Lower Plasma Vitamin B-6 is Associated with 2-Year Cognitive Decline in the Boston Puerto Rican Health Study

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ABSTRACT

Background: Despite its important role in cognitive development and regulation of nervous system function, vitamin B-6 has been under-studied in relation to cognitive aging.

Objective: We investigated whether plasma pyridoxal-5'-phosphate (PLP, vitamin B-6) concentrations were associated with cognitive function and subsequent cognitive decline.

Methods: In a longitudinal study of 949 participants (aged 45–75 y at baseline; 70% women) from the Boston Puerto Rican Health Study cohort, we examined the association between baseline plasma PLP and baseline cognitive function and 2-y cognitive decline. Cognitive function was assessed with an in-person 7-test cognitive battery, at baseline and 2-y follow-up. We also used logistic regression to estimate the odds of major 2-y decline in global cognitive function (defined as decline ≥ 1 SD below the mean), as well as decline in executive function and memory. We also used multivariable linear regression to calculate adjusted mean differences in cognitive scores, and 95% CIs, across tertiles of plasma PLP at baseline, as well as cross-sectional and longitudinal associations with individual test scores.

Results: In analyses adjusted for potential confounders, the OR of having a major 2-y decline in global cognitive function was 2.46 (95% CI: 1.49, 4.05; *P*-trend: 0.001) among participants in the lowest tertile of PLP compared with those in the top tertile of PLP. The association of PLP with cognition was stronger in participants older than 55 y at baseline (OR for bottom to top tertile: 4.58; 95% CI: 2.02, 10.35; *P*-interaction: 0.01) compared with those 55 y old or younger, as well as in ever smokers (OR for bottom to top tertile: 2.99; 95% CI: 1.45, 6.19; *P*-interaction: 0.02) compared with never smokers.

Conclusions: Lower baseline plasma PLP was associated with increased odds of 2-y cognitive decline in a cohort of Boston area Puerto Ricans. The association was stronger among older participants and among ever smokers. *J Nutr* 2019;149:635–641.

Keywords: epidemiology, nutrition, minority, aging, vitamin B-6, cognition, cohort, biomarker

Introduction

The recent WHO Ministerial Conference on Global Action Against Dementia identified top research priorities to reduce the global burden of dementia, 6 of which focused on identification,

prevention, and reduction in dementia risk (1). Nutrient deficiency is an important and modifiable target in the search for potential risk factors for cognitive decline and Alzheimer disease (2). Deficiency of B-vitamins has been suggested as a key risk factor linked to cognitive decline. Most prior studies of B-vitamins and cognition have focused on vitamin B-12, folate, and plasma homocysteine (3, 4). Few have examined whether vitamin B-6 deficiency may be related to cognitive decline independently of other B-vitamins and homocysteine (5–7). Most such studies have reported inconsistent findings (4, 8), largely due to variations in study design, outcome measures, and study populations. Therefore, there is a need for more evidence of the impact of vitamin B-6 on cognitive function, independent of other B-vitamins.

This work was supported by NIH grants P01 AG023394, P50 HL105185, and R01 AG055948 (to KLT). NP receives funding from NIH grant R01 NS097723.

Author disclosures: NP, TS, NS, XG, and KLT, no conflicts of interest.

Supplemental Tables 1–6 are available from the “Supplementary data” link in the online posting of the article and from the same link in the online table of contents at <https://academic.oup.com/jn/>.

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Abbreviations used: BPRHS, Boston Puerto Rican Health Study; CRP, C-reactive protein; MMSE, Mini Mental State Examination; PLP, plasma pyridoxal-5'-phosphate (vitamin B-6).

Vitamin B-6 is a key coenzyme involved in amino acid, lipid, and glucose metabolism, processes implicated in brain health and aging. It is also a strong antioxidant (9, 10). Low vitamin B-6 intake and plasma concentration may also contribute to systemic inflammation (11). Key dietary sources of vitamin B-6 include fortified breakfast cereal, chickpeas, meats, and some fruit and vegetables, including potatoes and bananas (12). Vitamin B-6 plays a key role in the synthesis of dopamine and serotonin, neurotransmitters important to cognitive function (13). Vitamin B-6 deficiency has, thus, been implicated as a potential modifiable risk factor for cognitive decline and Alzheimer disease.

