Supplemental Methods 1

Certain fat-soluble nutrients were calculated as composite variables:

- Serum carotenoids (CAR): sum of *alpha*-carotene, *beta*-carotene and *cis*- and *trans*-lycopene
- Serum xanthophylls (XAN): sum of *beta*-cryptoxanthin, lutein and zeaxanthin
- Plasma saturated fatty acids (SFA): sum of myristic [14:0], palmitic [16:0], stearic [18:0], arachidic [20:0], docosanoic [22:0] and lignoceric acid [24:0]
- Plasma monounsaturated fatty acids (MUFA): sum of myristoleic [14:1n5], palmitoleic [16:1n7], *cis*-vaccenic [18:1n7], oleic [18:1n9], eicosenoic [20:1-n9], docosenoic [22:1n9] and nervonic acid [24:1n9]
- Plasma polyunsaturated fatty acids (PUFA): sum of linoleic [18:2n6], *alpha*-linolenic [18:3n3], *gamma*-linolenic [18:3n6], eicosadienoic [20:2n6], *homo-gamma*-linolenic [20:3n6], arachidonic [20:4n6], eicosapentaenoic [20:5n3], docosatetraenoic [22:4n6], docosapentaenoic-3 [22:5n3], docosapentaenoic-6 [22:5n6] and docosahexaenoic acid [22:6n3]
- Plasma total fatty acids (tFA): sum of the above mentioned 24 fatty acids

Body iron (BI) was calculated from the ratio of soluble transferrin receptor (sTfR) to serum ferritin (FER) using the formula of Cook *et al.* (1):

Body iron (mg/kg) = -[log10 (sTfR * 1000 / FER) - 2.8229] / 0.1207.

The sTfR concentration in this formula represents an adjusted concentration to make the Roche sTfR concentrations (assay used in NHANES 2003–2006) equivalent to the Flowers assay (2) used in the development of the body iron model (3):

Flowers sTfR = 1.5 * Roche sTfR + 0.35 mg/L.

Calculation of estimated glomerular filtration rate (eGFR) and stages of chronic kidney disease (CKD):

The National Kidney Disease Education Program (NKDEP) recommends the use of eGFR as the best overall index of kidney function in CKD (4) using an equation developed from the Modification of Diet in Renal Disease (MDRD) Study (5):

$$\text{eGFR (mL/(min·1.73 m}^2\text{))} = 175 \times (\text{serum creatinine})^{-1.154} \times (\text{age})^{-0.203} \times (0.742 \text{ if female}) \times (1.210 \text{ if African American}) \text{ (conventional units).}$$

We considered persons with eGFR < 60 mL/(min·1.73 m^2) to have stage 3–5 CKD according to the National Kidney Foundation classification system (6). Stages 1 (eGFR ≥ 90 mL/(min·1.73 m^2)) and 2 (eGFR 60–89 mL/(min·1.73 m^2)) of apparent kidney damage were assessed by the presence of albuminuria (i.e., abnormal amounts of urine albumin assessed by the urine albumin to urine creatinine ratio [ACR]) (6). Microalbuminuria was defined as ACR of 17–250 mg/g for men or 25–355 mg/g for women. The definition of macroalbuminuria was ACR of > 250 mg/g for men or > 355 mg/g for women (6). Definition of stages 1 and 2 requires that persistent albuminuria is assessed from two urine samples. However, only one urine sample is available in NHANES and was therefore used for categorization.

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Supplemental Table 1. Sample sizes for biomarkers of diet and nutrition by physiological factor for adults ≥20 y, NHANES 2003–2006[1,2,3]

