Short Communication

Estimation of usual intake and food sources of choline and betaine in New Zealand reproductive age women

Vanessa L Mygind M Diet, Sophie E Evans M Diet, Meredith C Peddie PhD, Jody C Miller PhD, Lisa A Houghton PhD

Department of Human Nutrition, University of Otago, Dunedin, New Zealand

Recently, choline has been associated with neurodevelopment, cognitive function and neural tube defect incidence. However, data on usual intakes are limited, and estimates of dietary intakes of choline and its metabolite betaine, are not available for New Zealanders. The objective of the present study was to determine usual intake and food sources of choline and betaine in a group of New Zealand reproductive age women. Dietary intake data were collected from a sample of 125 women, aged 18-40 years, by means of a 3-day weighed food record, and usual choline and betaine intake distributions were determined. The mean (SD) daily intakes of choline and betaine were 316 (66) mg and 178 (66) mg, respectively. The total choline intake relative to energy intake and body weight was 0.18 mg/kcal and 5.1 mg/kg, respectively. Only 16% of participants met or exceeded the Adeuate Intake (AI) for adult women of 425 mg of choline. The top five major food contributors of choline were eggs, red meat, milk, bread and chicken; and of betaine were bread, breakfast cereal, pasta, grains and root vegetables (carrots, parsnips, beetroot, swedes). Our findings contribute towards the recent emergence of published reports on the range of dietary choline and betaine intakes consumed by free-living populations. In our sample of New Zealand women, few participants were meeting or exceeding the AI level. Given recent epidemiological evidence suggesting health benefits of increased choline and betaine intakes, recommendations should be made to encourage the consumption of choline and betaine-rich foods.

Key Words: choline, betaine, reproductive age women, usual intakes, major food contributors

INTRODUCTION

Choline is present in a wide variety of foods with eggs and meats representing the richest food sources. It is a precursor for a number of compounds including acetylcholine, phospholipids, and platelet activating factor.[1] Betaine, a derivative of choline, is important because of its role in the donation of methyl groups to homocysteine to form methionine – the universal methyl donor needed for methylation of DNA, RNA and proteins.[2] The relation of choline to lipid and one-carbon metabolism has led to several recent investigations examining the role of choline and betaine in neurodevelopment and cognitive function,[5-7] and pathogenesis of various chronic diseases including cancer and cardiovascular disease.[8-14] In reproductive-age women, a lower dietary intake of choline, and possibly betaine, in the periconceptional period is associated with an increased risk of having an neural tube defect-affected pregnancy, even after controlling for folate intake.[15]

In 1998, an Adequate Intake (AI) of 425 mg of choline per day for adult women (equivalent to 7 mg/kg body weight per day) was set instead of a Recommended Daily Allowance as sufficient evidence was not available at the time for calculating an Estimated Average Requirement.[16] Currently there is no recommendation set for dietary betaine. Additional data on the range of usual dietary intakes are needed to aid in the establishment of Nutrient Intake Values. Thus, the overall aim of this study was to describe the usual intake of choline and betaine in a group of healthy New Zealand reproductive age women. In addition, the major food contributors of choline and betaine in diets of the participants were identified to determine the commonly consumed foods that contribute significantly to the usual intake.

METHODS

This study was part of the baseline dietary data collection for a folate intervention trial conducted in New Zealand between July 2008 and May 2009.[19,20]In this study, 140 healthy, reproductive age women (18-40 years) were recruited from the staff and student population at the University of Otago, Dunedin, New Zealand, and from the local community. Women were excluded if they were pregnant, lactating, or were planning a pregnancy in the next 12 months. The population included in these analyses is composed of 125 women with complete 3-day weighed food records at the baseline visit. Ethical approval for the study was obtained from the Human Ethics

Corresponding Author: Dr Lisa Houghton, Department of Human Nutrition, University of Otago, New Zealand.
Tel: +64 3 479 7294; Fax: +64 3 479 7958
Email: lisa.houghton@otago.ac.nz
Manuscript received 16 February 2013. Initial review completed 27 February 2013. Revision accepted 11 March 2013.
doi:10.6133/apjcn.2013.22.2.19

Committee of the University of Otago, Dunedin, New Zealand, and all participants gave written, informed consent.

