

The Journal of Nutrition Supplement: The Controversial Role of Macronutrient Composition in Diabetes and Related Disorders

Differing Statistical Approaches Affect the Relation between Egg Consumption, Adiposity, and Cardiovascular Risk Factors in Adults^{1–4}

Theresa A Nicklas,5* Carol E O'Neil,6 and Victor L Fulgoni III7

⁵Department of Pediatrics, Baylor College of Medicine, USDA/Agricultural Research Service Children's Nutrition Research Center, Houston, TX; ⁶School of Nutrition and Food Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA; and ⁷Nutrition Impact LLC, Battle Creek, MI

Abstract

Background: Associations between food patterns and adiposity are poorly understood.

Objective: Two statistical approaches were used to examine the potential association between egg consumption and adiposity. **Methods:** Participants (n = 18,987) aged ≥ 19 y were from the 2001–2008 NHANES who provided 24-h diet recall data, body mass index (BMI) and waist circumference (WC)–determined adiposity measures, and blood pressure, circulating insulin, glucose, and lipid concentrations were considered cardiovascular risk factors (CVRFs). Covariate-adjusted least-squares means \pm SEs were generated. **Results:** The first statistical approach categorized participants into egg consumers or nonconsumers. Consumers had higher mean BMI (in kg/m²; 28.7 \pm 0.19; P = 0.006) and WC (98.2 \pm 0.43 cm; P = 0.002) than did nonconsumers (28.2 \pm 0.10 and 96.9 \pm 0.23 cm, respectively). Second, cluster analysis identified 8 distinct egg consumption patterns (explaining 39.5% of the variance in percentage of energy within the food categories). Only 2 egg patterns [egg/meat, poultry, fish (MPF)/grains/vegetables and egg/ MPF/grains], consumed by $\leq 2\%$ of the population, drove the association (compared with the no-egg pattern) between egg consumption and BMI and WC. Another analysis controlled for the standard covariates and the other food groups consumed with eggs in those 2 egg patterns. Only the egg/MPF/other-grains pattern remained associated with BMI and WC (both $P \leq 0.0063$). The pattern analyses identified associations between an egg pattern (egg/MPF/other grains/potatoes/other beverages) and diastolic blood pressure (DBP) and serum LDL cholesterol (both $P \leq 0.0063$). A final analysis was conducted by adding percentage of energy from fast foods and medication use for diabetes to the covariates. The association between the egg/MPF/grains pattern and BMI and the egg/MPF/potatoes/other beverages and DBP and LDL cholesterol disappeared.

Conclusions: Care needs to be taken with data interpretation of diet and health risk factors and the choice of statistical analyses and covariates used in the analyses because these studies are typically used to generate hypotheses. Additional studies are needed to better understand these relations. *J Nutr* 2015;145:170S–6S.

Keywords: adults, cardiovascular risk factors, egg consumption, statistical approaches, food patterns

Introduction

Nutritional epidemiology has involved research that 1) examines the role of nutrition in the etiology of disease, 2) monitors the nutritional status of populations, 3) develops and evaluates interventions designed to achieve and maintain healthier eating patterns among populations, and 4) examines the relation between nutrition and other behaviors on human health and disease. Until recently, nutritional epidemiology research has largely focused on the "traditional" or "reductionist" approach, with investigations into diet and health addressing the effects of

NUTRITION

OF

JOURNAL

¹ Presented at the conference "Advances and Controversies in Clinical Nutrition," held in Washington, DC, 5–7 December 2013. The conference was sponsored by the Egg Nutrition Center and the American Egg Board. The Supplement Coordinator for this supplement was Tia M Rains. Supplement Coordinator disclosures: Tia M Rains is an employee of The Egg Nutrition Center/American Egg Board. The Session Chair was Mitch Kanter. This supplement is the responsibility of the Guest Editor to whom the Editor of *The Journal of Nutrition* has delegated supervision of both technical conformity to the published regulations of *The Journal of Nutrition* and general oversight of the scientific merit of each article. The Guest Editor for this supplement was Harry D Dawson. Guest Editor disclosure: no conflicts of interest. Publication costs for this supplement were defraved in part by the payment of page charges. This publication must therefore be hereby marked "advertisement" in accordance with 18 USC section 1734 solely to indicate this fact. The opinions expressed in this publication are those of the authors and are not attributable to the sponsors or the publisher, Editor, or Editorial Board of *The Journal of Nutrition*.

² Supported by the USDA/Agricultural Research Service through specific cooperative agreement 58-6250-0-008. Partial support was received from the Egg Nutrition Board and from the USDA Hatch Project LAB 93951.

 $^{^{\}rm 3}$ Author disclosures: TA Nicklas, CE O'Neil, and VL Fulgoni III, no conflicts of interest.

⁴ The contents of this publication do not necessarily reflect the views or policies of the USDA, nor does mention of trade names, commercial products, or organizations imply endorsement from the U.S. government. The funding agencies had no input into the study design or interpretation of the data. *To whom correspondence should be addressed. E-mail: tnicklas@bcm.edu.

single nutrients or foods on health outcomes (1). Although this type of research has greatly advanced nutritional knowledge and generated valuable hypotheses for future studies and suggestions for public policy, it has also, at times, resulted in inconsistent relations between diet and chronic diseases. This is not surprising given the inherent complexities of these relations. Individuals do not consume single nutrients or foods but, rather, meals consisting of a variety of foods with complex combinations of foods and nutrients that are likely to be interactive or synergistic (2). It is unclear from the previous research whether observed relations between diet and health outcomes are due solely to individual foods or nutrients or whether they indirectly reflect other nutrients and foods consumed together in a dietary pattern. Thus, studies at the nutrient or food level often reported null associations or inconsistent findings. Moreover, lack of attention to important confounders, including other components of diet and behavioral variables, are clear limitations of earlier research.

In the past decade, there has been a paradigm shift in nutritional epidemiology to examine associations between diet and health. Dietary patterns research has been conducted as a more comprehensive way to characterize complex dietary exposures (3–10). There is a growing body of literature that has emerged to explore and assess the impact of dietary patterns on health outcomes. The case for using theoretically driven or empirically driven methods for identifying dietary patterns imposes challenges in data interpretation (5, 8). Limitations of data collection and analytic issues, such as the interrelation and correlation of foods within specific patterns, suggest the need for further research. Inconsistent findings are being found in the use of the dietary pattern approach with assessing the effect on health outcomes, which may reflect the statistical approach and the covariates used and assumptions made a priori in the analyses.

