Folate Adequacy and Diet Quality on the Royal Prince Alfred Hospital Allergy Unit Elimination Diet

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Declaration

The candidate, *Anita Tsui*, hereby declares that none of the work presented in this essay has been submitted to any other University or Institution for higher degree and that to the best of her knowledge contains no material written or published by another person, except where due reference is made in the text.

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Abstract

Aims: The current study aims to assess and compare folate adequacy and diet quality before and on the Elimination Diet. This study also aims to determine the impact of gluten exclusion and folic acid supplementation on folate intake on the Elimination Diet.

Methods: Four-day weighed food records were used in this study. The folate adequacy of baseline and Elimination Diet weighed food records were analysed on FoodWorks8, Xyris Software, and compared to the folate Estimated Average Requirement. Diet quality was assessed using the Healthy Eating Index for Australian Adults – 2013. Descriptive and inferential statistics were used to analyse the data with $p \leq 0.05$ considered as statistically significant.

Results: Mean folate intake significantly decreased on the Elimination Diet (105% Estimated Average Requirement) compared to baseline (169% Estimated Average Requirement). On the Elimination Diet, the proportion of gluten-excluding participants with inadequate folate intake more than doubled compared with gluten-including participants. Diet quality improved on the Elimination Diet compared to baseline. At baseline and on the Elimination Diet, all participants who took folic acid-containing supplements had folate intake above the Estimated Average Requirement.

Conclusion: Folate intake decreases on the Elimination Diet and gluten-excluding patients are most at risk of inadequate folate intake. It is therefore important to recommend intake of folate-rich sources allowed on the Elimination Diet. It may also be necessary to recommend the use of folic acid-containing supplements.

Key words: diet quality, Elimination Diet, folate, food intolerance, gluten

Introduction

Food intolerance is a non-immune reaction to certain naturally occurring food chemicals (e.g. salicylates, amines and glutamate), food additives and/or whole food groups (gluten, dairy, and soy). Food intolerance symptoms and severity vary between individuals and can affect one or more body systems including the skin, gastrointestinal tract (GIT), central nervous system and airways.[1] The Royal Prince Alfred Hospital (RPAH) Elimination Diet (ED) and Challenge Protocol is the sole method to reliably determine which food chemicals are responsible for the symptoms an individual experiences. The low chemical ED only allows foods that are low in chemicals that can trigger a reaction. In addition, patients are required to exclude gluten containing foods, dairy and/or soy products if they experience gastrointestinal symptoms.

The ED is followed for at least 2 weeks and after 5 consecutive days without symptoms, the patient goes onto food and/or capsule challenges to determine the reaction-causing chemicals. An individualised diet is then prescribed and liberalised under a dietitian's supervision to maintain the chemical load under the individual's threshold.[1] Even though the ED is a temporary diet, maintaining good nutrition on a restrictive diet is still important. However, for highly sensitive individuals, going on the ED may be life-long.

Despite the decrease in food choices on the ED (on-ED), diet quality was shown to improve on-ED compared to baseline. Discretionary food consumption decreased and macronutrient intake became more in-line with the Accepted Macronutrient Distribution Ranges (AMDRs) (Hooper, I. 2014; Nearchou, M. 2015; unpublished data). However,

on-ED, folate, calcium, vitamin A and thiamine were nutrients at risk of inadequate intake (Hooper, I. 2014; Nearchou, M. 2015; Tripodi, E. 2016; unpublished data).

The current study will focus on folate. Previous studies have found folate intake decreased on the low chemical ED, however average folate intake on-ED still met the Estimated Average Requirement (EAR).[2] The decrease in folate intake was speculated to be attributed to a majority of patients also following a gluten-excluding diet (Nearchou, M. 2015; Tripodi, E. 2016; unpublished data). Studies of patients with coeliac disease also confirm that folate intake is lower in patients following a gluten-free diet compared to the general population.[3,4]

Folate is a B group vitamin essential in DNA synthesis and nucleotide metabolism. Inadequate folate intake is the main cause of folate deficiency which is associated with macrocytosis, cardiovascular complications, neural tube defects (NTDs) in developing foetuses and neurological disorders.[5,6]

From 2009, folic acid fortification of all wheat flours for bread making was made mandatory in Australia.[7] However, it is not mandatory for non-wheat containing bread to be fortified, though manufacturers of non-wheat breads and other foods (e.g. breakfast cereals) may choose to voluntarily fortify their foods with folic acid. Post-fortification, regular breads and rolls contributed 36% to total Dietary Folate Equivalents (DFE) intake.[7] Other sources of DFE include vegetables, fruit and legumes.[8] On-ED, many folate-rich vegetables and fruits are restricted.