Participants were given a self-administered questionnaire to collect socio-demographic and general health information; height and weight were taken according to standardised procedures. BMI (kg/m^2) was classified using World Health Organisation (WHO) criteria as underweight (≤18.49), normal weight (18.5-24.9), overweight (25.0 – 29.9) and obese (≥30.0). Participant ethnicity was classified as New Zealand European, Maori and Pacific Peoples, Asian or other ethnicities; and education as secondary school or less, post-secondary education (university or college), or advanced degree.

Dietary intake of choline and betaine was quantified using 3-day weighed food records completed over two non-consecutive weekdays and one weekend day. Participants were provided with electronic digital kitchen scales accurate to 1 g (Vista Electronic 21 Kitchen Scale, model 3010, Salter Housewares Ltd, Tonbridge, England) and received instructions on how to weigh and record all food and beverages consumed.

Energy intakes were estimated from food and beverages consumed using the New Zealand Food Composition Database.[21] The New Zealand database does not contain nutrient-content for choline or betaine and these values were obtained for each food item from the US Department of Agriculture (USDA) National Nutrient Database for Standard Reference[17] or the USDA Database for the Choline Content of Common Foods Release 2.[18] The latter database contained values for choline-containing compounds, including water-soluble compounds (ie, free choline, glycerophosphocholine, and phosphocholine) and lipid-soluble compounds (ie, phosphatidylcholine and sphingomyelin); however, most foods were found only in the USDA Database for Standard Reference, which presents a single value for total choline. Commercial food products and foods unique to New Zealand that were not found in the USDA databases were assigned choline and betaine values from substitute foods with similar ingredients. Therefore, all foods were assigned total choline and betaine values (ie, no missing values) with the exception of Marmite™, a yeast extract spread, where the betaine content was unknown and no appropriate substitution was found.

To determine the most important food sources of choline and betaine, 7761 food entries were sorted into 97 food categories on the basis of nutrient similarities. Determination of the food categories that represented the highest percent contribution to dietary choline and betaine were calculated using the following formula:

[(Total choline/betaine provided by food category)÷(Total choline/betaine from all food categories)] × 100.

All statistical analysis was performed using Stata (version 11; Stata Corp, College Station, TX). Estimates of mean and percentiles of usual dietary intake for free choline, glycerophosphocholine, phosphocholine, phosphatidylcholine, sphingomyelin, total choline and total betaine were expressed in mg/d and were adjusted for the distribution of observed intakes to partially remove the day-to-day variability in choline and betaine intakes (within-person variation) using PC-SIDE version 1.012 (Department of Statistics, Iowa State University, Ames, 22 IA, USA).

RESULTS

The mean (SD) age of the participants was 24 (6) years. Most of the women were well-educated (88%; 110 of 125 were enrolled in or had completed a post-secondary degree) and were mainly New Zealand Europeans (75%; 94 of 125). The mean (SD) BMI was 24 (4), ranging from 17.4 to 39.0. Sixty-nine percent of participants were classified as normal weight, with the remaining participants classified as underweight (4%), overweight (22%) and obese (6%).

The mean (SD) daily energy intake was 1842 (354) kcal. The daily dietary intake for total choline [mean (SD)] was 316 (65) mgand percentiles of usual choline intakes are presented in Table 1. Total choline intake relative to reported energy intake and measured body weight was 0.18 mg/kcal and 5.1 mg/kg, respectively. Only 16% (20 of 125) of the participants met or exceeded the estimated choline requirement for adult women of 425 mg.

Table 2 presents data on the major contributors of choline in the diets of study participants. Of 7761 food entries sorted into 97 food categories, eggs and red meat, the top two items, constitute nearly one-quarter of the total choline intake. Forty-two percent of participants (53 of 125) reported consuming ≥1 egg on at least one of three diet record collection days, with 14% of participants (18 of 125) consuming ≥1 egg on two of the three days. For those participants reporting egg consumption, the average egg intake per day was 1.3 eggs, and ranged from 0.2 to 5.1 eggs, providing 25 to 638 mg of choline. Over half (58%) of the participants did not record eating at least one egg on any diet record collection day. Of those who reported red meat consumption, the average intake ranged from 24 to 355 g, providing 17 to 291 mg of choline. Other animal-based products such as milk, chicken,