Health issues related to egg consumption have been the subject of discussion primarily as it related to coronary heart disease (11, 12), stroke (12), and individual cardiovascular risk factors (CVRFs)⁸ (13)—namely, serum total cholesterol (14, 15), LDL cholesterol (14, 15), type 2 diabetes (16), and metabolic syndrome (17). There are no population studies in a recent nationally representative population assessing the relation between consumption of eggs with weight status. The contribution of egg consumption to health outcomes is poorly understood, and the findings are inconclusive because of methodologic and analytic flaws in approaches used in the past.

The goal of this study was to use 2 statistical approaches to examine the association between egg consumption, adiposity, and CVRFs in adults by using data from the 2001–2008 NHANES. Both limitations and advantages to each statistical approach are discussed along with recommendations for future research.

Methods

Study participants. Data from adults (n = 18,988) aged ≥ 19 y participating in the 2001–2008 NHANES were combined to increase sample size (18). Analyses included only individuals with dietary records deemed complete and reliable by the National Center for Health Statistics staff; women who were pregnant or lactating were excluded from analyses. The NHANES has strict protocols and procedures in place that ensure confidentiality and protect individual participants from identification using federal laws (19). This was a secondary data analysis

with a lack of personal identifiers; therefore, this study was exempted by the Baylor College of Medicine Institutional Review Board (20, 21). Physical activity was determined by using a questionnaire (22) that assessed sedentary, moderate, and vigorous physical activity in a typical week.

Intake data were obtained from What We Eat in America, which used in-person 24-h dietary recalls with an automated multiple-pass method (23–25). A single 24-h dietary recall was collected and released from each participant in 2001–2002; however, beginning in 2003– 2004, 2 d of intake were collected and released. To ensure consistency, only the data from the in-person interview (first recall) were used for this study. Detailed descriptions of the dietary interview methods are available (26). Intake of alcohol (g/d) was determined from the 24-h dietary recall.

Defining egg consumption and patterns of egg consumption. Egg consumption was defined as consumption of foods primary composed of whole eggs (eggs used in baking were not included). The USDA Food and Nutrient Databases for Dietary Studies (versions 2.0, 3.0, and 4.0) were used to identify 73 food codes that contained whole eggs (food codes 31101010–32202200).

Energy and nutrient intakes came directly from the total nutrient intake files. Food group equivalent intakes were determined by using MyPyramid Equivalents Database versions 1.0 (27) and 2.0 (28) and when necessary hand matching to similar foods in subsequent NHANES releases. Intakes for 2005-2008 NHANES were then determined by multiplying daily intake by food group equivalent composition for each food and summing over the day. Diet quality (29-31) was determined by the Healthy Eating Index-2005 (HEI-2005) score, which, in turn, was determined by using the downloadable code from the Center for Nutrition Policy and Promotion website (32). Briefly, HEI-2005 was designed to evaluate 9 major MyPyramid food groups and 3 nutrients. The 12 HEI-2005 components are summed for a total possible score of 100 points. Higher scores indicate greater dietary intake, except for SFAs, sodium, and solid fats/alcohol/added sugars, in which higher subcomponent scores indicate lower intake. Scores are adjusted on an energy-density basis (per 1000 kcal), which allowed for characterization of diet quality while controlling for diet quantity.

Physiologic measures. Height, weight, and waist circumference (WC) were obtained according to the NHANES Anthropometry Procedures Manual (33). The manual provides information about equipment, calibration, methods, quality control, and survey procedures. BMI was calculated as body weight (in kg) divided by height (in m) squared (34).

CVRFs. Health indices that were evaluated included BMI, WC, diastolic blood pressure (DBP) and systolic blood pressure, serum HDL cholesterol, serum LDL cholesterol, serum TGs, plasma glucose, plasma insulin, and serum C-reactive protein (CRP) concentrations. Three or 4 readings for systolic blood pressure and DBP were recorded in the NHANES; an average from each set of readings was used in this study. Venous blood was drawn in the mobile examination center and total HDL-cholesterol concentrations were determined in nonfasted individuals and LDL-cholesterol, TG, blood glucose, and insulin concentrations were determined in only fasted subjects; thus, not all individuals had laboratory values for all tests. Serum total cholesterol and plasma glucose were measured spectrophotometrically by using a series of enzymatic reactions (35). Serum LDL cholesterol was calculated according to the Friedewald equation and was reported only for fasting participants (35). Serum HDL cholesterol was measured by using enzymatic reactions in conjunction with the heparin-manganese precipitation method or a direct immunoassay technique (35). Serum insulin was measured by RIA (35), immunoenzymometric assay, or ELISA (35). High-sensitivity serum CRP was measured by particle-enhanced immunoassay with latex-enhanced nephelometry (35). An index of insulin resistance and sensitivity was calculated according to the HOMA formula = [fasting plasma or serum glucose (mmol/L) × fasting plasma or serum insulin (pmol/L)]/ 405 (36).

⁸ Abbreviations used: CRP, C-reactive protein; CVRF, cardiovascular risk factor; DBP, diastolic blood pressure; HEI-2005, Healthy Eating Index–2005; MPF, meat/poultry/fish; WC, waist circumference.

The Prescription Medication questionnaire was asked, in the home, by trained interviewers with the use of the Computer-Assisted Personal Interviewing system. Participants >16 y of age answered for themselves; a proxy provided information for survey participants who were ≤ 16 y of age and for individuals who could not answer the questions themselves. During the household interview, the participants were asked if they had taken medications in the past month for which they needed a prescription. Those who answered "yes" were asked to show the interviewer the medication containers of all the products used. For each medication reported, the interviewer entered the product's complete name from the container in a computer. If no container was available, the interviewer asked the participant to verbally report the name of the medication.

Statistical analyses. Two statistical approaches were used to assess the relation between egg consumption, adiposity, and CVRFs.