Analyte		Fasting			Inflammation status[4]			Renal function[5]				Pregnancy[6]		
(matrix)[7]	All	<3 h	3–<8 h	≥8 h	All	No	Yes	All	Normal	Stage 1 or 2	Stage 3–5	All	No	Yes
FOL (S)	8944	1667	2275	5002	8937	6562	2375	8771	6707	1107	957	2473	1966	507
FOL (RBC)	8980	1676	2274	5030	8910	6535	2375	8746	6689	1106	951	2489	1980	509
PLP (S)	4489	1403	960	2126	4484	3257	1227	4406	3393	574	439	1321	1014	307
4PA (S)	4488	1402	960	2126	4483	3256	1227	4405	3392	574	439	1321	1014	307
B-12 (S)	8878	1644	2271	4963	8872	6514	2358	8710	6664	1094	952	2453	1951	502
tHcy (P)	8999	1677	2290	5032	8953	6569	2384	8787	6717	1110	960	2488	1978	510
MMA (P)	4338	270	1276	2792	4313	3189	1124	4226	3207	519	500	1111	920	191
VIC (S)	8892	1662	2263	4967	8890	6533	2357	8759	6699	1107	953	2455	1951	504
VIA (S)	4440	1386	945	2109	4439	3230	1209	4385	3377	571	437	1307	1001	306
VIE (S)	4440	1386	945	2109	4439	3230	1209	4385	3377	571	437	1307	1001	306
CAR (S)	4387	1376	927	2084	4386	3192	1194	4333	3332	566	435	1288	986	302
XAN (S)	4416	1384	938	2094	4415	3213	1202	4361	3357	569	435	1301	996	305
25OHD (S)	8993	1678	2290	5025	8977	6585	2392	8811	6736	1113	962	2486	1976	510
SFA (P)	1687	ND[8]	ND	1687	1681	1233	448	1657	1256	211	190	437	357	80
MUFA (P)	1662	ND	ND	1662	1656	1219	437	1631	1248	200	183	430	351	79
PUFA (P)	1786	ND	ND	1786	1779	1307	472	1754	1338	219	197	466	381	85
tFA (P)	1445	ND	ND	1445	1440	1064	376	1420	1080	173	167	366	300	66
FER (S)	2539	542	564	1433	2538	1645	893	2508	2247	240	21	2470	1964	506
sTfR (S)	2513	539	560	1414	2512	1630	882	2482	2222	239	21	2444	1943	501
BI (S)	2509	538	558	1413	2508	1627	881	2478	2220	237	21	2440	1940	500
uI (U)	3031	592	759	1680	2937	2192	745	2917	2243	371	303	865	685	180

Analyte	Fasting				Inflammation status[4]			Renal function[5]				Pregnancy[6]		
(matrix)[7]	All	<3 h	3–<8 h	≥8 h	All	No	Yes	All	Normal	Stage 1 or 2	Stage 3–5	All	No	Yes
GEN (U)	2963	537	761	1665	2864	2092	772	2853	2204	351	298	870	679	191
DAZ (U)	2963	537	761	1665	2864	2092	772	2853	2204	351	298	870	679	191
EQU (U)	2958	536	761	1661	2859	2088	771	2848	2201	350	297	870	679	191
DMA (U)	2950	537	758	1655	2851	2081	770	2841	2195	349	297	861	670	191
ETD (U)	2963	537	761	1665	2864	2092	772	2853	2204	351	298	870	679	191
ETL (U)	2963	537	761	1665	2864	2092	772	2853	2204	351	298	870	679	191
HbAA (B)	4093	240	1213	2640	4064	3008	1056	3984	3020	498	466	1061	875	186
HbGA (B)	4152	243	1221	2688	4121	3056	1065	4042	3083	491	468	1073	884	189

[1] 25OHD, 25-hydroxyvitamin D; 4PA, 4-pyridoxic acid; B-12, total cobalamin; BI, body iron; CAR, carotenes; DAZ, daidzein; DMA, O-desmethylangolensin; EQU, equol; ETD, enterodiol; ETL, enterolactone; FER, ferritin; FOL, folate; GEN, genistein; HbAA, acrylamide hemoglobin adduct; HbGA, glycidamide hemoglobin adduct; MMA, methylmalonic acid; PLP, pyridoxal-5'-phosphate; sTfR, soluble transferrin receptor; tFA, total fatty acids; tHcy, total homocysteine; uI, urinary iodine; VIA, retinol; VIC, ascorbic acid; VIE, *alpha*-tocopherol; XAN, xanthophylls.

[2] Iron status indicators (FER, sTfR and BI) were only measured in women of reproductive age, thus our analysis was limited to women 20–49 y of age.

[3] MMA, SFA, MUFA, PUFA, tFA, HbAA, and HbGA data only available for NHANES 2003–2004; PLP, 4PA, VIA, VIE, CAR, and XAN data only available for NHANES 2005–2006.

[4] C-reactive protein (CRP, mg/L) was used to assess inflammation status (<5, no inflammation; ≥5, inflammation).

[5] Estimated glomerular filtration rate (eGFR) was used to assess renal function; normal was defined as eGFR ≥60 mL/(min·1.73 m^2) and absence of albuminuria; stage 1 or 2 chronic kidney disease (CKD) was defined as eGFR ≥60 mL/(min·1.73 m^2) and presence of albuminuria; stage 3–5 CKD was defined as eGFR <60 mL/(min·1.73 m^2).

[6] Women 20–49 y of age.

[7] S, serum; P, plasma; U, urine; B, whole blood.

[8] No data.