Table 1. Mean and percentiles of usual daily choline and betaine intakes among study participants (*n*=125)

	Mean±SD	Percentiles								
		1st	5th	10th	25th	50th	75th	90th	95th	99th
Total choline, mg/d	316±65	189	219	237	269	310	356	402	433	495
Free choline, mg/d	65±17	35	41	45	53	63	76	89	98	117
Choline from glycerophosphocholine, mg/d	51±18	22	28	32	39	49	61	75	84	106
Choline from phosphocholine, mg/d	13±4	6	8	9	10	12	15	17	19	22
Choline from phosphatidylcholine, mg/d	123±32	63	77	85	100	119	142	166	181	214
Choline from sphingomyelin, mg/d	14±5	4	6	8	9	13	17	21	23	28
Betaine, mg/d	178±66	77	96	107	131	166	212	263	300	388

Table 2. Major food contributors of choline and betaine in a group of New Zealand reproductive age women (*n*=25)

Rank	Choline			Betaine		
	Food Category	Percentage	Cumulative Percentage	Food Category	Percentage	Cumulative Percentage
1	Eggs	13.4	13.4	Bread	25.5	25.5
2	Red meat	10.0	23.4	Breakfast cereal	16.9	42.5
3	Milk	8.0	31.3	Pasta	13.0	55.4
4	Bread	5.5	36.8	Grains(Quinoa/couscous/semolina/bulgar)	8.4	63.8
5	Chicken	4.5	41.4	Root vegetables (Carrot/beetroot/parsnip/swedes)	4.8	68.6
6	Fish	4.2	45.6	Chicken	3.1	71.7
7	Fruits	4.1	49.7	Biscuits	2.5	74.3
8	Pork	3.5	53.2	Red meat	2.3	76.6
9	Cakes	3.0	56.2	Fish	2.3	78.9
10	Yoghurt	2.5	58.7	Cakes	2.1	81.0

fish and pork were also important contributors. Interestingly, bread ranked in the top 5 major contributors of choline in this sample of reproductive aged women, contributing approximately 6% of total dietary choline intake.

Mean (SD)daily betaine intake was 178 (66) mg (Table 1). Relative to energy ingested and body weight, the average betaine intake was 0.10 mg/kcal and 2.8 mg/kg, respectively. The food category contributing the most betaine in the diet was bread (26%), with breakfast cereals (17%) and pasta (13%) ranking second and third, respectively. The contribution of these foods combined provided more than one half of the total betaine intake (Table 2).

DISCUSSION

Using the food choline content from the USDA food composition database, we estimated that only a small proportion of our study participants met or exceeded the recommended intake level for choline. To our knowledge, this is the first study to assess the usual daily choline intakes of a sample of healthy New Zealand reproductive age women. While our data show that the usual mean intake in this sample of women is lower than the recommended daily intake, our estimates are consistent with other studies.[5,8,9,22-26] Nonetheless, the self-reported nature of the data may have led to an underestimation of choline due to an underreporting of food intake. For example, a study by Fischer *et al*,[26] found that estimated self-reported choline intakes from 3-day food records were 25% lower compared with measured intake from food consumed *ad libitum* in a clinical research setting; however, the difference between the observed and reported values was non-existent when choline intakes were normalized for energy. Remarkably, our energy-adjusted choline intakes [0.2 mg/kcal] were identical to those reported by Fischer *et al*,[26] inferring accurate determination of the choline composition of the diet but underreporting of energy.

The ability to rank the major food contributors to choline intake are less likely to be affected by underreporting of energy intakes. In this population of reproductive age women, eggs, red meat and milk were the top contributors of choline intake. In a recent national survey in Taiwan, eggs also ranked as the top contributor of choline, however, the contribution was much larger among Taiwanese adults, contributing 25% to total choline intake compared to only 13% in our study participants.[24] In contrast, eggs ranked fourth in a US study of adult men and women, contributing less than 8% to total diet while red meat ranked as the top food source, contributing 14%.[8] Animal products contain considerably more choline per unit weight than plants, however, when summed together, fruits and grain-based food products such as bread contributed an equally dominant source of total choline in the diet.