Puerto Ricans living in the United States represent a significant portion of the US Hispanic population, with well-documented health disparities compared with non-Hispanic whites as well as Mexican Americans. To our knowledge, no studies to date have focused on the association between plasma vitamin B-6 and cognition in Puerto Ricans. Previous studies have shown that Puerto Rican adults living in Massachusetts suffer disproportionately from depression, type 2 diabetes, heart disease, and cognitive impairment (14–17), compared with nonminority Caucasians. Thus, it is important to identify modifiable factors that might be contributing to these disparities. The Boston Puerto Rican Health Study Cohort (BPRHS) has collected extensive dietary and lifestyle data in addition to blood measures and baseline and 2-y follow-up cognitive assessments, making it ideal for examining the impact of plasma pyridoxal-5'-phosphate (PLP, vitamin B-6) on cognition. In this study, we examined whether low plasma PLP status was associated with baseline cognition and 2-y cognitive decline in the BPRHS.

Methods

Study design and participants

The BPRHS was approved by Institutional Review Boards at Tufts University and the University of Massachusetts Lowell. All study participants provided written informed consent. Study design, methodology, recruitment and selection of participants, procedures of blood collection, measurement of vitamin B-6 status, cognitive assessment, and information regarding relevant covariates were described in previous publications (9, 18–21). Briefly, 1499 participants in the Boston Puerto Rican Study were 45–75 y old at baseline. Eligible participants were enrolled if they self-identified as Puerto Rican, were able to answer questions in English or Spanish, and lived in the Boston, MA metropolitan area at the time of the study. Study participants were excluded if they had a low score (≤ 10) on the Mini Mental State Exam (MMSE) (22). This cutoff indicates severe dementia; we included this cutoff not for the purposes of excluding demented individuals but as an effort to limit floor effects for the other cognitive tests administered to this cohort. Participants completed cognitive testing at baseline and at the 2-y follow-up. The participants underwent anthropometric and blood-pressure measurements at study baseline and provided a blood sample. They completed cognitive testing at baseline and year 2 of follow-up, as detailed below.

Plasma vitamin B-6 exposure

Plasma vitamin B-6 was measured as PLP, using tyrosine decarboxylase, as described by Shin-Buehring et al. (23). The samples were light protected. The intra- and interassay CVs for the PLP assay were 5% and 6–8%, respectively. In our study sample, plasma PLP ranged from 7.2 to 737 nmol/L with a mean of 57.0 nmol/L.

Assessment of cognitive function

Study participants completed a cognitive testing battery administered by a trained research assistant in either English (2%) or Spanish (98%)

(20). The MMSE was administered to assess general cognitive function (possible score range: 0–30) (24). Cognitive tests were administered from the neuropsychological battery of Artiola I Fortuny et al. (25), which was designed and validated to assess Spanish-speaking residents of North America: a 16-word list learning task to assess verbal memory, with subscores including learning and immediate recall (the sum of words recalled over 5 learning trials; possible score range: 0–80), recognition (the number of words discriminated correctly from a longer word list after a 2.5–35 min delay; possible score range: 0–16), and percentage retention (calculated by dividing the number of words recalled after the delay by the number of correct responses on the fifth learning trial; possible score range: 0–100); digit span forward (possible score range: 0–16) and backward (possible score range: 0–14), to assess attention and working memory; the Stroop test (min score 0 and no upper limit), to measure cognitive flexibility, response inhibition, and processing speed; and letter fluency (min score 0 and no upper limit), to assess the speed at which one can provide exemplars to a category (three 1-min trials during which participants were asked to name as many words as they could that start with a given letter). In addition, clock drawing (possible range: 0–3) (26) and figure copying (possible range: 0–27) tasks were given, both to assess visual-spatial organization. Scores for figure copying were weighted for the complexity of the figure copied: 1 point for easy figures and 4 points for the most difficult. Higher scores on each of these tests signify better cognition. The same set of tests were administered at baseline and at the 2-y follow-up.

Cognitive outcome variables used in statistical analyses

Our primary analyses focused on the association between tertile categories of baseline plasma PLP and a global cognitive change score. This score, described in previous work (21), was calculated as a mean of the z scores for the 2-y changes in each of the following cognitive scores: MMSE, word list learning, recognition, percentage retention, Stroop, letter fluency, digit span forward and backward, clock drawing, and weighted figure copying. For analyses of 2-y global cognitive change, we created a binary variable representing “decline,” which was defined by a drop in global cognitive score ≥ 1 SD from the group mean (19, 27). We also performed cross-sectional analyses of the association between baseline plasma PLP and a baseline global cognitive score (average of the z scores of the baseline values of the following cognitive scores: MMSE, word list learning, recognition, percentage retention, Stroop, letter fluency, digit span forward and backward, clock drawing, and weighted figure copying).

