpose of our analysis, we assigned participants with serum folate concentrations >45.3nmol/L (>20 ng/mL) (the upper limit of quantification for the assay) a value of 45.3 nmol/L (20 ng/mL). For each individual with folate deficiency, we reported their observed demographic, clinical, and laboratory values.

RESULTS

The mean (±SD) age of 1546 participants was 65.4 ± 8.8 y; 54.1% of participants were white, and 60.0% of participants were women. A summary of the demographic characteristics of study participants is shown in Table 1.

The mean hemoglobin concentration of the entire sample was 13.6 g/dL. The lowest mean hemoglobin concentrations were shown in black women (12.7 g/dL), whereas the highest hemoglobin concentrations were shown in white men (14.6 g/dL) (Table 2). The prevalence of anemia in our sample by using the WHO criteria was 16.0% (95% CI: 14.1%, 18.1%); however, when anemia was classified by age, sex, and race cutoffs (Beutler's definition), the prevalence was 13.1% (95% CI: 11.4%, 15%) (17). The highest prevalence of anemia by using the WHO definition in our sample was in black women (25.3%), whereas the lowest was shown in white men (7.7%). When anemia was classified by using Beutler's definition (age, sex, and race cutoffs), the highest prevalence of anemia in our sample was shown in black men (20.0%), whereas the lowest prevalence of anemia in our sample was shown in white men (10.7%) (Table 2).

The median serum folate concentration was 35.6 nmol/L (15.1 ng/mL), and the prevalence of folate deficiency in our sample was 0.1% (95% CI: 0.03%, 0.5%), which was in 2 of 1546 study participants. We used the median measure because 18.2% of our sample had serum folate concentrations above the upper limit of the assay of 45.3 nmol/L (20 ng/mL).

With the use of the WHO definition of anemia in combination with folate deficiency, there was only one participant with folatedeficiency anemia (<0.1%; 95% CI: 0.01%, 0.5%). There were no cases of folate-deficiency anemia by using the combination of folate deficiency and anemia according to Beutler's definition (Table 2).

Characteristics of the 2 folate-deficient REGARDS participants are shown in Table 3. Both participants were black women, had normal serum vitamin B-12 concentrations (280.2 and 208.6 pmol/L), markedly elevated high-sensitivity C-reactive protein concentrations (43.8 and 17.4 mg/L), and macrocytosis (mean corpuscular volume of 113 and 99 fL). These participants had BMI (in kg/m^2) of 33.5 and 25.8, and both subjects had low daily caloric intakes (754 and 1056 kcal).

DISCUSSION

Although the contribution of mandatory flour fortification in North America to the reduction in NTDs has been documented (9, 12, 13, 25–27), not enough information has been published about its benefits in reducing folate-deficiency anemia in older adults. After mandatory folic acid fortification in the United States, there has been almost no folate deficiency and folate-deficiency anemia in an older population of adults in the REGARDS study. In our study, there were 2 individuals with folate deficiency, whereas only one individual had concurrent anemia.

The prevalence of folate deficiency was 0.1%, and prevalence of anemia associated with folate deficiency was <0.1%. Our folatedeficiency prevalence estimate was consistent with those in other population-based analyses of serum folate concentrations after fortification. For instance, in a recent study that used data from NHANES, Pfeiffer et al (19) showed that the postfortification prevalence of low serum folate, which was defined as <10 nmol/L (4.4 ng/mL), was 0.7% compared with 24% prefortification. Data from the Framingham Offspring Study cohort also showed a postfortification folate deficiency prevalence of 1.7% in nonsupplement users compared with 22% prefortification (14).

Our folate-deficiency anemia prevalence estimate was >100-fold less than prefortification estimates of 6.4% in the elderly US population (3, 28). In an analysis of NHANES III, Guralnik et al (3) showed that, in the prefortification period in adults aged ≥65 y who were anemic, 6.4–11.8% of the anemia could be attributed to folate deficiency. However, in subjects with anemia (by the WHO definition) in our REGARDS sample, only 0.4% of subjects had folate deficiency (1 of 247 participants). When we defined anemia by using Beutler's criteria, none of the participants with anemia had folate deficiency (0 of 202 participants). These results suggest that mandatory folic acid fortification has improved the health of the older Americans by nearly eliminating folate deficiency and folate-deficiency anemia.