Supplemental Table 2. Descriptive information for adults ≥20 y by physiological factor, NHANES 2003–2006[1]

Factor	Category	Full sample 2003–2006	Full sample 2003–2004	Full sample 2005–2006	Fatty acids fasting sample 2003–2004	Phytoestrogens 1/3 sample 2003–2006	Iodine 1/3 sample 2003–2006
Fasting	<3 h	19.5	5.22	33.7	0.00	18.6	20.1
	3–<8 h	24.1	29.2	19.1	0.00	24.4	23.9
	≥8 h	56.3	65.6	47.2	100	57.0	56.0
Inflammation status[2]	No	77.1	77.3	76.8	76.6	76.2	78.1
	Yes	22.9	22.7	23.2	23.4	23.8	21.9
Renal function[3]	Normal	81.4	82.0	80.8	82.4	81.8	80.8
	CKD Stage 1 or 2	10.5	9.96	10.9	10.2	10.4	11.1
	CKD Stage 3–5	8.15	8.01	8.29	7.39	7.86	8.07
Pregnancy status[4]	No	93.7	94.6	92.7	95.4	92.9	94.1
	Yes	6.33	5.39	7.28	4.60	7.10	5.91

[1] Values represent weighted percentage (%) by various Mobile Examination Center weights.

[2] C-reactive protein (CRP, mg/L) was used to assess inflammation status (<5, no inflammation; ≥5, inflammation).

[3] Estimated glomerular filtration rate (eGFR) was used to assess renal function; normal was defined as eGFR ≥60 mL/(min·1.73 m^2) and absence of albuminuria; stage 1 or 2 chronic kidney disease (CKD) was defined as eGFR ≥60 mL/(min·1.73 m^2) and presence of albuminuria; stage 3–5 CKD was defined as eGFR <60 mL/(min·1.73 m^2).

[4] Pregnancy status for women 20–49 y.

Supplemental Table 3. Spearman correlation coefficients describing bivariate associations between each biomarker of diet and nutrition and selected continuous physiological factors for adults ≥20 y, NHANES 2003–2006[1,2,3,4]

Analyte (matrix)[5]	Fasting time	Inflammation status[6]	Renal function[7]
FOL (S)	-0.05*	-0.03*	-0.18*
FOL (RBC)	-0.04*	0.09*	-0.24*
PLP (S)	-0.04*	-0.26*	-0.04*
4PA (S)	-0.07*	-0.03	-0.32*
B-12 (S)	-0.01	-0.04*	-0.03
tHcy (P)	0.01	0.02	-0.44*
MMA (P)	-0.12*	0.03	-0.40*
VIC (S)	-0.04*	-0.16*	-0.05*
VIA (S)	0.02	-0.11*	-0.36*
VIE (S)	-0.01	0.08*	-0.28*
CAR (S)	-0.03	-0.21*	-0.04
XAN (S)	0.00	-0.20*	-0.03
25OHD (S)	-0.01	-0.13*	-0.15*
SFA (P)	-0.00	0.22*	-0.09*
MUFA (P)	0.02	0.19*	-0.14*
PUFA (P)	0.05*	0.11*	-0.07*
tFA (P)	0.01	0.18*	-0.10*
FER (S)	0.00	0.13*	-0.11*
sTfR (S)	0.02	0.11*	-0.01
BI (S)	0.02	0.07*	-0.06*
uI (U)	-0.03	0.07*	-0.09*
GEN (U)	0.04	-0.04	-0.01
DAZ (U)	0.03	-0.05	-0.03
EQU (U)	0.07*	0.01	-0.01
ODMA (U)	0.06*	-0.08*	-0.08*
ETD (U)	0.12*	-0.02	0.00
ETL (U)	0.07*	-0.13*	-0.08*
HbAA (B)	-0.01	-0.04*	0.16*
HbGA (B)	0.01	0.07*	0.17*

[1] 25OHD, 25-hydroxyvitamin D; 4PA, 4-pyridoxic acid; B-12, total cobalamin; BI, body iron; CAR, carotenes; DAZ, daidzein; DMA, O-desmethylangolensin; EQU, equol; ETD, enterodiol; ETL, enterolactone; FER, ferritin; FOL, folate; GEN, genistein; HbAA, acrylamide hemoglobin adduct; HbGA, glycidamide hemoglobin adduct; MMA, methylmalonic acid; PLP, pyridoxal-5'-phosphate; sTfR, soluble transferrin receptor; tFA, total fatty acids; tHcy, total homocysteine; uI, urinary iodine; VIA, retinol; VIC, ascorbic acid; VIE, *alpha*-tocopherol; XAN, xanthophylls.

[2] Iron status indicators (FER, sTfR and BI) were only measured in women of reproductive age, thus our analysis was limited to women 20–49 y of age.