The current study aims to assess and compare the adequacy and intake of folate and diet quality of participants at baseline and during the ED. This study also aims to determine the impact of gluten exclusion and folic acid supplementation on folate intake.

Consequently, results from this study will be used to inform clinical practice to ensure patients are able to achieve folate adequacy on the ED.

Methods

The current study analysed data collected from a 5-year prospective observational study at the RPAH Allergy Unit looking at the nutritional adequacy and diet quality of patients on the ED. The study was approved by the Ethics Review Committee (RPAH Zone) of the Sydney Local Health District, protocol no: X13-0208. The study conforms to the provisions of the Declaration of Helsinki (as revised in Edinburg 2008).

A detailed methodology of participant recruitment and data entry could be found in previous unpublished reports (Nearchou, M. 2015; Tripodi, E. 2016; unpublished data). In brief, eligible participants were recruited prior to their initial appointment at the Allergy Unit. Participants were required to complete a 4-day weighed food record (WFR) consisting of 3 weekdays and 1 weekend day to capture their baseline diet before their initial appointment.

At the initial appointment, baseline WFRs were collected and height and weight were measured. One month after commencing the ED, participants were asked to complete an additional WFR.

WFRs were entered into and analysed on FoodWorks8 (Xyris Software) using food composition databases AUSNUT 2011-2013, AusFood 2015 and Aus Brands 2015 to obtain folate intake. Folate intake was compared as a percentage of the folate EAR.[8] Patients with WFRs excluding gluten, dairy and/or soy were also tagged for analysis. Participants who took folic acid (FA)-containing supplements were tagged and WFRs were analysed with supplements excluded as well as with supplements included.

Food and drink items were coded as core or non-core based on the Australian Bureau of Statistics Discretionary Food List which was derived from the Australian Dietary Guidelines 2013 (ADGs) and used in the Australian Health Survey 2011-13.[9] Mixed dishes were further broken down into individual ingredients which were then coded into food groups using an amplified version of the Healthy Eating Index for Australians (HEIFA) developed based on the ADGs and Australian Guide to Healthy Eating (AGHE).[10]

A total HEIFA score ranging from 0 to 100 was calculated to determine diet quality with a higher score indicating a better diet quality. The HEIFA score was calculated as the sum of 11 sub-scores which were based on the adherence to ADG recommendations on intake of different food groups and nutrients.

Descriptive and inferential statistics were performed on Microsoft Excel 2007, and GraphPad Prism 7. A p-value ≤ 0.05 was considered statistically significant.

Results

A total of 102 participants, with an age range of 19 – 80 years, were included in the study sample. Forty-one participants had a baseline WFR only and 61 had both a baseline and on-ED WFR. The total sample had a mean age of 43, mean BMI of 24.3, and 80 of the 102 participants were female. No statistical difference existed between the mean age, mean BMI and gender proportion of participants with only a baseline WFR and participants with both WFRs.

Seventy-eight (76%) participants presented to the Allergy Unit with one or more dietary restrictions. The most frequently restricted food groups were milk and milk products (43%), gluten-containing breads, cereals and pasta (38%), vegetables (25%), and fruit and fruit juices (25%). The most commonly reported foods that trigger a reaction were gluten-containing breads, cereals and pasta (45%), vegetables (45%), fresh or dried fruit (44%) and dairy and dairy products (40%). Overall, 92 (90%) participants had two or more body systems affected with the GIT, CNS, respiratory system and skin being affected in 87%, 71%, 62% and 61% of participants respectively.

The following results on folate adequacy have excluded supplements from the analysis unless otherwise specified.