Other countries that have instituted mandatory folic acid fortification have reported findings similar to those from the United States. For instance, in the Canadian Health Measures Survey, which is a population-based cohort, MacFarlane et al (29) and Colapinto et al (30) showed a <1% prevalence of folate deficiency postfortification. A hospital-laboratory–based study from Australia also showed a 77% decrease (from 9.3% to 2.1%) in the incidence of low serum folate concentrations after mandatory folic acid fortification compared with prefortification (31). Hospital-based studies have also documented low to nonexistent folate deficiency and folate-deficiency anemia after mandatory fortification so much so that questions have been raised about the utility of screening for folate deficiency in hospitalized patients with anemia (16, 32, 33).

One of the strengths of this study was that our sample was obtained from a large prospective cohort. Another strength was that clinical data we had of participants with folate deficiency were based on information collected by the REGARDS study. This collection enabled us to examine the possibility of other comorbidities such as cancer, diabetes, hypertension, and markers of chronic inflammation that may have contributed to anemia and possibly serum folate concentration in the 2 folate-deficient individuals.

Our study was not without limitations. Within the REGARDS cohort, only white and black races were represented. Although the REGARDS cohort was selected from a nationwide list of 250 million individuals in the United States, the cohort was over-sampled from the stroke belt region and excluded persons with selected medical conditions such as cognitive impairment, persons residing in or awaiting admission to a nursing home, as well as persons who do not speak English. Thus, the generalizability of our study to the entire US population may be limited. However; despite these limitations, our results were consistent with a recent analysis of a nationally representative sample of the US population in NHANES that showed that the prevalence of low serum folate was <1% irrespective of age, sex, or race (19). Also, because REGARDS data were only collected after mandatory folic acid fortification in the United States, we did not have prefortification estimates from the same population for comparison. Hence, we had to use prefortification estimates from other studies such as the NHANES. In addition, we defined folate-deficiency anemia as the concurrent appearance of low serum folate with anemia, and therefore, we could not be certain that the anemia was a result of folate deficiency. The participant who had both folate deficiency and anemia also had an elevated C-reactive protein concentration, and as such, chronic inflammation and anemia of chronic disease could not be ruled out as potential causes of anemia. The 2 participants in our sample with folate deficiency also had a previous history of stroke on entry to the study. In addition, they both had low caloric intake and, thus, poor diet or undernutrition may have played a role in the cause of their folate deficiency and folate-deficiency anemia. As a result, our estimate of the prevalence of folate-deficiency anemia after mandatory folic acid fortification as being <0.1% may have been an overestimate. Information on folate-only supplements was not available in our REGARDS dataset. Although 36% of our sample reported the consumption of multivitamin supplements, this amount was not sufficient to explain the overall low prevalence of folate deficiency in our population. Also, we could not be certain that all these multivitamin supplements contained folic acid.

In conclusion, both folate deficiency and anemia associated with folate deficiency may have been eliminated postfortification in an ambulatory and relatively healthy population. Although other factors such as increased supplement use and perhaps improved nutrition may have contributed to a decline in folatedeficiency anemia, reductions in both the prevalence of folate deficiency and folate-deficiency anemia are an added benefit of mandatory flour fortification. In addition to preventing spina bifida and anencephaly, mandatory folic acid fortification has contributed to the near elimination of folate deficiency and folatedeficiency anemia in individuals aged ≥50 y. Thus, regulators in countries around the world considering folic acid fortification need to take into account the benefits of mandatory folic acid fortification on both newborns in preventing NTDs and in older adults in preventing folate-deficiency anemia.