We also examined associations between baseline plasma PLP and 2-y change in each individual test score, as well as calculating a difference in change in cognitive test score per 1 unit decrease in $\ln(\text{PLP})$. Changes in individual test scores were calculated by subtracting the score at baseline from the score at 2 y for each test, thus negative values represented deterioration and positive values represented improvement in cognitive function. Cross-sectional analyses of the association between baseline plasma PLP and individual cognitive tests were also conducted.

The BPRHS also conducted MMSE, but not other cognitive testing, during the 5-y follow-up. We thus examined the association between baseline plasma PLP and 5-y changes in the MMSE. Because a sizable proportion of participants (43%) did not attempt question 8 of the MMSE (counting down from 100 in 7s), for these analyses we recoded the MMSE scores at baseline and year 5, assigning a value of 0 to component 8 of the MMSE total score if it was not attempted.

In addition, and as described previously (21), we used principal components analysis to quantify 2 major cognitive function factors: 1) executive function and 2) memory, and linked plasma PLP to change in these 2 factors. Based on loadings for individual baseline test scores on the 2 principal components, the individual components were designated as measuring the cognitive domains of executive function and memory. Composite scores were also computed for year 2 follow-up testing data, using the means, SDs, and scoring coefficients from the baseline principal components analysis. Difference scores for baseline–follow-up were calculated for each domain, and descriptive statistics were obtained for these difference scores. For each cognitive domain, “major decline” was defined by a drop of ≥ 1 SD from the group

TABLE 1 Characteristics of 949 study participants of the Boston Puerto Rican Health Study with complete baseline plasma PLP (vitamin B-6) data and complete cognitive data at baseline and 2-y follow-up¹

	Tertile 1 (<i>n</i> = 315) ²	Tertile 2 (<i>n</i> = 317) ²	Tertile 3 (<i>n</i> = 317) ²	<i>P</i> -trend
Plasma PLP, ³ [mean (range)], nmol/L	20.9 (7.2–31.3)	40.4 (31.4–51.8)	82.0 (52.0–217.6)	
Age, y	56.1 ± 7.2	56.2 ± 7.5	56.9 ± 7.9	0.22
BMI, kg/m ²	32.4 ± 7.4	32.3 ± 6.4	30.7 ± 5.5	0.006
Women	75	75	70	0.10
Past smoker	30	33	31	0.87
Current smoker	30	23	16	<0.001
Less than high school education	41	40	42	0.93
Current alcohol use	36	43	52	<0.001
Past alcohol use	33	28	24	0.002
Diabetes	40	33	34	0.04
High CRP (≥3 mg/dL)	67	62	41	<0.001
Hypertension ⁴	67	66	67	0.74
Plasma homocysteine, μmol/L	9.3 ± 4.7	9.0 ± 5.8	8.3 ± 3.0	0.001
Serum vitamin B-12, pg/mL	482 ± 240	522 ± 245	636 ± 322	<0.001
Serum folate, ng/mL	16.2 ± 6.6	18.4 ± 7.6	22.9 ± 10.5	<0.001
Plasma vitamin C, mg/dL	0.7 ± 0.4	0.8 ± 0.4	0.9 ± 0.4	<0.001
Multivitamin user	24	28	39	<0.001
Presence of <i>APOE</i> ⁵	18	20	22	0.35
Household income (US\$)	14,788 ± 14,959	17,833 ± 16,959	20,786 ± 25,461	<0.001
Family size (inc. self)	2.4 ± 1.5	2.4 ± 1.5	2.1 ± 1.3	0.14
Married and living with spouse	25	35	31	0.10
Ever worked	89	90	92	0.40
MMSE	23.7 ± 3.0	23.7 ± 2.9	24.2 ± 3.1	0.03
Word list learning	38.8 ± 10.6	37.8 ± 10.7	39.0 ± 9.4	0.50
Recognition	13.0 ± 2.6	13.0 ± 2.6	13.4 ± 2.3	0.07
Percentage retention	81.3 ± 32.1	81.0 ± 31.7	86.2 ± 27.3	0.05
Stroop	22.6 ± 10.1	23.9 ± 10.4	24.9 ± 10.4	<0.001
Letter fluency	23.7 ± 9.2	24.8 ± 10.4	25.6 ± 10.4	0.01
Digit span forward	7.2 ± 1.8	7.5 ± 2.0	7.6 ± 1.9	0.002
Digit span backward	3.4 ± 1.4	3.5 ± 1.5	3.7 ± 1.6	0.01
Clock drawing	2.2 ± 1.0	2.2 ± 0.9	2.1 ± 0.9	0.78

¹Values are means ± SDs or percentages unless otherwise indicated. CRP, C-reactive protein; PLP, plasma pyridoxal-5'-phosphate.