- Approach 1. Least-squares means \pm SEs were calculated by using PROC REGRESS of SUDAAN (SAS Institute) for energy, nutrients, food groups, diet quality, and CVRFs for consumers and nonconsumers of whole eggs. Statistical differences for variables of interest among egg consumers were determined via *t* test compared with the nonconsumers of eggs. A probability of *P* < 0.05 was considered significant. Covariates were as follows: age, gender, race/ethnicity, poverty-income ratio grouped into 3 categories (<1.25, 1.25–3.49, and >3.49), physical activity level (sedentary, moderate, and vigorous), smoking status (yes or no), alcohol intake (g/d), and energy intake (kcal). The HEI-2005 and total energy intake were not controlled for energy intake, because many of the target intakes in HEI-2005 are based on per-1000 kcal, which controls for energy.
- Approach 2. Egg consumption patterns were identified by using SAS 9.2 PROC CLUSTER (SAS Institute) with the use of NHANES 2001–2008 dietary day 1 weights for all analyses. Patterns of egg consumption were established by taking the food codes and separating them into 20 food groupings. All food codes fit in only one of the food groupings. For each participant, the percentage of egg consumption kilocalories from each of the 20 food groupings was determined.
- Egg consumption patterns were defined by the percentage of energy provided by each food group (only foods that contributed $\geq 5\%$ of kcal were reported). The use of this method resulted in 8 readily identifiable egg consumption patterns (including no egg consumption), accounting for ~39.5% of the variance in total energy intake. With egg consumption patterns identified, each participant was placed into one egg consumption pattern on the basis of their food intake. Least-squares means \pm SEs calculated by using PROC REGRESS of SUDAAN for dietary intake, diet quality (HEI-2005), and physiologic measures were determined for participants in each egg consumption pattern. Covariates were as follows: age, gender, race/ethnicity, poverty-income ratio grouped into 3 categories (<1.25, 1.25–3.49, and >3.49), physical activity level (sedentary, moderate, and vigorous), smoking status (yes or no), alcohol intake (g/d), and energy intake for nutrient-related variables (not for energy intake itself, HEI-2005, or physiologic measures). For non-weight-related physiologic variables, BMI was also included as a covariate. Statistical differences for variables of interest were determined via t test comparing with the no-egg-consumption group. A Bonferroni correction was applied for the multiple comparisons (0.05/8 clusters), so the effective *P* value was P < 0.0063.
- Food group equivalents that made up 3 egg consumption patterns that had differences from the no-egg pattern [meat/poultry/fish (MPF), other grains, potatoes, and other beverages; MPF, other grains, and vegetables; and MPF and other grains, respectively] were also included as covariates in the analyses (the no-egg-consumption group was also adjusted for these additional covariates). In addition, further exploration of other possible confounding variables identified medication use and percentage of energy from fast foods as possible mediators of relations initially reported. Thus, these variables were also added to the models as additional covariates.

Results

THE JOURNAL OF NUTRITION

 \geq

Demographic and lifestyle characteristics of the sample The total sample consisted of 18,988 adults aged \geq 19 y (51% women, 72% non-Hispanic whites, 11% non-Hispanic blacks, 12% Mexican-American/Hispanic) (**Table 1**). Sixty-three percent reported sedentary to moderate physical activity, 25% reported smoking, and 27% reported intake of alcohol. Overall, diet quality was poor, reflected in a HEI score of 51 of 100 possible points.

Statistical approaches

Approach 1. The association between egg consumption and CVRFs is presented in **Table 2.** Adults in the egg consumption group had significantly higher BMI (P = 0.0060) and WC (P = 0.0018) than did nonconsumers.

Approach 2. Table 3 describes the 8 egg consumption patterns (includes the no-egg-consumption pattern). Eighty percent of adults reported not consuming eggs in the 24-h recall. For the no-egg-consumption group, the main energy sources were MPF/ other grains/other beverages. Eight percent had an egg consumption pattern that included MPF/other grains/potatoes/fruit/vegetables. Less than 3% consumed 1 of the 6 remaining egg consumption patterns.

The association between the egg consumption patterns and CVRFs is presented in **Table 4**. Adults in egg consumption patterns 4 (egg/MPF/other grains/vegetables) and 7 (egg with meat/MPF/other grains) had significantly higher BMI and WC than did the adults in the no-egg-consumption pattern. Data suggest that the association between egg consumption and BMI and WC found by using the first statistical approach was driven by these 2 specific egg consumption patterns. The use of this second statistical approach resulted in a new finding. Adults in egg consumption pattern 3 (egg/MPF/other grains/potatoes/other beverages) had significantly higher DBP and LDL cholesterol compared with adults in the no-egg-consumption pattern.

The association between egg consumption patterns and CVRFs, adjusted for the standard covariates in addition to the food groups consumed with eggs, in patterns 3, 4, and 7 is presented in Table 5. After adjusting for the associated food groups in these 3 egg consumption patterns (patterns 3, 4, and 7), the association between egg consumption pattern 4 was no longer associated with BMI and WC compared with the no-egg-

TABLE 1 Demographic and lifestyle characteristics of adults aged \geq 19 y (*n* = 18,988) participating in the NHANES 2001–2008¹

Demographic variables	Total weighted sample
Gender, %	
Male	49.2 ± 0.39
Female	50.9 ± 0.39
Race/ethnicity, %	
Non-Hispanic white	72.1 ± 1.58
Non-Hispanic black	11.3 ± 0.94
Mexican American/Hispanic	11.7 ± 1.05
Other	4.9 ± 0.38
Mean poverty-income ratio	3.0 ± 0.04
Physical activity intensity, %	
Sedentary	30.7 ± 0.04
Moderate	32.7 ± 0.53
Vigorous	36.7 ± 0.81
Current smoker, %	24.8 ± 0.63
Alcohol consumer, %	26.6 ± 0.82
Healthy Eating Index score	51.2 ± 0.27

¹ Values are least-squares means ± SEs.