[3] MMA, SFA, MUFA, PUFA, tFA, HbAA, and HbGA data only available for NHANES 2003–2004; PLP, 4PA, VIA, VIE, CAR, and XAN data only available for NHANES 2005–2006.

[4] Sample sizes for each biomarker by variable can be found in Supplemental Table 1.

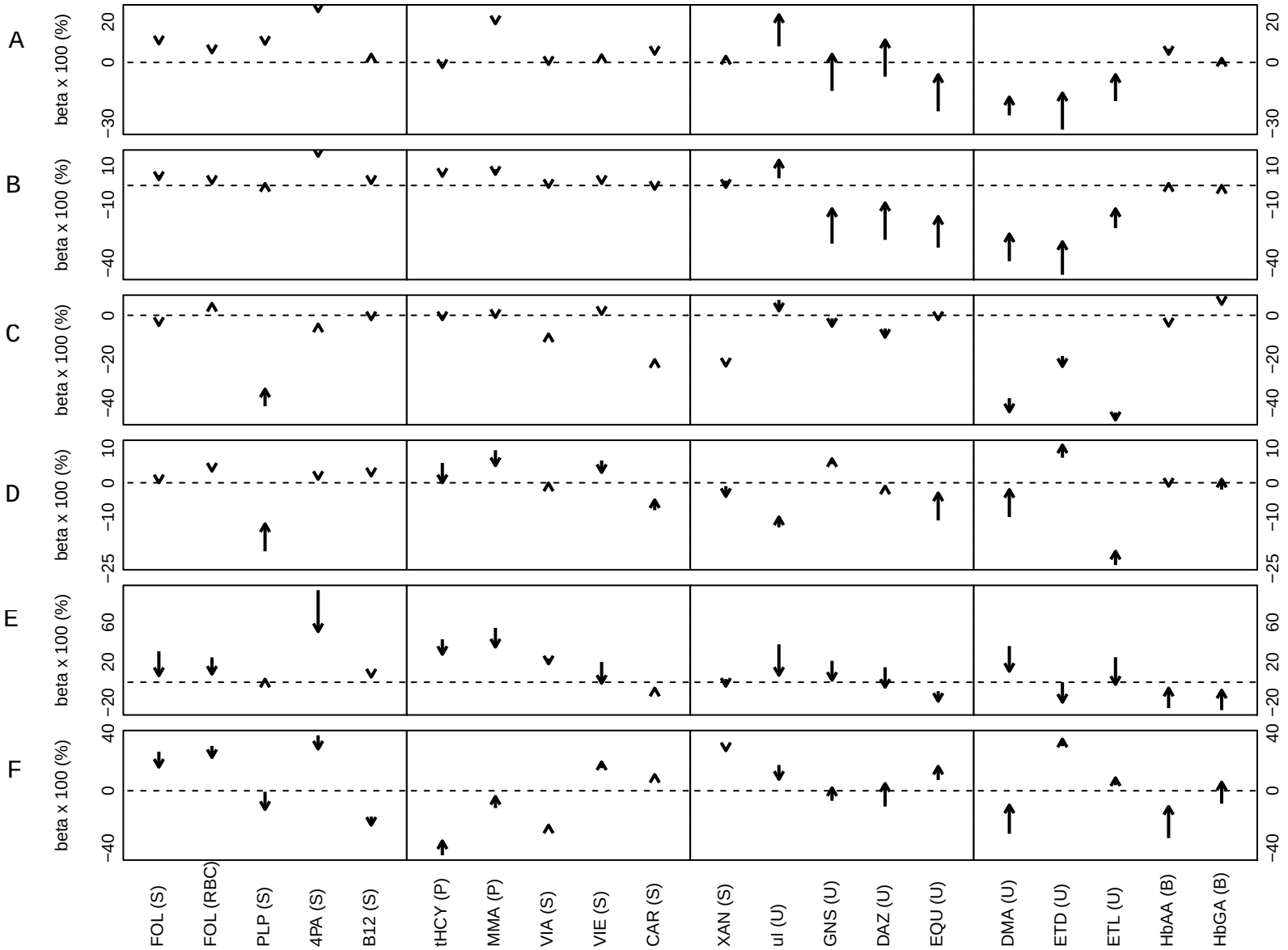
[5] S, serum; P, plasma; U, urine; B, whole blood.

[6] C-reactive protein (CRP, mg/L) was used to assess inflammation status (<5, no inflammation; ≥5, inflammation).