²Tertiles are not perfectly even because of overlapping PLP values among participants.

³PLP values presented were back-transformed.

⁴Participants with mean systolic blood pressure ≥140 mm Hg or mean diastolic blood pressure ≥90 mm Hg, or those taking medication for hypertension, were classified as having hypertension.

⁵Apolipoprotein E.

mean difference score (19, 27). After creating the executive function and memory component scores, we used logistic regression to model the OR of “major decline”, in the executive function and memory component scores, respectively, by tertile of plasma PLP. Tertiles were used to maximize power in the analyses of decline.

Covariates

On the baseline study questionnaire, participants provided information on age, years of education, household income, marital status, employment status, family size, acculturation, migration history, smoking status, and alcohol intake (Table 1). Trained field workers measured body weight and height during the interview, and BMI (in kg/m²) was computed. The field workers also measured blood pressure at 3 time points during the study. Participants with mean systolic blood pressure ≥140 mm Hg or a mean diastolic blood pressure ≥90 mm Hg, or those taking medication for hypertension, were classified as having hypertension. A certified phlebotomist drew fasting (12-h) blood samples in the home of the study participants on a day after the interview. Participants with fasting plasma glucose >7.0 mmol/L or those who reported taking medications for diabetes (insulin or oral medicines) were classified as having type 2 diabetes (28). The TaqMan single nucleotide polymorphism genotyping assay (Applied Biosystems) was used to assess Apolipoprotein E (*APOE*) genotype, with a success

rate of 95%. Serum vitamin D was measured in fasting blood samples obtained at baseline as 25-hydroxyvitamin D, by extraction followed by 251 radioimmunoassay with a Packard COBRA II Gamma Counter (DiaSorin Inc.). Plasma folate and vitamin B-12 concentrations were determined by a radioassay method using a commercially available kit from Biorad. Total homocysteine in plasma was determined by a method based on the principles described by Araki and Sako (29). Ascorbate (vitamin C) was analyzed by paired-ion, reversed-phase High Performance Liquid Chromatography (HPLC) coupled with electrochemical detection.

Statistical analyses

Statistical analyses were performed with R version 3.3.1 (R Foundation). All analyses were restricted to participants with complete cognitive data at baseline and 2-y follow-up and nonmissing baseline plasma PLP concentration. Participants with extreme PLP values (>220, representing the 97.5th percentile in our baseline data) were excluded from analyses (*n* = 29). The final number of participants, after exclusions for missing or extreme exposure or missing outcome, was 949.

Participants were categorized according to their baseline plasma PLP tertile. For our cross-sectional analyses of PLP and baseline cognition, we used multivariable linear regression and calculated the adjusted

TABLE 2 Cross-sectional associations between baseline cognitive function and baseline plasma PLP (vitamin B-6) concentration among participants of the Boston Puerto Rican Health Study¹

		Tertile 1 (n = 315)	Tertile 2 (n = 317)	Tertile 3 (n = 317)	P-trend
PLP ² range, nmol/L		7.2–31.3	31.4–51.8	52.0–217.6	
Global cognitive score	Model 1 ³	−0.13 (−0.20, −0.05)	−0.08 (−0.16, −0.01)	REF	0.003
	Model 2 ⁴	−0.12 (−0.20, −0.04)	−0.09 (−0.16, −0.01)	REF	0.007
	Model 3 ⁵	−0.07 (−0.15, 0.02)	−0.05 (−0.13, 0.03)	REF	0.19

¹n = 949. PLP, plasma pyridoxal-5'-phosphate.

²PLP values presented were back-transformed.

³Model 1 adjusted for age in years, sex, and education (<5th grade, 5–8th grade, 9–12th grade, some college or college degree, some graduate school).

⁴Model 2 adjusted for age in years, sex, education, diabetes (yes/no), smoking (never/past/current), alcohol use (never/past/current), hypertension (yes/no), BMI (normal weight/overweight/obese), and presence of Apolipoprotein E (APOE) 4 (yes/no).