Figure 1 shows folate intake as a percentage of the folate EAR at baseline and on-ED as a whole group, and separated as gluten-excluding, and gluten-including participants. Overall, mean folate intake was significantly lower on-ED compared to baseline, 105% EAR and 169% EAR respectively. A greater proportion of participants were not

meeting the folate EAR on-ED, 34 of 61 (56%), compared to baseline, 12 of 102 (12%). On-ED, participants excluding gluten had a significantly lower mean folate intake (87% EAR) compared to participants including gluten (135% EAR). Also, on-ED, a greater proportion of participants excluding gluten, 27 of 38 (71%), had inadequate folate intake compared to participants including gluten, 7 of 23 (30%).

When paired baseline and on-ED WFRs were analysed, mean folate intake decreased on-ED in all participants. Mean folate intake significantly decreased on-ED in participants who included gluten at baseline and excluded gluten on-ED (169% EAR vs 86% EAR) (Appendix I).

Diet quality, as a HEIFA score, was significantly higher on-ED compared to baseline overall (57.9 vs. 53.0). Overall on-ED compared to baseline, the fruit component score was significantly lower (1.3 vs. 2.6), dairy and alternative score was significantly lower (3.3 vs. 4.7) and non-core components score was significantly higher (31.6 vs. 24.6). Mean fruit variety score was low at baseline and on-ED, 0.4 and 0.0 respectively, out of a maximum of 5. The HEIFA sub-score did not differ significantly between baseline and on-ED for vegetables, grains, meat and alternatives, unsaturated fat and fluid (Appendix II). On-ED, gluten-including participants scored significantly higher than gluten-excluding participants for dairy and alternatives (5.1 vs. 2.2).

In paired baseline and on-ED HEIFA scores, total HEIFA score significantly increased in participants who included gluten at baseline and excluded gluten on-ED. In this

group of participants, dairy and alternatives score was significantly lower and non-core components score was significantly higher on-ED (Table 1).

At baseline and on-ED, core ingredients of core foods contributed the most to folate intake. At baseline, core and non-core ingredients contributed to folate in core foods. On-ED, non-core ingredients contributed negligible amounts of folate to core foods. At baseline and on-ED, core and non-core ingredients contributed to folate in non-core foods (Table 2).

At baseline, vegetables were the primary source of folate for participants excluding gluten, and gluten-containing grains was the major folate source for participants including gluten. On-ED, vegetables and gluten-free grains contributed approximately equally as the major sources of folate in gluten-excluding participants. Gluten-containing grains remained the major source of folate for gluten-including participants on-ED, however the proportion of contribution decreased (Table 3).

At baseline and on-ED, when FA-containing supplements were not taken, not all participants had folate intake above the EAR. Gluten-excluding participants who were not taking FA-containing supplements were most at risk of inadequate folate intake with 15 out of 19 (79%) participants not meeting the folate EAR. When FA-containing supplements were included in the nutrient analysis, all participants taking FA-containing supplements exceeded the folate EAR (Figure 2).

Discussion

The current study contributes to the wider literature by investigating and providing preliminary results on the effect of the RPAH ED on folate intake.

On presentation to the Allergy Unit, the majority of patients had already made restrictions to their diet. The most frequently restricted foods correlated with the most frequently reported food triggers which, in agreement with Tripodi, E. (2016; unpublished data), were gluten-containing breads and cereals, vegetables, fruits and dairy products. These are all sources of folate, which may explain the lower mean folate intake of 169% EAR in study participants at baseline compared to 191% EAR in the general Australian population 19 years and over.[11] On presentation to the Allergy Unit, the GIT was the most commonly affected body system. This implies that the majority of patients will further restrict gluten, dairy and soy on-ED and thus have further impact on folate intake.

At baseline the majority of participants had folate intake above the EAR regardless of gluten intake. On-ED overall, mean folate intake was significantly reduced but was still above the EAR which was consistent with previous findings (Nearchou, M. 2015; Tripodi, E. 2016; unpublished data). However, comparison of results from the current study with previous studies (Nearchou, M. 2015; Tripodi, E. 2016; unpublished data) need to be made with caution. It was identified that several items, including gluten-free bread and cereals, in the FoodWorks8 database had incorrect DFE values. The items were found to have a DFE value of a fortified product when the product itself was not fortified. Manufacturers of the products were contacted to confirm any fortification

practices. The identified items were replaced with a closely matched item with a more accurate DFE value which was not done previously.