		Egg consumption			
		Yes		No	
Cardiovascular risk factors	n	Value	п	Value	Р
BMI, ² kg/m ²	4301	28.7 ± 0.19	14,688	28.2 ± 0.10	0.0060
Waist circumference, ² cm	4301	98.2 ± 0.43	14,688	96.9 ± 0.23	0.0018
Diastolic blood pressure, ³ kPa	4301	9.59 ± 0.04	14,688	9.51 ± 0.03	0.19
Systolic blood pressure, ³ kPa	4301	16.4 ± 0.05	14,688	16.4 ± 0.03	0.62
Serum C-reactive protein, ³ mg/L	4301	4.10 ± 0.10	14,688	4.10 ± 0.10	0.73
Serum LDL cholesterol, ³ mmol/L	2086	3.06 ± 0.03	7202	3.02 ± 0.02	0.19
Serum HDL cholesterol, ³ mmol/L	4301	1.37 ± 0.01	14,688	1.36 ± 0.004	0.12
Serum TGs, ³ mmol/L	2086	1.57 ± 0.06	7202	1.64 ± 0.03	0.24
Plasma glucose, ³ mmol/L	2086	5.77 ± 0.04	7202	5.69 ± 0.02	0.13
Plasma insulin, ³ pmol/L	2086	69.8 ± 1.92	7202	67.3 ± 1.14	0.16
HOMA-IR ³	2086	3.18 ± 0.11	7202	2.98 ± 0.06	0.09

 1 Values are least-squares means \pm SEs. Systolic/diastolic blood pressure, kPa = mm Hg \times 0.1333; C-reactive protein, mg/L = mg/dL \times 10; HDL cholesterol, mmol/L = mg/dL \times 0.002586; LDL cholesterol, mmol/L = mg/dL \times 0.002586 \times mg/dL; TGs, mmol/L = mg/dL \times 0.01129; glucose, mmol/L = mg/dL \times 0.05551; insulin, pmol/L = mU/L \times 6.

 $^{\rm 2}$ Adjusted for ethnicity, age, poverty-income ratio, physical activity, smoking, and alcohol.

³ Adjusted for ethnicity, age, poverty-income ratio, physical activity, smoking, alcohol, and BMI.

consumption pattern group. However, the relation between egg consumption pattern 7 and BMI and WC and egg consumption pattern 3 and DBP and LDL cholesterol remained significant after controlling for the other food groups in those 2 egg consumption patterns.

Demographic and lifestyle characteristics of egg consumption patterns 3 and 7 are presented in **Table 6**. Compared with the no-egg-consumption pattern group, adults in egg pattern 3 were younger and had a higher percentage of non-Hispanic blacks, smokers, and alcohol consumers (alcohol intake was also significantly higher) and a lower percentage of females and adults with a BMI (kg/m²) >40 or taking diabetes medication. Adults in egg consumption pattern 7 consumed significantly less alcohol and a significantly higher percentage of energy from fast food than did adults in the no-egg-consumption group.

Compared with the nonconsumers, a final analysis was conducted with egg consumption patterns 3 and 7, controlling for standard covariates, the associated food groups, percentage of energy from fast food, and taking medication for diabetes (e.g., insulin or oral hypoglycemics). The relation between egg consumption pattern 7 was no longer associated with BMI; yet, the association with WC remained. The relation between egg consumption pattern 3 and DBP and LDL cholesterol no longer remained significant (data not shown).

Discussion

NUTRITION

THE JOURNAL OF

 \geq

Results from this study varied considerably depending on what statistical approach was used in the analyses. Evaluating all statistical approaches used in this study, it appears that the association found between egg consumption and BMI and WC (approach 1) was specifically driven by 2 of 8 egg consumption patterns: egg/MPF/grains/vegetables (pattern 4) and egg/MPF/

						Whole eggs, 9	% of energy	Grain snacks/		Whole fruit		Vegetables	
Egg clusters	Cluster number	u	Population, %	Other dairy, % of energy	MPF, % of energy	Without meat	With Meat	desserts, % of energy	Other grains, % of energy	and juices, % of energy	Potatoes, % of energy	and legumes, % of energy	Other beverages, % of energy
No egg	0	14,686	80.2	5.7	19.6	0.0	0.0	9.3	23.1	4.6	4.5	6.7	11.2
Egg/MPF/grains	-	527	2.1	3.9	22.7	7.3	0.2	5.0	32.8	4.5	2.7	4.7	5.9
Egg/other grains	2	533	2.4	4.7	4.7	7.5	2.4	5.4	48.0	4.8	4.1	4.2	4.9
Egg/MPF/other grains/potatoes/	ę	646	2.9	3.3	15.9	7.1	2.0	3.0	18.1	2.3	6.2	3.7	30.2
other beverages													
Egg/MPF/other grains/vegetables	4	465	2.1	4.0	43.8	7.7	0.5	4.9	11.5	2.9	3.0	5.6	6.1
Egg/MPF/other grains/desserts	ŋ	291	1.4	4.7	9.6	7.4	0.3	32.3	14.1	4.9	3.0	4.8	6.6
Egg/MPF/other grains/potatoes/	9	1575	7.8	7.7	16.2	8.2	0.8	7.2	14.8	5.6	6.2	9.0	7.0
fruit/vegetables													
Egg with meat/MPF/other grains	7	264	1.2	6.4	14.4	0.9	23.4	7.9	13.2	4.2	5.4	4.2	8.3