⁵Model 3 is equivalent to model 2 with additional adjustment for ln plasma vitamin C, ln serum vitamin D, ln serum vitamin B-12, ln serum folate, multivitamin supplement use (yes/no), and ln plasma homocysteine.

mean differences and 95% CIs in cognitive decline across tertiles of plasma PLP. Logistic regression was used to estimate the odds of major global cognitive decline, executive function decline, and memory decline according to tertiles of PLP. For tests of trend, we used the median value of each tertile as a continuous variable to minimize the influence of outliers. Plasma and serum values of biomarkers were ln transformed before analysis to improve normality, as appropriate.

We built our multivariable models in sequential steps, as presented in [Tables 2–4](#). First, a model adjusted for age in years, sex, and education (<5th grade, 5–8th grade, 9–12th grade, some college or college degree, some graduate school) was fitted (Model 1 in [Tables 2–4](#)). Subsequently, health-related risk factors for cognitive decline including diabetes (yes/no), smoking (never/past/current), alcohol use (never/past/current), hypertension (yes/no), BMI [normal weight (<25)/overweight (25–<30)/obese (≥30)], and presence of APOE 4 (yes/no) were added to the model (Model 2 in [Tables 2–4](#)). Finally, in a third model, other relevant nutrients were added as covariates including ln plasma vitamin C, ln serum vitamin D, ln serum vitamin B-12, ln serum folate, multivitamin supplement use (yes/no), ln plasma homocysteine, and time in days between the baseline and year 2 visits (Model 3 in [Tables 2](#) and [3](#)). Longitudinal models of 2-y cognitive change were additionally adjusted for the appropriate baseline cognitive score. We used the missing indicator method to address the missingness of all covariates, other than the exposure (plasma PLP), in our models. Participants missing plasma PLP or those missing the cognitive outcome were excluded from all analyses.

We further examined whether the association between plasma PLP and cognitive decline was modified by age (≤55 y compared with >55 y), smoking, and sex by including multiplicative terms in our regression models and conducting stratified analyses. Interaction analyses were performed for the global cognitive decline logistic regression models by adding an interaction term between the relevant effect modifier and plasma PLP to the fully adjusted model (Model 3), as appropriate, and using the likelihood ratio test.

We conducted sensitivity analyses, adjusting for C-reactive protein (CRP) (<3 compared with ≥3) ([30](#)), a marker of inflammation. In these models, white blood cell count (10³/μL) was included with CRP because of the possibility of CRP elevation with recent infection. We also performed additional sensitivity analyses adjusting for depressive symptomatology using the Center for Epidemiologic Studies Depression scale (CESD) score, household income, marital status (married and living with spouse compared with not), employment status (ever worked compared with not), and psychological acculturation scale, sequentially and together. We performed additional sensitivity analyses using a category of vitamin B-6 defined as low [PLP concentration (c_{PLP}) <30 nmol/L as opposed to ≥30 nmol/L], as well as 3 categories of sufficient (c_{PLP} ≥30 nmol/L), insufficient (20 ≤ c_{PLP} <30 nmol/L), and deficient (c_{PLP} <20 nmol/L) instead of tertiles ([31](#), [32](#)). To address the possibility of selection bias due to only including participants with complete baseline and 2-y cognitive assessments, we conducted a cross-sectional sensitivity analysis among all participants who had cognitive and vitamin B-6 data at baseline (n = 1274).

TABLE 3 Association between tertiles of plasma PLP (vitamin B-6) and 2-y change in global cognitive function, as odds of ≥1 SD decline among participants of the Boston Puerto Rican Health Study¹

		Tertile 1	Tertile 2	Tertile 3	P-trend ²
Declined ³		n = 64	n = 52	n = 41	
Did not decline		n = 251	n = 265	n = 276	
PLP ⁴ range: nmol/L		7.2–31.3	31.4–51.8	52.0–217.6	
OR (95% CI) for decline	Model 1 ⁵	1.94 (1.24, 3.04)	1.37 (0.87, 2.17)	REF	0.03
	Model 2 ⁶	2.04 (1.26, 3.28)	1.47 (0.91, 2.37)	REF	0.004
	Model 3 ⁷	2.46 (1.49, 4.05)	1.65 (1.09, 2.51)	REF	0.001

¹n = 949. PLP, plasma pyridoxal-5'-phosphate.