Acknowledgments

We thank Vijaya Kancherla of the Center for Spina Bifida Research, Prevention and Policy, Emory University, for her valuable comments and content editing.

REFERENCES

1. McLean E, Cogswell M, Egli I, Wojdyla D, de Benoist B. Worldwide prevalence of anaemia, WHO Vitamin and Mineral Nutrition Information System, 1993–2005. Public Health Nutr. 2009; 12:444–54. [PubMed: 18498676]
2. Zakai NA, Katz R, Hirsch C, Shlipak MG, Chaves PHM, Newman AB, Cushman M. A prospective study of anemia status, hemoglobin concentration, and mortality in an elderly cohort: the Cardiovascular Health Study. Arch Intern Med. 2005; 165:2214–20. [PubMed: 16246985]
3. Guralnik JM, Eisenstaedt RS, Ferrucci L, Klein HG, Woodman RC. Prevalence of anemia in persons 65 years and older in the United States: evidence for a high rate of unexplained anemia. Blood. 2004; 104:2263–8. [PubMed: 15238427]
4. Oakley GP Jr. Global prevention of all folic acid-preventable spina bifida and anencephaly by 2010. Community Genet. 2002; 5:70–7. [PubMed: 14960902]
5. Zittoun J. [Anemias due to disorder of folate, vitamin B12 and transcobalamin metabolism] Rev Prat. 1993; 43:1358–63. (in French). [PubMed: 8235383]
6. Food and Drug Administration. Food standards: amendment of standards of identity for enriched grain products to require addition of folic acid. Fed Regist. 1996; 61:8761–97.
7. Yang Q, Cogswell ME, Hamner HC, Carriquiry A, Bailey LB, Pfeiffer CM, Berry RJ. Folic acid source, usual intake, and folate and vitamin B-12 status in US adults: National Health and Nutrition Examination Survey (NHANES) 2003–2006. Am J Clin Nutr. 2010; 91:64–72. (Published erratum appears in Am J Clin Nutr 2010:92:1001.). [PubMed: 19828716]
8. Flour fortification initiative. Apr. 2013 Available from: http://www. ffinetwork.org/global_progress/index.php (cited 26 April 2013)
9. Centers for Disease Control and Prevention. Ten great public health achievements–United States, 2001–2010. MMWR Morb Mortal Wkly Rep. 2011; 60:619–23. [PubMed: 21597455]
10. Centers for Disease Control and Prevention (CDC). Spina bifida and anencephaly before and after folic acid mandate–United States, 1995–1996 and 1999–2000. MMWR Morb Mortal Wkly Rep. 2004; 53:362–5. [PubMed: 15129193]
11. Dietrich M, Brown CJP, Block G. The effect of folate fortification of cereal-grain products on blood folate status, dietary folate intake, and dietary folate sources among adult non-supplement users in the United States. J Am Coll Nutr. 2005; 24:266–74. [PubMed: 16093404]
12. Boulet SL, Yang Q, Mai C, Kirby RS, Collins JS, Robbins JM, Meyer R, Canfield MA, Mulinare J. Trends in the postfortification prevalence of spina bifida and anencephaly in the United States. Birth Defects Res A Clin Mol Teratol. 2008; 82:527–32. [PubMed: 18481813]