small. MPF, meat/poultry/fish

/ery

	Egg pattern							
				3: Egg/MPF/		_	6: Egg/MPF/other	
			2. En /	other grains/	4: Egg/MPF/	5: Egg/MPF/	grains/potatoes/	7. [
Cardiovascular risk	0: No eaas ²	1: Egg/IVIPF/ grains	2: Egg/other grains	potatoes/other	other grains/ venetables	other grains/ desserts	vegetables	7: Egg with meat/MPE/
factors	(<i>n</i> = 14,686)	(<i>n</i> = 527)	(<i>n</i> = 533)	(<i>n</i> = 646)	(<i>n</i> = 465)	(<i>n</i> = 291)	(<i>n</i> = 1575)	other grains ($n = 264$)
BMI, ³ kg/m ²	28.2 ± 0.10	29.0 ± 0.37	28.8 ± 0.38	28.2 ± 0.29	30.4 ± 0.55*	27.6 ± 0.41	28.3 ± 0.24	30.7 ± 0.81*
Waist circumference, ³ cm	96.9 ± 0.23	98.9 ± 0.96	98.2 ± 0.97	97.2 ± 0.73	$101.7 \pm 1.19^*$	95.5 ± 1.03	97.3 ± 0.59	101.9 ± 1.56*
Diastolic blood Pressure, ⁴ kPa	9.54 ± 0.03	9.56 ± 0.09	9.54 ± 0.10	$9.93 \pm 0.10^*$	9.61 ± 0.13	9.45 ± 0.10	9.53 ± 0.05	9.51 ± 0.10
Systolic blood pressure, ⁴ kPa	16.4 ± 0.03	16.4 ± 0.12	16.4 ± 0.15	16.6 ± 0.15	16.6 ± 0.15	16.3 ± 0.14	16.3 ± 0.08	16.1 ± 0.14
Serum C-reactive protein, ⁴ mg/L	4.10 ± 0.10	5.00 ± 0.50	3.50 ± 0.30	4.40 ± 0.40	4.20 ± 0.50	4.30 ± 0.60	4.00 ± 0.20	3.90 ± 0.40
Serum LDL cholesterol, ⁴ mmol/L	3.02 ± 0.02	3.09 ± 0.09	2.88 ± 0.07	$3.22 \pm 0.07^*$	2.99 ± 0.06	3.13 ± 0.19	2.99 ± 0.04	3.27 ± 0.10
Serum HDL cholesterol, ⁴ mmol/L	1.36 ± 0.004	1.39 ± 0.02	1.39 ± 0.03	1.35 ± 0.02	1.37 ± 0.03	1.34 ± 0.03	1.39 ± 0.01	1.35 ± 0.02
Serum TGs, ⁴ mmol/L	1.64 ± 0.03	1.54 ± 0.09	1.56 ± 0.08	1.50 ± 0.08	1.67 ± 0.12	1.62 ± 0.10	1.57 ± 0.12	1.50 ± 0.10
Plasma glucose, ⁴ mmol/L	5.69 ± 0.02	5.64 ± 0.16	5.85 ± 0.19	5.73 ± 0.07	5.97 ± 0.16	5.62 ± 0.11	5.81 ± 0.06	5.53 ± 0.07
Plasma insulin, ³ pmol/L	67.3 ± 1.14	74.6 ± 10.9	70.6 ± 4.32	76.1 ± 5.64	65.0 ± 5.52	72.0 ± 4.68	67.1 ± 2.28	68.3 ± 4.14
HOMA-IR ³	2.98 ± 0.06	3.21 ± 0.46	3.16 ± 0.24	3.44 ± 0.31	3.15 ± 0.36	3.25 ± 0.28	3.13 ± 0.17	2.92 ± 0.20

¹ Values are least-squares means \pm SEs. *Different from no eggs, $P \leq 0.0063$ (with Bonferroni correction). Systolic/diastolic blood pressure, kPa = mm Hg \times 0.1333; C-reactive protein, mg/L = mg/dL \times 10; HDL cholesterol, mmol/L = mg/dL \times 0.002586; LDL cholesterol, mmol/L = mg/dL \times 0.002586 \times mg/dL; TGs, mmol/L = mg/dL \times 0.01129; glucose, mmol/L = mg/dL \times 0.05551; insulin, pmol/L = mU/L \times 6. MPF, meat/poultry/fish.

² Reference group

NUTRITION

THE JOURNAL OF

 \geq

³ Adjusted for ethnicity, age, poverty-income ratio, physical activity, smoking, and alcohol.

⁴ Adjusted for ethnicity, age, poverty-income ratio, physical activity, smoking, alcohol, and BMI.

grains (pattern 7). Moreover, with pattern analysis, a new finding emerged: egg consumption pattern 3 (egg/MPF/grains/ potatoes/beverages) was associated with DBP and LDL cholesterol. This raised the question of whether these associations between the 3 egg consumption patterns and the selected CVRFs were due to the other food groups consumed with eggs rather than specifically to egg consumption.

Additional analyses, controlling for the food groups in egg consumption patterns 3, 4, and 7, were conducted. Thus, with these analyses, the association between egg consumption pattern 4 and BMI and WC disappeared. However, the relations between egg consumption patterns 3 (with DBP and LDL cholesterol) and 7 (with BMI and WC) continued to be significant compared with the no-egg-consumption pattern.

A striking result was the significantly higher percentage of energy from fast food in egg consumption pattern 7 compared with the no-egg-consumption pattern. Given that egg consumption pattern 7 was associated with BMI and WC, it was possible that the higher percentage of energy from fast food mediated that association. Thus, when the percentage of energy from fast food was controlled for in the analyses, the relation between egg consumption pattern 7 and BMI disappeared, but the relation

TABLE 5 Cardiovascular risk factors in adults \geq 19 y (*n* = 18,988) by egg pattern: NHANES 2001–2008¹