²P-trend was calculated using the median value of each quantile as a continuous variable.

³n = number of participants who met the criterion for “decline”: ≥1 SD decline in global cognitive score.

⁴PLP values presented were back-transformed.

⁵Model 1 is adjusted for age in years, sex, and education (<5th grade, 5–8th grade, 9–12th grade, some college or college degree, some graduate school).

⁶Model 2 is adjusted for age in years, sex, education (<5th grade, 5–8th grade, 9–12th grade, some college or college degree, some graduate school), diabetes (yes/no), smoking (never/past/current), alcohol use (never/past/current), hypertension (yes/no), BMI (normal weight/overweight/obese), and presence of Apolipoprotein E (APOE) 4 (yes/no).

⁷Model 3 is equivalent to model 2 with additional adjustment for ln plasma vitamin C, ln serum vitamin D, ln serum vitamin B-12, ln serum folate, multivitamin supplement use (yes/no), plasma homocysteine, and time in days between baseline and year 2 visits.

TABLE 4 Association between tertiles of plasma PLP (vitamin B-6) and 2-y change in global cognitive function, as odds of ≥ 1 SD decline, stratified by age, BMI, smoking status, and sex among participants of the Boston Puerto Rican Health Study¹

	<i>n</i>		Tertile 1	Tertile 2	Tertile 3	<i>P</i> -trend ²	<i>P</i> -interaction ³
PLP ⁴ range nmol/L			7.2–31.3	31.4–51.8	52.0–217.6		
Age							0.01
Older (>55 y)	477	Model 1 ⁵	3.05 (1.58, 5.85)	1.83 (0.94, 3.55)	Ref	0.0008	
		Model 2 ⁶	4.58 (2.02, 10.35)	2.45 (1.15, 5.22)	Ref	0.0003	
Younger (≤ 55 y)	472	Model 1 ⁵	1.30 (0.69, 2.46)	1.04 (0.54, 2.00)	Ref	0.24	
		Model 2 ⁶	1.65 (0.80, 3.41)	1.36 (0.71, 2.61)	Ref	0.21	
Smoking							0.02
Never	432	Model 1 ⁵	1.25 (0.65, 2.39)	1.22 (0.69, 2.19)	Ref	0.99	
		Model 2 ⁶	1.77 (0.92, 3.23)	1.63 (1.58, 1.82)	Ref	0.61	
Ever	515	Model 1 ⁵	2.78 (1.46, 5.28)	1.58 (0.80, 3.10)	Ref	0.002	
		Model 2 ⁶	2.99 (1.45, 6.19)	1.80 (0.10, 3.34)	Ref	0.01	
Sex							0.72
Female	254	Model 1 ⁵	1.89 (1.10, 3.28)	1.24 (0.77, 2.02)	Ref	0.04	
		Model 2 ⁶	2.18 (2.10, 2.26)	1.35 (0.67, 3.02)	Ref	0.11	
Male	695	Model 1 ⁵	1.98 (0.85, 4.65)	1.66 (0.71, 3.90)	Ref	0.29	
		Model 2 ⁶	2.56 (0.98, 6.70)	2.13 (0.83, 5.51)	Ref	0.06	

¹Values are OR (95% CI) unless indicated otherwise. *n* = 949. PLP, plasma pyridoxal-5'-phosphate.

²*P*-trend was calculated using the median value of each quantile as a continuous variable.

³*P*-interaction calculated with the likelihood ratio test comparing a model with the interaction term to a model without the interaction term.

⁴PLP values presented were back-transformed.

⁵Adjusted for age in years, sex, and education (<5th grade, 5–8th grade, 9–12th grade, some college or college degree, some graduate school).

⁶Adjusted for age in years, sex, education (<5th grade, 5–8th grade, 9–12th grade, some college or college degree, some graduate school), diabetes (yes/no), smoking (never/past/current), alcohol use (never/past/current), hypertension (yes/no), BMI (normal weight/overweight/obese), presence of Apolipoprotein E (APOE) 4, In plasma vitamin C, In serum vitamin D, In serum vitamin B-12, In serum folate, multivitamin supplement use (yes/no), plasma homocysteine, and time in days between baseline and year 2 visits.