13. Centers for Disease Control and Prevention (CDC). Spina bifida and anencephaly before and after folic acid mandate–United States, 1995–1996 and 1999–2000. MMWR Morb Mortal Wkly Rep. 2004; 53:362–5. [PubMed: 15129193]
14. Jacques PF, Selhub J, Bostom AG, Wilson PW, Rosenberg IH. The effect of folic acid fortification on plasma folate and total homocysteine concentrations. N Engl J Med. 1999; 340:1449–54. (doi). [PubMed: 10320382]
15. Pfeiffer CM, Caudill SP, Gunter EW, Osterloh J, Sampson EJ. Biochemical indicators of B vitamin status in the US population after folic acid fortification: results from the National Health and Nutrition Examination Survey 1999–2000. Am J Clin Nutr. 2005; 82:442–50. [PubMed: 16087991]
16. Joelson DW, Fiebig EW, Wu AH. Diminished need for folate measurements among indigent populations in the post folic acid supplementation era. Arch Pathol Lab Med. 2007; 131:477–80. [PubMed: 17516752]
17. Lawrence JM, Petitti DB, Watkins M, Umekubo MA. Trends in serum folate after food fortification. Lancet. 1999; 354:915–6. [PubMed: 10489953]
18. Ganji V, Kafai MR. Hemoglobin and hematocrit values are higher and prevalence of anemia is lower in the post-folic acid fortification period than in the pre-folic acid fortification period in US adults. Am J Clin Nutr. 2009; 89:363–71. [PubMed: 19056553]
19. Pfeiffer CM, Hughes JP, Lacher DA, Bailey RL, Berry RJ, Zhang M, Yetley EA, Rader JI, Sempos CT, Johnson CL. Estimation of trends in serum and RBC folate in the U.S. population from pre- to post-fortification using assay-adjusted data from the NHANES 1988–2010. J Nutr. 2012; 142:886–93. [PubMed: 22437563]
20. Howard VJ, Cushman M, Pulley LV, Gomez CR, Go RC, Prineas RJ, Graham A, Moy CS, Howard G. The reasons for geographic and racial differences in stroke study: objectives and design. Neuroepidemiology. 2005; 25:135–43. [PubMed: 15990444]
21. Zakai NA, McClure LA, Prineas R, Howard G, McClellan W, Holmes CE, Newsome BB, Warnock DG, Audhya P, Cushman M. Correlates of anemia in american blacks and whites. Am J Epidemiol. 2009; 169:355–64. [PubMed: 19066309]
22. Howard VJ, McClure LA, Meschia JF, Pulley LV, Orr SC, Friday GH. High prevalence of stroke symptoms among persons without a diagnosis of stroke or transient ischemic attack in a general population: the REasons for Geographic And Racial Differences in Stroke (REGARDS) study. Arch Intern Med. 2006; 166:1952–8. [PubMed: 17030827]
23. WHO. Nutritional anaemias. Report of a WHO scientific group. World Health Organ Tech Rep Ser. 1968; 405:5–37. [PubMed: 4975372]
24. Beutler E, Waalen J. The definition of anemia: what is the lower limit of normal of the blood hemoglobin concentration? Blood. 2006; 107:1747–50. [PubMed: 16189263]
25. Williams LJ, Mai CT, Edmonds LD, Shaw GM, Kirby RS, Hobbs CA, Sever LE, Miller LA, Meaney FJ, Levitt M. Prevalence of spina bifida and anencephaly during the transition to mandatory folic acid fortification in the United States. Teratology. 2002; 66:33–9. [PubMed: 12115778]
26. Mathews, T. Trends in spina bifida and anencephalus in the United States, 1991–2005. National Center for Health Statistics; Hyattsville, MD: 2007.
27. Honein MA, Paulozzi LJ, Mathews T, Erickson JD, Wong LYC. Impact of folic acid fortification of the US food supply on the occurrence of neural tube defects. JAMA. 2001; 285:2981–6. [PubMed: 11410096]
28. Bross MH, Soch K, Smith-Knuppel T. Anemia in older persons. Am Fam Physician. 2010; 82:480–7. [PubMed: 20822082]
29. MacFarlane AJ, Greene-Finestone LS, Shi Y. Vitamin B-12 and homocysteine status in a folate-replete population: results from the Canadian Health Measures Survey. Am J Clin Nutr. 2011; 94:1079–87. [PubMed: 21900461]
30. Colapinto CK, O'Connor DL, Tremblay MS. Folate status of the population in the Canadian Health Measures Survey. CMAJ. 2011; 183:E100–6. [PubMed: 21149516]

31. Brown RD, Langshaw MR, Uhr EJ, Gibson JN, Joshua DE. The impact of mandatory fortification of flour with folic acid on the blood folate levels of an Australian population. Med J Aust. 2011; 194:65–7. [PubMed: 21241218]