Dietary intake of betaine in our study population was somewhat lower than values published from larger cohorts in the United States (208 mg),[8] Greece (314 mg)[9] and the Netherlands (241 mg) [22] although the intakes in our study were greater than those reported from the 1993-1996 Nutrition and Health Survey in Taiwan (78 mg).[24] In most studies, betaine intake was calculated with the use of a self-reported food frequency questionnaire (FFQ). FFQs are typically designed to rank participants from highest to lowest intake rather than quantifying the absolute intake of choline. In contrast to choline, which is predominantly found in animal sources, betaine is rich in plant-based foods such as wheat bran, quinoa, beets and spinach[27] as reflected in the top five food sources in our study contributing nearly two-thirds to total betaine intake. The top food contributors of betaine among our study participants were similar to those recently reported in an American study, with bread ranking as the highest contributor to betaine intake (USA, 22% compared to our study, 26%).[8] Conversely, dark leafy vegetables ranked as the major contributor of betaine in Taiwan, with bread contributing only 5% to total betaine intake.[24] At present, there is no recommended intake level for betaine. Humans can obtain betaine either from diet or endogenous synthesis from choline. Establishing a recommended intake level will require a better understanding of the role of betaine in one-carbon metabolism as well as its interrelationship with choline, folate and methionine. For example, a higher intake of folate appears to have a sparing effect on betaine, as shown by an increase in plasma betaine concentrations following supplementation with folic acid.[28]

While weighed diet records are considered the most accurate method to measure usual nutrient intake,[29] several limitations of our study should be acknowledged. First, the New Zealand food composition tables do not contain values for choline and betaine, rather these were derived from similar foods listed in the USDA Nutrient Database.[17] Substitutions were especially difficult for local foods, which may not be typically consumed in a North American diet. For example, silverbeet, a commonly consumed leafy green in New Zealand was replaced with spinach. Spinach is an excellent source of betaine, however, the differences in the betaine composition of spinach compared to silverbeet is unknown. In addition, Marmite™, a yeast extract spread with no assigned betaine value, was consumed by one-quarter of participants (33 of 125) on at least one day. Yeast extract spreads contain high amounts of B-vitamins including choline and are likely to contain appreciable amounts of betaine. The possibility that betaine intakes may have been underestimated based on the missing value for this commonly consumed food needs to be considered. In addition, various food processing and cooking methods have been found to affect betaine content with boiling resulting in losses of up to 80%.[27,30] Future studies should undertake the compositional analysis of locally consumed foods. Secondly, our small sample size of predominantly university-educated women who identified mostly as New Zealand Europeans does not permit generalization of our results to the general population. Nationally representative survey data on choline and betaine intakes are needed. Nevertheless, our study is strengthened by the adjustment of observed intakes to remove the day-to-day within-person variation.

In conclusion, our findings contribute toward recent reports suggesting that choline intakes may be suboptimal for a large percentage of the population. However, more work is needed to better understand dietary choline and betaine requirements. For example, the choline AI for non-pregnant, non-lactating women, set in 1998, was extrapolated from a single study conducted in men and designed to prevent liver dysfunction.[16] In the meantime, obtaining choline and betaine from the food supply is attainable if proper food choices are made and would be naturally aligned with dietary recommendations.

ACKNOWLEDGEMENT
We thank Elizabeth Gray for her assistance in assigning choline and betaine values to existing foods in the New Zealand Food Composition table. This study was supported by University of Otago Research Grant.

AUTHOR DISCLOSURES
None of the authors had a personal or financial conflict of interest.