[7] Estimated glomerular filtration rate (eGFR) was used to assess renal function; normal was defined as eGFR ≥60 mL/(min·1.73 m^2) and absence of albuminuria; stage 1 or 2 chronic kidney disease (CKD) was defined as eGFR ≥60 mL/(min·1.73 m^2) and presence of albuminuria; stage 3–5 CKD was defined as eGFR <60 mL/(min·1.73 m^2).

* Significant correlation; P <0.05.

Supplemental Figure 1. Change in beta coefficient for physiological variables from simple to the multiple linear regression model



A: fasting, <3 h vs. ≥8 h; B: fasting, 3–8 h vs. ≥8 h; C: inflammation, yes (CRP ≥5 mg/L) vs. no (CRP <5 mg/L); D: renal function, CKD stage 1 or 2 vs. normal; E: renal function, CKD stage 3–5 vs. normal; F: pregnancy, yes vs. no (women 20–49 y).

In each panel, *beta* × 100 (%) can be interpreted as the approximate percent change in the biomarker for a change in the respective covariate while holding any other variables in the model constant.

Sorted by class of biomarker (water-soluble, fat-soluble, phytoestrogens, iodine, hemoglobin adducts of acrylamide); arrows point in the direction of the change of the *beta* coefficient from simple linear regression (model 1) to multiple linear regression (model 2); reference line at zero; to simplify visual appearance, horizontal lines have been added.

Multiple linear regression model (panels A–E) controlled for age, sex, race-ethnicity, smoking, supplement use, fasting, inflammation, and renal function (and urinary creatinine for urine biomarkers). Multiple linear regression model for women 20–49 y (panel F) controlled for pregnancy in addition to the above mentioned variables.

4PA, 4-pyridoxic acid; B, whole blood; B-12, total cobalamin; CAR, carotenes; CKD, chronic kidney disease; CRP, C-reactive protein; DAZ, daidzein ; DMA, O-desmethylangolensin; EQU, equol; ETD, enterodiol; ETL, enterolactone; FOL, folate; GEN, genistein; HbAA, acrylamide hemoglobin adduct; HbGA, glycidamide hemoglobin adduct; MMA, methylmalonic acid; P, plasma; PLP, pyridoxal-5'-phosphate; S, serum; tHcy, total homocysteine; U, urine; uI, urine iodine; VIA, retinol; VIE, *alpha*-tocopherol; XAN, xanthophylls.

Supplemental Methods 2

Information on prevalence of abnormal biomarker concentrations by physiological variables

To provide additional information on whether the prevalence of abnormal biomarker concentrations is associated with common physiological variables (fasting, inflammation, renal function, and pregnancy), we conducted a descriptive bivariate analysis using cross-sectional data from the adult US population participating in NHANES 2003–2006 (women 20–49 y of age for iron status indicators and the pregnancy variable).

Methods

For biomarkers with accepted cutoff values for abnormal concentrations we calculated prevalence estimates by variable categories and used the Wald *Chi*-Square test to compare prevalences across categories (tests whether at least 1 of the estimates across the categories was significantly different). In addition, if the overall *Chi*-Square test was significant, we performed pairwise comparisons when there were more than 2 categories. We used the following cutoff values to calculate prevalence estimates: S-FOL[1] <2 µg/L (4.5 nmol/L) (1); RBC-FOL <95 µg/L (215 nmol/L) (1); PLP <20 nmol/L (2); B-12 <200 ng/L (148 pmol/L) (3); tHcy >13 µmol/L (4); MMA >271 nmol/L (5); VIC <11.4 µmol/L (6); VIA <20 µg/dL (0.7 µmol/L) (7); VIE <500 µg/dL (11.6 µmol/L) (8); 25OHD <30 nmol/L (9); FER <15 µg/L (33.7 pmol/L) (10); sTfR >4.4 mg/L (no generally accepted conversion factor available) (11); BI <0 mg/kg (12). We used the relative standard error (RSE) as a criterion for prevalence estimates of sufficient precision. Prevalence estimates were flagged if 30%≤RSE<40%. Estimates were not provided if they were associated with an RSE ≥40%. We did not present prevalence estimates for S-FOL, RBC-FOL, VIA, and VIE because they were very low (≤1%) and most estimates had to be suppressed or flagged because of high RSE.

[1] Abbreviations: 25OHD, 25-hydroxyvitamin D; B-12, total cobalamin; BI, body iron; FER, ferritin; MMA, methylmalonic acid; PLP, pyridoxal-5'-phosphate; RBC-FOL, red blood cell folate; RSE, relative standard error; S-FOL, serum folate; sTfR, soluble transferrin receptor; tHcy, total homocysteine; VIA, retinol; VIC, ascorbic acid; VIE, *alpha*-tocopherol.