Gluten-excluding and gluten-including participants were analysed separately in the light that wheat-containing breads and rolls were the major contributor to DFE intake in the general Australian population.[7] The current study has identified that participants excluding gluten on-ED were more than twice as likely to have inadequate folate intake compared with participants including gluten in their diet.

The paired analysis showed that regardless of whether a participant was excluding or including gluten at baseline, the majority of gluten-excluding participants on-ED had inadequate folate intake (Appendix I). Many folate-rich vegetables are not allowed on-ED due to their chemical content. For example, broccoli is a high chemical food and is an exceptional source of folate with half a cup providing over 60% of the EAR for an adult.[12] For participants including gluten at baseline and excluding gluten on-ED, folate intake significantly decreased. This was due to the restriction of folate-rich vegetables as well as fortified breads and cereals.

Studies on coeliac disease patients also found that patients on a gluten-free diet had significantly lower folate intake compared to a gluten-containing diet pre-diagnosis.[3,4] Not only are the majority of patients attending the Allergy Unit required to exclude gluten, the ED restrictions themselves also impose reductions in folate-rich sources. Therefore, tailored clinical recommendations are necessary to ensure patients achieve adequate folate intake from alternative sources.

Gluten-including participants are at lower risk of inadequate folate intake, however, some participants still had inadequate folate intake on-ED. These included participants who included oats in their diet, but restricted all other forms of gluten-containing foods. It also included participants who were limiting gluten but still included small amounts of gluten-containing foods in their diet. Therefore, patients allowed gluten on-ED should also be educated on folate-rich sources allowed on-ED.

The ED is followed for up to 6 weeks before the chemical challenges are commenced. During the challenges, patients are required to consume specified doses of a particular chemical through food and/or capsules whilst maintaining the low chemical diet. Patients are recommended to start with a gluten challenge and if no reactions occur, gluten-containing foods can remain in the diet provided they are low in trigger chemicals. If gluten can remain in the diet, the inclusion of regular breads will decrease the risk of folate inadequacy. However, patients who react to gluten remain at risk of inadequate folate intake. Highly sensitive patients may maintain a gluten-excluding diet after liberalisation. Prolonged inadequate folate intake will cause folate deficiency and the associated complications such as macrocytosis, cardiovascular complications, neural tube defects (NTDs) in developing foetuses and neurological disorders[5,6].
To prevent folate deficiency, which occurs after 3-4 months of inadequate folate intake,[13] it is recommended that patients are educated on adequate folate intake during the initial consultation.

The ED is a restrictive diet, however it is possible to achieve adequate folate intake through food alone, regardless of further food group restrictions. Folate-rich sources

allowed on the low chemical ED include legumes and lentils, Brussels sprout, and quinoa and fortified cereals such as Sanitarium Gluten-Free Weet-Bix and Kellogg's Gluten-Free Special K.[12]

To help patients achieve folate intake above the EAR, it is necessary to educate patients in choosing folate-rich sources allowed on-ED. Also, providing patients with a ready reckoner such as the one in the second volume Allergy Unit handbook[12] could help patients calculate their folate intake. Appendix III shows how an adult aged 19 years and over could achieve folate intake above the EAR.

Patients including wheat-containing bread in their diet on-ED have a reduced risk of inadequate folate intake. One slice of regular bread provides approximately 40% of the EAR for folate[12]. This highlights the importance of folic acid fortification in folate intake in populations with restricted food intake such as patients on the RPAH ED. Currently folate fortification of gluten-free breads is voluntary[14] and very few manufacturers fortify their products. It is therefore a point of consideration to convince more manufacturers to fortify gluten-free breads with folate to provide individuals who exclude gluten an additional folate-rich source.

The current diet quality findings are consistent with previous findings (Nearchou, M. 2015; Tripodi, E. 2016; unpublished data). Total grain score was not significantly different on-ED or between gluten-excluding and gluten-including participants. This was because gluten-free and gluten-containing grains were not differentiated and both contributed towards the total grain score. The dairy and alternatives sub-score was

significantly lower on-ED due to the concomitant dairy and soy exclusion with gluten exclusion. In addition, the consumption of calcium-fortified rice milk on-ED was low. On-ED, patients did not score for fruit variety as only pears were allowed and a minimum of two types of fruits were required for patients to receive a score for fruit variety.