Results

For our analysis, 949 participants had complete cognitive data at baseline and at the 2-y follow-up, as well as nonmissing plasma PLP concentration. Compared with participants who were excluded owing to missing data on exposure or outcome, included participants were younger (mean $\pm$ SD age 56.4 ± 7.5 compared with 58.0 ± 7.3 ; $P < 0.001$), had higher baseline MMSE score (mean $\pm$ SD: 23.9 ± 3.0 compared with 21.1 ± 3.8 ; $P < 0.001$), and a higher proportion were female (73.2% compared with 60.0%; $P < 0.001$).

At baseline, the study population overall had relatively low education, and high prevalence of diabetes, hypertension, and overweight or obesity (Table 1). Participants in the higher tertiles of plasma PLP had lower BMI (*P*-trend 0.006), lower prevalence of current smoking (*P*-trend < 0.001), higher prevalence of current (*P*-trend < 0.001) and past (*P*-trend = 0.002) alcohol use, lower prevalence of high CRP (*P*-trend < 0.001), lower prevalence of diabetes (*P*-trend 0.04), lower concentration of plasma homocysteine (*P*-trend 0.001), and higher household income (*P*-trend < 0.001) compared with those in the lowest tertile. Participants in the higher tertiles of plasma PLP had higher plasma vitamin B-12 (*P*-trend < 0.001), folate (*P*-trend < 0.001), and vitamin C concentrations (*P*-trend < 0.001) and were more likely to be multivitamin users at baseline (*P*-trend < 0.001) compared with those in the lower tertiles. Supplemental Table 1 presents the baseline and 2-y mean and SD of each of the cognitive measures administered in this study. Supplemental Table 2 presents the normative cognitive *t* scores for our study participants.

Plasma PLP was not associated with global cognition at baseline after adjusting for potential confounders. In fully adjusted models, the mean difference in baseline global cognitive score, comparing the lowest to the highest tertile of plasma PLP, was -0.07 (95% CI: $-0.15, 0.02$), and when comparing the middle to the highest tertile it was -0.05 (95%

CI: $-0.13, 0.03$). These results are presented in Table 2. For context, the mean difference in cognition associated with 1 y of aging in this cohort, after adjustment for sex and education, was -0.018 (95% CI: $-0.022, -0.013$).

Longitudinally, we found that the odds of major decline, defined as being ≥ 1 SD below the mean 2-y decline in global cognition, was 2.46 (95% CI: 1.49, 4.05) when comparing participants in the lowest to those in the highest tertile of PLP (*P*-trend across tertiles: 0.001) (Table 4). The association between low PLP and cognition was stronger among participants older than 55 y (OR for comparing the lowest to the highest PLP tertile: 4.58; 95% CI: 2.02, 10.35; *P*-trend: 0.0003), and was not as strong among those 55 y and younger (*P*-trend: 0.21) (*P*-interaction with age: 0.01). The association was also stronger among ever smokers (OR for comparing the top to the bottom tertile: 2.99; 95% CI: 1.45, 6.19; *P*-trend: 0.01) than among never smokers (*P*-trend: 0.61) (*P*-interaction with smoking: 0.02). We did not observe an interaction of the vitamin B-6–cognition relation with sex (*P*-interaction with sex: 0.72).

We found association, both at baseline and over 2 y, between PLP concentration and performance on several individual cognitive tests. In fully adjusted cross-sectional analyses, baseline plasma PLP was positively associated with performance on the Stroop test (*P*-trend: 0.03) (Supplemental Table 3). Lower baseline plasma PLP was also associated with negative 2-y changes in recognition (*P*-trend: 0.05) (Supplemental Table 4). Several other associations were marginal, but did not reach statistical significance. We did not observe a significant association between baseline plasma PLP concentration and 5-y change in MMSE score (*P*-trend: 0.74) (Supplemental Table 5).

We did not observe statistically significant associations between baseline PLP concentration and decline in executive function or memory component scores. The odds of a ≥ 1 SD decline in executive function for participants in the lowest compared with the highest tertile of plasma PLP were 1.51 (95%

CI: 0.62, 3.69), with a statistically nonsignificant *P*-trend of 0.19. Similarly, in logistic regression analyses using a ≥ 1 SD decline in the memory component score as the outcome, the OR for participants in the lowest compared with the highest tertile of plasma PLP was 1.32 (95% CI: 0.79, 2.32), with a statistically nonsignificant *P*-trend of 0.26. These results are presented in **Supplemental Table 6**. Owing to small case numbers, we were only able to perform analyses adjusted for age, sex, and education because addition of other covariates led to model overfitting, and therefore these analyses should be interpreted with caution.