Results

Fasting. Fasting was only associated with the prevalence of abnormal tHcy concentrations (**Supplemental Table 4**). We found a higher prevalence of abnormal tHcy concentrations in persons with intermediate fasting time (3–<8 h) compared to the other 2 fasting categories.

Inflammation. We found a higher prevalence of abnormal PLP, tHcy, VIC, and 25OHD concentrations in the presence of inflammation (**Supplemental Table 5**). We found a lower prevalence of abnormal FER and a higher prevalence of abnormal sTfR in the presence of inflammation, but no difference in prevalence for BI.

Renal function. Impaired renal function was significantly associated with the prevalence of abnormal biomarker concentrations for several indicators, but generally with small differences except for tHcy and MMA where the prevalence was highly different between normal renal function vs. stage 3–5 CKD: 5% vs. 37% and 5% vs. 28%, respectively (Supplemental Table 5).

Pregnancy. Consistent with observations made based on mean concentrations, we also found lower prevalence of abnormal MMA and higher prevalence of abnormal PLP, FER and BI in pregnant women (**Supplemental Table 6**).

Summary. Generally, our observations based on biomarker mean concentrations were consistent with those based on prevalence of abnormal concentrations; however, in some cases the "magnitude of the effect" seemed larger when cutoff values were considered. For example, we found prevalence rates that were at least double: for abnormal PLP and 25OHD in the presence of inflammation; for abnormal tHcy and MMA in the presence of impaired renal function; and for abnormal FER in pregnancy. This illustrates how sometimes the "effect" is different at the center and the tail of the distribution. On the other hand, because of very low prevalence estimates of abnormal folate, vitamins A and E concentrations, we could not assess whether there were differences by physiological variable categories.

Supplemental Table 4. Unadjusted prevalence of abnormal biomarker concentrations by fasting categories for adults ≥20y, NHANES 2003–2006[1,2,3,4,5]

Analyte (matrix)[6]	Overall	Fasting			*P*-value[7]
		<3 h	3–<8 h	≥8 h	
Water-soluble vitamins and related metabolites					
PLP (S)	12.9 (11.1 – 15.0)	12.0 (9.75 – 14.6)	14.0 (11.1 – 17.5)	13.1 (10.6 – 16.0)	0.53
B-12 (S)	2.49 (2.07 – 2.99)	2.50 (1.74 – 3.59)	2.52 (1.84 – 3.44)	2.47 (1.95 – 3.13)	0.99
tHcy (P)	7.71 (6.77 – 8.77)	6.75[a] (5.14 – 8.83)	9.81[b] (8.24 – 11.6)	7.13[a] (6.08 – 8.35)	0.0079
MMA (P)	7.51 (6.09 – 9.23)	14.1[8] (6.79 – 26.9)	7.67 (6.16 – 9.51)	6.93 (5.21 – 9.18)	0.10
VIC (S)	6.99 (5.74 – 8.47)	6.13 (4.29 – 8.69)	6.84 (4.93 – 9.41)	7.35 (6.08 – 8.86)	0.56
Fat-soluble vitamins					
25OHD (S)	9.24 (7.63 – 11.1)	8.87 (7.14 – 11.0)	8.71 (6.91 – 10.9)	9.59 (7.78 – 11.8)	0.42
Trace elements					
FER (S)	13.2 (11.8 – 14.8)	11.3 (8.53 – 14.7)	15.4 (11.7 – 20.1)	13.1 (11.0 – 15.5)	0.33
sTfR (S)	19.4 (17.3 – 21.7)	17.6 (12.8 – 23.8)	21.2 (17.0 – 26.2)	19.3 (16.8 – 22.1)	0.60
BI (S)	9.57 (8.57 – 10.7)	7.64 (5.24 – 11.0)	11.6 (8.41 – 15.8)	9.52 (8.07 – 11.2)	0.27

[1] Estimates represent percent prevalence and 95% CI. Within a variable, labeled percentages in a row without a common letter differ based on pairwise comparison, $P < 0.05$.

[2] Biomarker cutoff values used to calculate prevalence estimates: PLP (pyridoxal-5'-phosphate), <20 nmol/L; B-12 (total cobalamin), <200 ng/L (148 pmol/L); tHcy (total homocysteine), >13 µmol/L; MMA (methylmalonic acid), >271 nmol/L; VIC (ascorbic acid), <11.4 µmol/L; 25OHD (25-hydroxyvitamin D), <30 nmol/L; FER (ferritin), <15 µg/L (33.7 pmol/L); sTfR (soluble transferrin receptor), >4.4 mg/L; BI (body iron), <0 mg/kg.