Egg pattern					
Addition grains	nally adjusted for MPF/ s/potatoes/beverages	Additionally grain	/ adjusted for MPF/ s/vegetables	Additiona MPF/0	Ily adjusted for other grains
0: No eggs (<i>n</i> = 14,686)	3: Egg/MPF/grains/potatoes/ beverages (n = 646)	0: No eggs (<i>n</i> = 14,686)	4: Egg/MPF/grains/ vegetables (<i>n</i> = 465)	0: No eggs (<i>n</i> = 14,686)	7: Egg/MPF/grains (<i>n</i> = 264)
28.2 ± 0.10	28.3 ± 0.29	28.2 ± 0.10	29.7 ± 0.58	28.2 ± 0.10	30.7 ± 0.81*
96.9 ± 0.23	97.2 ± 0.76	96.9 ± 0.23	101.48 ± 1.28	96.9 ± 0.23	101.9 ± 1.57*
9.54 ± 0.03	9.90 ± 0.10	9.54 ± 0.03	9.58 ± 0.13	9.54 ± 0.03	9.51 ± 0.10
16.4 ± 0.03	16.5 ± 0.14	16.4 ± 0.03	16.6 ± 0.15	16.4 ± 0.03	16.1 ± 0.14
4.10 ± 0.10	4.20 ± 0.40	4.10 ± 0.10	4.30 ± 0.50	4.10 ± 0.10	3.80 ± 0.40
1.36 ± 0.004	1.38 ± 0.02	1.36 ± 0.004	1.36 ± 0.03	1.36 ± 0.005	1.35 ± 0.02
3.02 ± 0.02	3.22 ± 0.07	3.02 ± 0.02	2.98 ± 0.06	3.02 ± 0.02	3.28 ± 0.10
1.64 ± 0.03	1.46 ± 0.09	1.64 ± 0.03	1.66 ± 0.11	1.64 ± 0.03	1.50 ± 0.10
5.69 ± 0.02	5.79 ± 0.07	5.69 ± 0.02	5.93 ± 0.16	5.69 ± 0.02	5.53 ± 0.07
67.3 ± 1.20	69.7 ± 5.94	67.3 ± 1.20	61.3 ± 5.40	67.3 ± 1.20	68.5 ± 4.08
2.98 ± 0.06	3.47 ± 0.32	2.98 ± 0.06	2.95 ± 0.35	2.98 ± 0.06	2.90 ± 0.20
	Addition grains 0: No eggs $(n = 14,686)$ 28.2 \pm 0.10 96.9 \pm 0.23 9.54 \pm 0.03 16.4 \pm 0.03 4.10 \pm 0.10 1.36 \pm 0.004 3.02 \pm 0.02 1.64 \pm 0.03 5.69 \pm 0.02 67.3 \pm 1.20 2.98 \pm 0.06	Additionally adjusted for MPF/ grains/potatoes/beveragesGr No eggs3: Egg/MPF/grains/potatoes/ $0: No eggs$ $3: Egg/MPF/grains/potatoes/(n = 14,686)beverages (n = 646)28.2 \pm 0.1028.3 \pm 0.2996.9 \pm 0.2397.2 \pm 0.769.54 \pm 0.039.90 \pm 0.1016.4 \pm 0.0316.5 \pm 0.144.10 \pm 0.104.20 \pm 0.401.36 \pm 0.0041.38 \pm 0.023.02 \pm 0.023.22 \pm 0.071.64 \pm 0.031.46 \pm 0.095.69 \pm 0.025.79 \pm 0.0767.3 \pm 1.2069.7 \pm 5.942.98 \pm 0.063.47 \pm 0.32$	$\begin{array}{ c c c c } \hline Egg parametric p$	$ \begin{array}{ $	$\begin{tabular}{ c c c c } \hline Egg \mbox{pattern} \\ \hline Additionally adjusted for MPF/ grains/potatoes/beverages (n = 646) (n = 14,686) (n = 1,64 \pm 0.03) (n = 14,64 \pm 0.03) (n $

¹ Values are least-squares means \pm SEs. *Different from no eggs, $P \le 0.0063$ (with Bonferroni correction). Systolic/diastolic blood pressure, kPa = mm Hg \times 0.1333; C-reactive protein, mg/L = mg/dL \times 10; HDL cholesterol, mmol/L = mg/dL \times 0.002586; LDL cholesterol, mmol/L = mg/dL \times 0.002586 \times mg/dL; TGs, mmol/L = mg/dL \times 0.01129; glucose, mmol/L = mg/dL \times 0.05551; insulin, pmol/L = mU/L \times 6. MPF, meat/poultry/fish.

² Adjusted for ethnicity, age, poverty income ratio, physical activity, smoking, and alcohol and all other food groups in the pattern (excluding egg consumption).

³ Adjusted for ethnicity, age, poverty income ratio, physical activity, smoking, alcohol, and BMI and all other food groups in the pattern (excluding egg consumption).

		Egg pattern	
Demographic/lifestyle variables	0: No eggs ² (<i>n</i> = 14,686)	3: Egg/MPF/grains/potatoes/beverages (<i>n</i> = 646)	7: Egg/MPF/grains ($n = 264$)
Age, y	45.8 ± 0.31	43.0 ± 0.54*	47.5 ± 1.50
Female, %	51.9 ± 0.47	34.0 ± 2.09*	49.2 ± 5.19
Race/ethnicity, %			
White	73.2 ± 1.53	62.6 ± 3.02	66.5 ± 4.36
African American	10.9 ± 0.92	16.2 ± 1.84*	10.7 ± 2.66
Hispanic American	10.8 ± 0.92	16.8 ± 2.37	20.4 ± 3.83
Mean poverty-income ratio	3.04 ± 0.04	2.8 ± 0.10	2.9 ± 0.18
Physical activity intensity, %			
Sedentary	30.0 ± 0.71	36.1 ± 2.54	32.0 ± 4.08
Light	33.1 ± 0.63	29.4 ± 2.63	28.4 ± 4.25
Moderate-vigorous	36.9 ± 0.92	34.5 ± 2.49	39.7 ± 4.79
Current smoker, %	25.4 ± 0.69	39.2 ± 2.77*	19.7 ± 3.93
Alcohol consumer, %	26.2 ± 0.86	$64.9 \pm 2.65^*$	20.8 ± 3.99
Alcohol, g/d	10.6 ± 0.41	48.0 ± 2.57*	5.1 ± 1.24*
Energy from fast food, %	12.9 ± 0.40	12.2 ± 1.15	21.6 ± 2.26*
Food security, %			
Marginal/low	17.2 ± 0.68	25.2 ± 2.03*	18.7 ± 3.07
BMI >40 kg/m ² , %	5.44 ± 0.27	$2.83 \pm 0.68^*$	10.1 ± 3.16
Medication for diabetes, %	5.24 ± 0.26	2.36 ± 0.52*	6.84 ± 2.15

¹ Values are least-squares means \pm SEs. *Different from no eggs, $P \leq 0.001$. MPF, meat/poultry/fish.

² Reference group.

with WC remained. This remaining association may be due to residual confounding (which will be addressed later in the discussion) or to other potentially significant variables not measured in the NHANES. It is unlikely that a single ingredient in a food pattern is responsible for a relation with WC. This is particularly true given that only 1.9-ounce equivalents of eggs were consumed in pattern 7. Among all subjects, the clustering explained 0.10% of the variation in WC across subjects after adjustment for covariates.

Another noteworthy finding was that there were differences in the percentage of the population taking diabetes medication. However, once medication use was controlled for in the final analysis, the association between egg consumption pattern 3 and DBP and LDL cholesterol disappeared. To further complicate the findings, the percentage of smokers and alcohol consumers was higher in egg consumption pattern 3, which may explain the elevated DBP and LDL cholesterol found in this egg consumption pattern. There is sufficient evidence that smoking and excessive alcohol consumption are adversely associated with blood pressure and LDL-cholesterol concentrations (37-44); thus, smoking and alcohol may be mediators in the associations that were found. However, smoking and alcohol intake were included as covariates in the analyses. A possible explanation for a discrepancy in our interpretation of the results may be due to residual confounding.