In sensitivity analyses, adding CRP to our full regression models did not significantly affect the results (OR for 2-y decline: 2.43; 95% CI: 1.48, 4.01; *P*-trend: 0.01), and there was no effect modification by CRP (*P*-interaction: 0.40). Additional adjustment for depressive symptomatology using the Center for Epidemiologic Studies Depression Scale (CES-D) score, household income, marital status (married and living with spouse compared with not), employment status (ever worked compared with not), or psychological acculturation scale did not substantially alter the results. Our cross-sectional sensitivity analysis, including all participants ($n = 1274$) at baseline with available cognitive scores, regardless of the availability of 2-y cognitive scores, was not substantially different from our primary analyses.

Discussion

In this study of Puerto Rican adults living in the greater Boston area, we found evidence favoring an association between lower plasma PLP concentration and cognitive decline over 2 y, which is in general agreement with prior studies. The Veterans Affairs Normative Ageing Study of men reported a significant association between low (PLP <46 nmol/L) vitamin B-6 and cognitive decline over 3 y (4). A recent study in an Irish population of 155 participants, aged 60–88 y, also reported significant associations between plasma vitamin B-6 status and cognitive decline (8). In the MacArthur Study of Successful Aging, vitamin B-6 was associated, cross-sectionally, with better cognitive performance, although prospective results focusing on cognitive decline did not show significance (33).

Our study has several strengths, including the careful assessment of exposures and outcomes. Vitamin B-6 was measured as plasma PLP, which is a more objective measure of vitamin B-6 status than an estimate of dietary intake from questionnaires. Cognitive function was measured with an extensive neuropsychological battery of Artiola I Fortuny et al.'s (25), administered by trained study personnel in the native language of the study participants. Another important strength is the prospective nature of the study, which allowed us to examine change in cognitive function over 2 y as the primary outcome of interest. We were able to adjust for a wide range of potential confounders, including serum concentrations of other B-vitamins and homocysteine. We considered the possibility that the effect of vitamin B-6 on cognition may be through its impact on systemic inflammation. There was limited evidence of mediation by CRP, and adding CRP and white blood cell count to our models did not substantially alter the results. However, the interrelation between vitamin B-6, inflammation, and cognition is complex (11, 34) and there is some evidence that inflammation lowers plasma PLP concentrations (35), which must also be considered.

The focus on a US mainland Puerto Rican population is also a unique strength of this study, because it provides information on a modifiable dietary risk factor in a distinct and vulnerable population. However, because of the focus on Puerto Rican adults, our study is not entirely generalizable to other populations. Although we adjusted for a wide range of potential confounders, there may be other unmeasured variables that could mediate the association between plasma PLP concentration and cognitive decline. Also, the study did not include clinical assessments of the participants, so we did not have clinical designations of “normal,” “Mild Cognitive Impairment (MCI),” or “dementia.” Finally, because of the observational nature of this study, causal associations between plasma vitamin B-6 and cognitive function and decline cannot be established. There remains a possibility that the observed results are due to reverse causation, or residual confounding by other nutrients, deprivation, or socioeconomic status. Longer follow-up, in this and other cohorts, with better adjustment for factors associated with deprivation and socioeconomic status, will be needed to address this issue. There also remains a possibility of selection bias if participants with low PLP concentration and declining cognition selectively dropped out of the study between the baseline and year-2 visits. We conducted cross-sectional sensitivity analyses among all participants who had available PLP and cognitive data at baseline ($n = 1274$), regardless of whether they had participated in the 2-y follow-up. The results of these analyses were not substantially different from our main results. Finally, there is a possibility of a learning effect over 2 y due to participants learning tasks after completing the baseline assessment.

In our study population, 12% of the population was deficient in vitamin B-6 (PLP <20 nmol/L) and an additional 17% was insufficient ($20 \leq \text{PLP} \leq 30$ nmol/L). Given the findings of this study, more work is needed in both Puerto Rican and other populations to further understand the nature and extent of the association between vitamin B-6 and cognitive decline. If this association is confirmed, efforts should be taken to reduce vitamin B-6 deficiency.

Acknowledgments

We thank Esther Carver and Oladimeji (Jimmy) Akinlawon for assistance with this manuscript. The authors' contributions were as follows—NP, KLT, TS, and XG: designed and conducted the research; NP and NS: analyzed the data; NP: wrote the paper and had primary responsibility for final content; and all authors: read and approved the final manuscript.

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