[3] Prevalence estimates for S-FOL (serum folate), RBC-FOL (RBC folate), VIA (serum retinol), and VIE (serum *alpha*-tocopherol) were very low (≤1%) and are not presented because most estimates had to be suppressed or flagged because of high relative standard error (RSE).

[4] MMA data only available for NHANES 2003–2004; PLP data only available for NHANES 2005–2006.

[5] Sample sizes for each biomarker by covariate categories can be found in Supplemental Table 1.

[6] S, serum; P, plasma.

[7] *P*-value based on Wald *Chi*-Square test, which tests whether at least 1 of the estimates across the categories is significantly different.

[8] Estimate flagged because 30%≤RSE<40%.

Supplemental Table 5. Unadjusted prevalence of abnormal biomarker concentrations by inflammation and renal function categories for adults ≥20y, NHANES 2003–2006[1,2,3,4,5]

Analyte		Inflammation[6]				Renal function[7]			
(matrix)[8]	Overall	No	Yes	*P*-value[9]	Overall	Normal	CKD Stage 1 or 2	CKD Stage 3–5	*P*-value[9]
Water-soluble vitamins and related metabolites									
PLP (S)	12.9	9.47	24.2	<0.0001	12.7	11.6[a]	18.1[b]	16.0[a,b]	0.0147
	(11.1 – 15.0)	(7.61 – 11.7)	(20.8 – 28.0)		(10.9 – 14.7)	(9.32 – 14.3)	(14.6 – 22.4)	(12.3 – 20.7)	
B-12 (S)	2.49	2.46	2.57	0.81	2.44	2.34	2.29	3.57	0.09
	(2.07 – 2.99)	(1.97 – 3.08)	(1.93 – 3.42)		(2.02 – 2.94)	(1.87 – 2.93)	(1.49 – 3.50)	(2.56 – 4.95)	
tHcy (P)	7.68	7.13	9.52	0.0026	7.62	4.60[a]	8.16[b]	37.0[c]	<0.0001
	(6.74 – 8.74)	(6.19 – 8.21)	(8.06 – 11.2)		(6.67 – 8.69)	(3.70 – 5.69)	(6.16 – 10.7)	(32.2 – 42.2)	
MMA (P)	7.47	7.49	7.37	0.91	7.46	5.32[a]	8.20[b]	28.4[c]	<0.0001
	(6.04 – 9.20)	(5.98 – 9.35)	(5.34 – 10.1)		(6.06 – 9.17)	(4.29 – 6.59)	(5.67 – 11.7)	(22.9 – 34.5)	
VIC (S)	6.99	6.31	9.29	0.004	6.92	6.93[a,b]	8.24[a]	5.16[b]	0.0497
	(5.75 – 8.48)	(5.11 – 7.77)	(7.33 – 11.7)		(5.69 – 8.40)	(5.53 – 8.65)	(6.36 – 10.6)	(3.62 – 7.31)	
Fat-soluble vitamins									
25OHD (S)	9.18	7.74	14.0	<0.0001	9.13	8.56[a]	13.1[b]	9.82[a,b]	0.0008
	(7.59 – 11.1)	(6.37 – 9.38)	(11.4 – 17.1)		(7.55 – 11.0)	(6.96 – 10.5)	(10.4 – 16.2)	(7.46 – 12.8)	
Trace elements									
FER (S)	13.2	14.4	10.5	0.0268	13.2	12.3[a]	22.2[b]	NR[10]	0.0411
	(11.8 – 14.8)	(12.6 – 16.3)	(8.11 – 13.4)		(11.8 – 14.8)	(10.5 – 14.3)	(16.4 – 29.4)		
sTfR (S)	19.4	17.8	23.1	0.023	19.3	18.3[a]	26.9[b]		0.0181
	(17.3 – 21.7)	(15.4 – 20.5)	(19.5 – 27.1)		(17.2 – 21.6)	(16.1 – 20.9)	(21.5 – 33.2)	NR	
BI (S)	9.57	10.2	8.08	0.16	9.57	8.99	15.2	NR	0.22
	(8.57 – 10.7)	(8.91 – 11.6)	(6.02 – 10.8)		(8.59 – 10.6)	(7.61 – 10.6)	(10.1 – 22.2)		

[1] Estimates represent weighted percent prevalence and 95% CI (by various Mobile Examination Center weights). Within a variable, labeled percentages in a row without a common letter differ based on pairwise comparison, $P < 0.05$.