32. Ashraf MJ, Cook J, Rothberg M. Clinical utility of folic acid testing for patients with anemia or dementia. J Gen Intern Med. 2008; 23:824–6. [PubMed: 18414954]

33. Shojania AM, von Kuster K. Ordering folate assays is no longer justified for investigation of anemias, in folic acid fortified countries. BMC Res Notes. 2010; 3:22. [PubMed: 20181007]

TABLE 1

Characteristics of study participants in the REGARDS study, 2003–2007[1]

Characteristics	All (*n* = 1546) summary
Demographics	
Age (y)	65.37 ± 8.8[2]
White [*n* (%)]	837 (54.1)
Female [*n* (%)]	928 (60.0)
Education [*n* (%)]	
College	550 (35.7)
Some college	419 (27.2)
High school	381 (24.7)
Less than high school	192 (12.5)
Income [*n* (%)]	
<$20,000	272 (17.6)
$20,000–$34,999	350 (22.6)
$35,000–$74,999	490 (34.7)
≥$75,000	229 (14.8)
Not reported	205 (13.3)
Multivitamin use [*n* (%)][3]	559 (36.1)
Comorbidity	
C-reactive protein[4]	2.17 (4.2)
Previous stroke [*n* (%)]	93 (6.0)
Previous transient ischemic attacks [*n* (%)]	59 (4.1)

[1] The REGARDS study had 30,239 participants; from this larger cohort, we selected a random sample of 1547 REGARDS participants aged ≥50 y with a baseline complete blood count and measured their serum folate and cobalamin concentrations for this analysis. One participant was excluded because of an insufficient stored sample for serum folate testing; the final analytic sample size was 1546. REGARDS, Reasons for Geographic and Racial Differences in Stroke.

[2] Mean ± SD.

[3] On the basis of a medication inventory.

[4] Median; IQR in parentheses. The median is reported because the variable C-reactive protein was not normally distributed. This variable was assessed by using statistical tests for normal distribution.

TABLE 2

Hematologic variables of study participants in the REGARDS study, 2003–2007[1]

Variable	All (*n* = 1546)	White (*n* = 837)		Black (*n* = 709)		*P*
		M (*n* = 363)	F (*n* = 474)	M (*n* = 255)	F (*n* = 454)	
Hemoglobin (g/dL)	13.6 ± 1.4[2]	14.6 ± 1.2	13.4 ± 1.2	13.8 ± 1.4	12.7 ± 1.2	<0.001
Anemia by using the WHO definition [*n* (%)]	247 (16.0)	28 (7.7)	42 (8.9)	62 (24.3)	115 (25.3)	<0.001
Anemia by using Beutler's criteria [*n* (%)][3]	202 (13.1)	39 (10.7)	52 (11.0)	51 (20.0)	60 (13.2)	0.005
Corpuscular volume (fL)	89.7 ± 6.0	91.9 ± 4.5	91.0 ± 4.9	88.7 ± 6.9	87.2 ± 6.6	<0.001
Macrocytosis [*n* (%)][4]	282 (18.2)	92 (25.3)	93 (18.6)	46 (18.0)	51 (11.2)	<0.001
Serum folate (nmol/L)[5]	34.2 (15.1)	35.6 (15.7)	37.8 (16.7)	29.9 (13.2)	13.9	<0.001
Folate deficiency [*n* (%)][6]	2 (0.1)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.4)	0.12
Serum vitamin B-12 (pmol/L)	661.7 (353.8)	559.5 (292.1)	632.4 (331.8)	699.5 (374.3)	752.7 (383.3)	<0.001
Vitamin B-12 deficiency [*n* (%)][7]	41 ± 2.7	12 ± 3.3	15 ± 3.2	4 ± 1.6	10 ± 2.2	0.12
Folate deficiency and anemia [*n* (%)]						
WHO criteria	1 (<0.1)	0.0	0.0	0.0	1 (0.9)	—
Beutler's criteria	0.0	0.0	0.0	0.0	0.0	—