Residual confounding reflects the inability to measure accurately a potentially confounding variable; thus, correction for confounding is incomplete in the analyses. Measurement error associated with variables can lead to residual confounding. Confounding can also be caused by variables that are associated with both exposure and outcome (45-47). There are 3 potential causes of residual confounding factors that were not considered or there was no attempt to adjust for them because data on these factors were not collected, 2) data on confounding variables were not precise enough (e.g., smoking status, physical activity),

or 3) there may be errors in the classification of the subjects with respect to confounding variables.

To our knowledge, this is the first study to demonstrate differing effects of statistical approaches to assess the relation between an exposure variable and health outcomes. The relation between egg consumption and CVRFs varied considerably depending on the statistical approach and the covariates used in the analyses. All traditional dietary analyses in epidemiology share one strong but incorrect assumption: that exposures, such as foods or nutrients, were measured with great accuracy. Care needs to be taken with data interpretation of diet and health risk factors and the choice of statistical approaches because these epidemiologic studies are used to generate hypotheses. More studies are needed to develop statistical methods that reduce bias when evaluating dietary hypotheses in more detail. A number of methods are being explored, yet they are usually complicated and will not provide a simple solution (46, 47).

Acknowledgments

We thank Lori Briones for help in obtaining research articles and in preparing the manuscript. VLF analyzed the data; TAN and CEO wrote the manuscript; and TAN had primary responsibility for final content. All authors read and approved the final manuscript.

References

- Hoffmann I. Transcending reductionism in nutrition research. Am J Clin Nutr 2003;78(Suppl):5145–65.
- Committee on Diet and Health, Food and Nutrition Board, Commission on Life Sciences, National Research Council. Diet and health: implications for reducing chronic disease risk. Washington (DC): National Academies Press; 1989.
- Hu FB. Dietary pattern analysis: a new direction in nutritional epidemiology. Curr Opin Lipidol 2002;13:3–9.
- Byers T. The role of epidemiology in developing nutritional recommendations: past, present, and future. Am J Clin Nutr 1999;69:1304S–8S.

- 5. Newby PK, Tucker KL. Empirically derived eating patterns using factor or cluster analysis: a review. Nutr Rev 2004;62:177–203.
- 6. Kant AK. Dietary patterns and health outcomes. J Am Diet Assoc 2004;104:615-35.
- Jones-McLean EM, Shatenstein B, Whiting SJ. Dietary patterns research and its applications to nutrition policy for the prevention of chronic disease among diverse North American populations. Appl Physiol Nutr Metab 2010;35:195–8.
- 8. Tucker KL. Dietary patterns, approaches, and multicultural perspective. Appl Physiol Nutr Metab 2010;35:211–8.
- Slattery ML. Analysis of dietary patterns in epidemiological research. Appl Physiol Nutr Metab 2010;35:207–10.
- 10. Kant AK. Dietary patterns: biomarkers and chronic disease risk. Appl Physiol Nutr Metab 2010;35:199–206.
- 11. McNamara DJ. Dietary cholesterol, heart disease risk and cognitive dissonance. Proc Nutr Soc 2014;73:161-6.
- Shin JY, Xun P, Nakamura Y, He K. Egg consumption in relation to risk of cardiovascular disease and diabetes: a systematic review and metaanalysis. Am J Clin Nutr 2013;98:146–59.
- McNamara DJ. The impact of egg limitations on coronary heart disease risk: do the numbers add up? J Am Coll Nutr 2000;19:540S–8S.
- Gray J, Griffin B. Eggs and dietary cholesterol: dispelling the myth. Nutr Bull 2009;34:66–70.
- Kanter MMK-EP, Fernandez ML, Vickers KC, Katz DL. Exploring the factors that affect blood cholesterol and heart disease risk: is dietary cholesterol as bad for you as history leads us to believe? Adv Nutr 2012;3:711–7.
- Tran NL, Barraj LM, Heilman JM, Scrafford CG. Egg consumption and cardiovascular disease among diabetic individuals: a systematic review of the literature. Diabetes Metab Syndr Obes 2014;98:146–59.
- Blesso CN, Andersen CJ, Barona J, Volek JS, Fernandez ML. Whole egg consumption improves lipoprotein profiles and insulin sensitivity to a greater extent than yolk-free substitute in individuals with metabolic syndrome. Metabolism 2013;62:400–10.
- National Center for Health Statistics. Analytic and reporting guidelines: the National Health and Nutrition Examination Survey (NHANES). 2006 [cited 2014 Mar 25]. Available from: http://www.cdc.gov/nchs/ data/nhanes/nhanes_03_04/nhanes_analytic_guidelines_dec_2005.pdf.
- National Center for Health Statistics; Centers for Disease Control and Prevention. Welcome NHANES participants: is my information confidential? [cited 2014 Mar 25]. Available from: http://www.cdc.gov/nchs/ nhanes/participant.htm.
- Centers for Disease Control and Prevention. National Health and Nutrition Examination Survey. Is my survey information confidential? April 2013 [cited 2014 Mar 25]. Available from: http://www.cdc.gov/ nhanes/pQuestions.htm#.
- 21. U.S. Department of Health and Human Services; Office of Extramural Research; National Institutes of Health. NIH exploratory/developmental research grant award (R21). 2012 [cited 2014 Feb 5]. Available from: http://www.grants.nih.gov/grants/funding/r21.htm.
- 22. National Health and Nutrition Examination Survey. Physical activity and physical fitness. [cited 2014 Mar 12]. Available from: http://www.cdc.gov/nchs/data/nhanes/nhanes_07_08/paq07_08_eng.pdf.
- 23. Moshfegh AJ, Rhodes DG, Baer DJ, Murayi T, Clemens JC, Rumpler WV, Paul DR, Sebastian RS, Kuczynski KJ, Ingwersen LA, et al. The US Department of Agriculture Automated Multiple-Pass Method reduces bias in the collection of energy intakes. Am J Clin Nutr 2008;88:324–32.
- Moshfegh AJ, Raper N, Ingwersen I, Cleveland L, Anand J, Goldman J, LaComb R. An approved approach to 24-hour dietary recall methodology. Ann Nutr Metab 2001;45:156.
- Conway JM, Ingwersen LA, Vinyard BT, Moshfegh AJ. Effectiveness of the US Department of Agriculture 5-step multiple-pass method in assessing food intake in obese and nonobese women. Am J Clin Nutr 2003;77:1171–8.
- 26. Centers for Disease Control and Prevention; National Center for Health Statistics. National Health and Nutrition Examination Survey (NHANES). NHANES dietary interview component. [cited 2014 Mar 11]. Available from: www.cdc.gov/nchs/data/nhanes/nhanes_09_10/dietaryrecall_f.pdf.
- Friday J, Bowman S. 2006. MyPyramid Equivalents Database for USDA survey food codes, 1994–2002. Version 1.0. [cited 2014 Mar 11]. Available from: http://www.barc.usda.gov/bhnrc/cnrg.
- Bowman SA, Friday JE, Moshfegh A. 2006. MyPyramid Equivalents Database, 2.0 for USDA survey foods, 2003–2004 Food Surveys Research Group. 2006 [cited 2014 Mar 11]. Available from: http:// www.ars.usda.gov/ba/bhnrc/fsrg.