[2] Biomarker cutoff values used to calculate prevalence estimates: PLP (pyridoxal-5'-phosphate), <20 nmol/L; B-12 (total cobalamin), <200 ng/L (148 pmol/L); tHcy (total homocysteine), >13 µmol/L; MMA (methylmalonic acid), >271 nmol/L; VIC (ascorbic acid), <11.4 µmol/L; 25OHD (25-hydroxyvitamin D), <30 nmol/L; FER (ferritin), <15 µg/L (33.7 pmol/L); sTfR (soluble transferrin receptor), >4.4 mg/L; BI (body iron), <0 mg/kg.

[3] Prevalence estimates for S-FOL (serum folate), RBC-FOL (RBC folate), VIA (serum retinol), and VIE (serum *alpha*-tocopherol) were very low (≤1%) and are not presented because most estimates had to be suppressed or flagged because of high relative standard error (RSE).

[4] MMA data only available for NHANES 2003–2004; PLP data only available for NHANES 2005–2006.

[5] Sample sizes for each biomarker by covariate categories can be found in Supplemental Table 1.

[6] S, serum; P, plasma.

[7] C-reactive protein (CRP, mg/L) was used to assess inflammation status (<5, no inflammation; ≥5, inflammation).

[8] Estimated glomerular filtration rate (eGFR) was used to assess renal function; normal was defined as eGFR ≥60 mL/(min·1.73 m^2) and absence of albuminuria; stage 1 or 2 chronic kidney disease (CKD) was defined as eGFR ≥60 mL/(min·1.73 m^2) and presence of albuminuria; stage 3–5 CKD was defined as eGFR <60 mL/(min·1.73 m^2).

[9] *P*-value based on Wald *Chi*-Square test, which tests whether at least 1 of the estimates across the categories is significantly different.

[10] Not reported due to small sample size (*n* <42).

Supplemental Table 6. Unadjusted prevalence of abnormal biomarker concentrations by pregnancy categories for women 20–49 y, NHANES 2003–2006[1,2,3,4,5]

Analyte (matrix)[6]	Overall	Pregnant		*P*-value[7]
		No	Yes	
Water-soluble vitamins and related metabolites				
PLP (S)	15.8 (12.7 – 19.4)	15.2 (12.3 – 18.7)	23.3 (14.9 – 34.5)	0.05
B-12 (S)	2.50 (1.77– 3.51)	2.41 (1.62 – 3.56)	3.81 (2.39 – 6.03)	0.23
tHcy (P)	2.55 (1.77 – 3.66)	2.72 (1.89 – 3.89)	NR[8]	<0.0001
MMA (P)	3.79 (2.39 – 5.95)	3.93 (2.47 – 6.19)	1.18 (0.66 – 2.11)	0.0064
VIC (S)	6.41 (4.85 – 8.43)	6.79 (5.16 – 8.88)	NR	<0.0001
Fat-soluble vitamins				
25OHD (S)	11.3 (9.15 – 14.0)	11.5 (9.25 – 14.2)	8.77 (5.17 – 14.5)	0.24
Trace elements				
FER (S)	13.3 (11.9 – 14.8)	12.5 (11.1 – 14.1)	25.3 (19.9 – 31.6)	0.0003
sTfR (S)	19.4 (17.2 – 21.6)	19.5 (17.2 – 21.9)	17.7 (13.3 – 23.0)	0.52
BI (S)	9.54 (8.54 – 10.6)	9.13 (8.03 – 10.4)	16.1 (12.3 – 20.8)	0.0058

[1] Estimates represent weighted percent prevalence and 95% CI (by various Mobile Examination Center weights).

[2] Biomarker cutoff values used to calculate prevalence estimates: PLP (pyridoxal-5'-phosphate), <20 nmol/L; B-12 (total cobalamin), <200 ng/L (148 pmol/L); tHcy (total homocysteine), >13 µmol/L; MMA (methylmalonic acid), >271 nmol/L; VIC (ascorbic acid), <11.4 µmol/L; 25OHD (25-hydroxyvitamin D), <30 nmol/L; FER (ferritin), <15 µg/L (33.7 pmol/L); sTfR (soluble transferrin receptor), >4.4 mg/L; BI (body iron), <0 mg/kg.

[3] Prevalence estimates for S-FOL (serum folate), RBC-FOL (RBC folate), VIA (serum retinol), and VIE (serum *alpha*-tocopherol) were very low (≤1%) and are not presented because most estimates had to be suppressed or flagged because of high relative standard error (RSE).

[4] MMA data only available for NHANES 2003–2004; PLP data only available for NHANES 2005–2006.

[5] Sample sizes for each biomarker by covariate category can be found in Supplemental Table 1.

[6] S, serum; P, plasma.

[7] *P*-value based on Wald *Chi*-Square test, which tests whether at least 1 of the estimates across the categories is significantly different.

[8] Not reported due to estimate suppressed because RSE ≥40%.

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