REFERENCES
1. Zeisel SH, Blusztajn JK. Choline and human nutrition. Annu Rev Nutr. 1994;14:269-96.
2. McKeever MP, Weir DG, Molloy A, Scott JM. Betaine-homocysteine methyltransferase: organ distribution in man, pig and rat and subcellular distribution in the rat. Clin Sci (Lond). 1991;81:551-6.
3. Jacob RA, Jenden DJ, Allman-Farinelli MA, Swendseid ME. Folate nutriture alters choline status of women and men fed low choline diets. J Nutr. 1999;129:712-7.
4. Ueland PM. Choline and betaine in health and disease. J Inherit Metab Dis. 2011;34:3-15.
5. Poly C, Massaro JM, Seshadri S, Wolf PA, Cho E, Krall E et al. The relation of dietary choline to cognitive performance and white-matter hyperintensity in the Framingham Offspring Cohort. Am J Clin Nutr. 2011;94:1584-91.
6. Strain JJ, McSorley EM, van Wijngaarden E, Kobrosly RW, Bonham MP, Mulhern MS et al. Choline status and neurodevelopmental outcomes at 5 years of age in the Seychelles Child Development Nutrition Study. Br J Nutr. 2013. Epub Jan 9:1-7
7. Wu BT, Dyer RA, King DJ, Richardson KJ, Innis SM. Early second trimester maternal plasma choline and betaine are related to measures of early cognitive development in term infants. PLoS One. 2012;7:e43448.
8. Cho E, Zeisel SH, Jacques P, Selhub J, Dougherty L, Colditz GA et al. Dietary choline and betaine assessed by food-frequency questionnaire in relation to plasma total homocysteine concentration in the Framingham Offspring Study. Am J Clin Nutr. 2006;83:905-11.
9. Detopoulou P, Panagiotakos DB, Antonopoulou S, Pitsavos C, Stefanadis C. Dietary choline and betaine intakes in relation to concentrations of inflammatory markers in healthy adults: the ATTICA study. Am J Clin Nutr. 2008;87: 424-30.
10. Lever M, George PM, Atkinson W, Molyneux SL, Elmslie JL, Slow S et al. Plasma lipids and betaine are related in an acute coronary syndrome cohort. PLoS One. 2011;6:e21666.
11. Xu X, Gammon MD, Zeisel SH, Bradshaw PT, Wetmur JG, Teitelbaum SL et al. High intakes of choline and betaine reduce breast cancer mortality in a population-based study. FASEB J. 2009;23:4022-8.
12. Cho E, Holmes M, Hankinson SE, Willett WC. Nutrients involved in one-carbon metabolism and risk of breast cancer among premenopausal women. Cancer Epidemiol Biomar. 2007;16:2787-90.
13. Lee JE, Giovannucci E, Fuchs CS, Willett WC, Zeisel SH, Cho E. Choline and betaine intake and the risk of colorectal cancer in men. Cancer Epidemiol Biomar. 2010;19:884-7.
14. Cho E, Holmes MD, Hankinson SE, Willett WC. Choline and betaine intake and risk of breast cancer among post-menopausal women. Brit J Cancer. 2010;102:489-94.
15. Shaw GM, Carmichael SL, Yang W, Selvin S, Schaffer DM. Periconceptional dietary intake of choline and betaine and neural tube defects in offspring. Am J Epidemiol. 2004;160:102-9.
16. Institute of Medicine, Food and Nutrition Board. Dietary Reference Intakes for Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin and Choline. Washington, DC: National Academy Press; 1998.
17. United States Department of Agriculture (USDA), Agricultural Research Service. USDA National Nutrient Database for Standard Reference, release 24. 2011 [accessed November 2012]. http://www/ars/usda/gov/food-composition/usda-nutrient-data-laboratory.
18. United States Department of Agriculture. USDA Database for the Choline Content of Common Foods. Release 2. http//www.ars.usda.gov/nutrientdata. Beltsville, Maryland: US Department of Agriculture; 2008.
19. Houghton LA, Gray AR, Rose MC, Miller JC, Hurthouse NA, Gregory JF, 3rd. Long-term effect of low-dose folic acid intake: potential effect of mandatory fortification on the prevention of neural tube defects. Am J Clin Nutr. 2011;94: 136-41.
20. Hursthouse NA, Gray AR, Miller JC, Rose MC, Houghton LA. Folate status of reproductive age women and neural tube defect risk: the effect of long-term folic acid supplementation at doses of 140 μg and 400 μg per day. Nutrients. 2011;3:49-62.