- USDA; Center for Nutrition Policy and Promotion. Healthy Eating Index-2005 development and evaluation technical report support files [cited 2014 Mar 11]. Available from: http://www.cnpp.usda.gov/HealthyEatingIndex-2005report.htm.
- Kennedy ET, Ohls J, Carlson S, Fleming K. The Healthy Eating Index: design and applications. J Am Diet Assoc 1995;95:1103–8.
- Guenther PM, Reedy J, Krebs-Smith SM, Reeve BB. Evaluation of the Healthy Eating Index-2005. J Am Diet Assoc 2008;108:1854–64.
- 32. USDA; Center for Nutrition Policy and Promotion. Calculation of the Healthy Eating Index-2005 component and total scores for a population, subpopulation, or group [cited 2013 Dec 20]. Available from: www.cnpp.usda.gov/Publications/HEI/HEI-2005/Readme_HEI2005_ NHANES0102PopulationScore.doc.
- 33. Centers for Disease Control and Prevention; National Center for Health Statistics. National Health and Nutrition Examination Survey. Anthropometry manual [cited 2014 Mar 12]. Available from: http://www.cdc. gov/nchs/data/nhanes/bm.pdf.
- 34. National Cholesterol Education Program; National Heart, Lung, and Blood Institute; National Institutes of Health. 2002. Detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III) [cited 2014 Mar 12]. Available from: http://www.nhlbi.nih.gov/ about/ncep/.
- 35. Centers for Disease Control and Prevention; National Center for Health Statistics. NHANES. Laboratory methods [cited 2014 Mar 12]. Available from: http://www.cdc.gov/nchs/nhanes/nhanes1999–2000/lab99_00.htm; http://www.cdc.gov/nchs/nhanes/nhanes2001–2002/lab01_02.htm; http:// www.cdc.gov/nchs/nhanes/nhanes2003–2004/lab03_04.htm; http:// www.cdc.gov/nchs/nhanes/nhanes2005–2006/lab_methods_05_06.htm; http:// www.cdc.gov/nchs/nhanes/nhanes2007–2008/lab_methods_07_08.htm; http://www.cdc.gov/nchs/nhanes/nhanes2009–2010/lab_methods_09_ 10.htm; http://www.cdc.gov/nchs/nhanes/nhanes2011–2012/lab_methods_ 11_12.htm.
- Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and betacell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 1985;28:412–9.
- 37. Menotti A, Keys A, Blackburn H, Kromhout D, Karvonen M, Nissinen A, Pekkanen J, Punsar S, Fidanza F, Giampaoli S, et al. Comparison of multivariate predictive power of major risk factors for coronary heart diseases in different countries: results from eight nations of the Seven Countries Study, 25-year follow-up. J Cardiovasc Risk 1996;3:69–75.
- Jonas MA, Oates JA, Ockene JK, Hennekens CH. Statement on smoking and cardiovascular disease for health care professionals. American Heart Association. Circulation 1992;86:1664–9.
- Kannel WB. Importance of hypertension as a major risk factor in cardiovascular disease. In: Genest J, Koiw E, Kuchel O, editors. Hypertension: physiopathology and treatment. New York: McGraw-Hill Book Company; 1977:888–910.
- 40. U.S. Department of Health and Human Services. The health consequences of smoking: a report of the Surgeon General. Atlanta (GA): U.S. Department of Health and Human Services, Centers for Disease Control and Prevention, National Center for Chronic Disease Prevention and Health Promotion, Office on Smoking and Health; 2004 [cited 2014 Mar 11]. Available from: http://www.ncbi.nlm.nih.gov/books/n/smokeuse/pdf/.
- Bagnardi V, Zatonski W, Scotti L, La Vecchia C, Corrao G. Does drinking pattern modify the effect of alcohol on the risk of coronary heart disease? Evidence from a meta-analysis. J Epidemiol Community Health 2008;62:615–9.
- Cleophas TJ. Wine, beer and spirits and the risk of myocardial infarction: a systematic review. Biomed Pharmacother 1999;53:417–23.
- Djoussé L, Gaziano JM. Alcohol consumption and heart failure: a systematic review. Curr Atheroscler Rep 2008;10:117–20.
- 44. Di Castelnuovo A, Rotondo S, Iacoviello L, Donati MB, De Gaetano G. Meta-analysis of wine and beer consumption in relation to vascular risk. Circulation 2002;105:2836–44.
- Greenland S, Robins JM. Identifiability, exchangeability, and epidemiologic confounding. Int J Epidemiol 1986;15:413–9.
- Fraser GE. A search for truth in dietary epidemiology. Am J Clin Nutr 2003;78:5215–55.
- Flanders WD, Khoury MJ. Indirect assessment of confounding: graphic description and limits on effect of adjusting for covariates. Epidemiology 1990;1:239–46.

NUTRITION

OF

JOURNAL

THE

 \geq