[1] The REGARDS study had 30,239 participants; from this larger cohort, we selected a random sample of 1547 REGARDS participants aged ≥50 y with a baseline complete blood count and measured their serum folate and cobalamin concentrations for this analysis. One participant was excluded because of an insufficient stored sample for serum folate testing; the final analytic sample size was 1546. *P* values for differences between black and white races were analyzed by using the chi-square test for categorical variables and the independent *t* test for continuous variables. REGARDS, Reasons for Geographic and Racial Differences in Stroke.

[2] Mean ± SD (all such values).

[3] Beutler's criteria for anemia were defined as follows: white male aged 20–59 y, 13.7 g/dL; white male aged ≥60 y, 13.2 g/dL; white female aged ≥20 y, 12.2 g/dL; black male aged 20–59 y, 12.9 mg/dL; black male aged ≥60, 12.7 g/dL; and black female aged ≥20 y, 11.5 g/dL.

[4] Defined as a mean corpuscular volume >95 fL.

[5] All values are medians; IQRs in parentheses.

[6] Defined as a serum folate concentration <6.8 nmol/L (<3 ng/mL)

[7] Cobalamin deficiency was defined as a serum cobalamin concentration ≤148 pmol/L.

TABLE 3

Characteristics of the 2 folate-deficient participants in the REGARDS study, 2003–2007[1]

Characteristics	Participant 1	Participant 2
Demographics		
Age (y)	54	66
Sex	F	F
Race	Black	Black
Region[2]	Stroke buckle	Stroke belt
Education	Some college	High school
BMI (kg/m^2)	33.5	25.8
Smoking	Current	Never
Alcohol use	Past	Never
Dietary intake		
Daily energy intake (MJ)	314.8×10^5	442.3×10^5
Estimated folate intake from fortified foods (FFQ) (μg/d)	50.3	169.9
Estimated folate intake from natural foods (FFQ) (μg/d)	97.3	67.5
Multivitamin use		
Multivitamin use in medication inventory	No	No
Multivitamin use in FFQ	No	Yes
Hematologic variables		
Serum folate [nmol/L (ng/mL)]	6.1 (2.7)	4.3 (1.9)
Serum vitamin B-12 (pmol/L)[3]	280.2	208.6
Hematocrit (%)	36.3	36.1
Hemoglobin concentration (g/dL)	12.6	11.8
MCV(fL)[4]	113	99
Anemia by using the WHO definition	No	Yes
Anemia by using Beutler's definition (24)	No	No
C-reactive protein concentration (mg/L)[5]	43.8	17.4
Past medical history		
Participant reported stroke at baseline	Yes	Yes
History of cancer diagnosis	No	—
Self-reported health	Good	Good
Self-reported hypertension	Yes	No
Self-reported diabetes	No	No

[1] The REGARDS study had 30,239 participants; from this larger cohort, we selected a random sample of 1547 REGARDS participants aged ≥50 y with a baseline complete blood count and measured their serum folate and cobalamin concentrations for this analysis. FFQ, food-frequency questionnaire; MCV, mean corpuscular volume; REGARDS, Reasons for Geographic and Racial Differences in Stroke.

[2] The stroke buckle is a region within the stroke belt that has a higher stroke mortality rate than the rest of the stroke belt. The stroke buckle includes the coastal plains of Georgia, North Carolina, and South Carolina. The stroke belt is a region in the Southeastern part of the United States with high stroke mortality. It comprises the following 8 states: Georgia, North Carolina, South Carolina, Mississippi, Tennessee, Alabama, Louisiana, and Arkansas.

[3] Vitamin B-12 deficiency was defined as a serum vitamin B-12 concentration ≤148 pmol/L.

[4] MCV >95 fL was used as the cutoff for macrocytosis.

[5] Elevated C-reactive protein concentration was defined as >10 mg/L.