21. New Zealand Institute for Plant and Food Research Ltd. FOODfiles. Data files of the New Zealand Food Composition Database 2010 version 02. Palmerston North, New Zealand.
22. Dalmeijer GW, Olthof MR, Verhoef P, Bots ML, van der Schouw YT. Prospective study on dietary intakes of folate, betaine, and choline and cardiovascular disease risk in women. Eur J Clin Nutr. 2008;62:386-94.
23. Bidulescu A, Chambless LE, Siega-Riz AM, Zeisel SH, Heiss G. Usual choline and betaine dietary intake and incident coronary heart disease: the Atherosclerosis Risk in Communities (ARIC) study. BMC Cardiovasc Disord. 2007; 7:20.
24. Chu DM, Wahlqvist ML, Chang HY, Yeh NH, Lee MS. Choline and betaine food sources and intakes in Taiwanese. Asia Pac J Clin Nutr. 2012;21:547-57.
25. Guerrerio AL, Colvin RM, Schwartz AK, Molleston JP, Murray KF, Diehl A, et al. Choline intake in a large cohort of patients with nonalcoholic fatty liver disease. Am J Clin Nutr. 2012;95:892-900.
26. Fischer LM, Scearce JA, Mar MH, Patel JR, Blanchard RT, Macintosh BA et al. Ad libitum choline intake in healthy individuals meets or exceeds the proposed adequate intake level. J Nutr. 2005;135:826-9.
27. Slow S, Donaggio M, Cressey P, Lever M, George P, Chambers S. The betaine content of New Zealand foods and esitmated intake in the New Zealand diet. J Food Composit Anal. 2005;18:473-85.
28. Melse-Boonstra A, Holm PI, Ueland PM, Olthof M, Clarke R, Verhoef P. Betaine concentration as a determinant of fasting total homocysteine concentrations and the effect of folic acid supplementation on betaine concentrations. Am J Clin Nutr. 2005;81:1378-82.
29. Gibson RS. Measuring food composition of individuals. Principles of nutritional assessment. 2nd ed. New York: Oxford University Press; 2005. pp. 41-60.
30. de Zwart FJ, Slow S, Payne RJ, Lever M, George PM, Gerrard JA et al. Glycine betaine and glycine betaine analogues in common foods. Food Chem. 2003;83:197-204.

Short Communication

Estimation of usual intake and food sources of choline and betaine in New Zealand reproductive age women

Vanessa L Mygind M Diet, Sophie E Evans M Diet, Meredith C Peddie PhD, Jody C Miller PhD, Lisa A Houghton PhD

Department of Human Nutrition, University of Otago, Dunedin, New Zealand

紐西蘭育齡婦女膽鹼及甜菜鹼的日常攝取量及食物來源評估

近年來，膽鹼被認為與神經發展、認知功能及神經管缺陷的發生有關。然而，紐西蘭人的膽鹼日常攝取資料不足，且缺乏膽鹼及其代謝物甜菜鹼的飲食攝取量評估。本研究的目的為估算紐西蘭育齡婦女，其膽鹼及甜菜鹼的日常攝取量及食物來源。飲食攝取數據是來自 125 名年齡 18-40 歲的婦女樣本，收集三天的秤重食物記錄，估算膽鹼及甜菜鹼攝取的分布值。膽鹼及甜菜鹼每日攝取平均(標準差)量分別為 316(66)毫克及 178(66)毫克。總膽鹼攝取量相對於熱量攝取及體重分別為 0.18 mg/kcal 及 5.1 mg/kg。只有 16%的參與者符合或超過成年女性膽鹼的足夠攝取量(AI) 425 毫克。膽鹼的前五項主要食物來源為蛋類、紅肉類、奶類、麵包及雞肉；甜菜鹼的來源為麵包、早餐穀片、麵條類、穀類及根莖類(紅蘿蔔、歐洲防風草根、甜菜根、瑞典蕪菁)。近期出現較多對於一般人群的膽鹼及甜菜鹼飲食攝取量範圍的報告，本研究結果將對此有貢獻。在這群紐西蘭女性樣本中，少數參與者符合或是超過 AI 量。鑑於近期流行病學證實增加膽鹼及甜菜鹼攝取的健康效益，應提出建議鼓勵攝取富含膽鹼及甜菜鹼的食物。

關鍵字：膽鹼、甜菜鹼、育齡婦女、日常攝取量、主要食物貢獻者