The use of FA-containing supplements enabled all supplementing participants to achieve folate intake above the EAR. Whilst increasing folate intake through food is most ideal, some patients may dislike the recommended foods and will therefore require supplementation as an alternative source of folate. Supplementation would be beneficial for all patients on-ED, but especially for gluten-excluding patients. Supplements that contain folic acid include Blackmores Sustained Release Multi + Antioxidants[15] and Cenovis Once Daily Women's Multi.[16]

Strengths of the current study included having the same patients complete a baseline and on-ED WFR. This enabled paired data analysis which increased the power of the results. Also, WFRs are considered the gold standard in collecting dietary intake.[13] Participants who only completed a baseline WFR were not statistically different to patients who completed a WFR at baseline and on-ED in terms of age, BMI and gender proportion. Selection bias was minimised by including participants who only completed a baseline WFR in the study. The majority of the participants were female which is representative of patients with food intolerance.

The current study did have limitations in that only participants on the low chemical ED were analysed. The majority of patients going on the ED would be advised to follow the low chemical approach, however, due to various reasons such as practicality or less severe symptoms, patients may also choose to follow a less restrictive approach of the diet. Participants on a less restrictive approach were not included in the study due to insufficient sample size. Future studies may consider investigating the folate adequacy of patients following a less restrictive approach of the ED. If sample size remains insufficient, it may be useful to use diet modelling to generate a larger sample size.

Dairy and soy were often restricted together with gluten and would have therefore also contributed the reduced folate intake. This study was therefore unable to assess the impact of gluten-exclusion alone on-ED. However, this is not warranted as very few patients exclude gluten alone on-ED.

Also, the HEIFA tool was validated in a study sample with an age range of 19 to 30 years, however, the current study included participants ranging from 19 to 80 years of age.

Thiamin intake was found to decrease on-ED (Nearchou, M. 2015; Tripodi, E. 2016; unpublished data) and it is also mandatory to fortify wheat flour for bread-making with thiamin.[17] Therefore, future research on the effect of the ED and gluten-exclusion on thiamin adequacy could be considered. Future studies on the RPAH ED need to take caution in entering food items into FoodWorks8 and ensure the nutrient values of the selected item is reflective of the food eaten.

In summary, the current study showed diet quality improved on-ED. Folate intake decreased on-ED and a large proportion of participants on-ED had inadequate folate intake. Participants excluding gluten on-ED were at particularly high risk of inadequate folate intake. Lastly, supplementation reduced the risk of inadequate folate intake in all participants. Therefore, it is recommended that patients going on-ED are educated on folate-rich sources allowed on-ED which include legumes and lentils, Brussels sprouts, quinoa, and fortified cereals (e.g. Gluten-Free Weet-Bix). It is also recommended to encourage patients to take a FA-containing supplement to further reduce the risk of inadequate folate intake. This study has highlighted that fortification of wheat-containing breads provided a valuable source of folate for gluten-including participants. Thus, it could be considered to convince manufacturers of gluten-free bread to fortify their breads with folic acid to provide an additional folate-rich source for patients excluding gluten.

References

1. Swain AR, Soutter VL, Loblay RH. RPAH Elimination Diet Handbook: With Food & Shopping Guide. 2nd ed. Allergy Unit, Royal Prince Alfred Hospital; 2011. 1-134 p.
2. NHMRC. What are Nutrient Reference Values? [Internet]. 2015 [cited 2016 Sep 27]. Available from: https://www.nrv.gov.au/introduction
3. Hallert C, Grant C, Grehn S, Grännö C, Hultén S, Midhagen G, et al. Evidence of poor vitamin status in coeliac patients on a gluten-free diet for 10 years. Aliment Pharmacol Ther. 2002;16(7):1333–9.
4. Shepherd SJ, Gibson PR. Nutritional inadequacies of the gluten-free diet in both recently-diagnosed and long-term patients with coeliac disease. J Hum Nutr Diet. 2013;26(4):349–58.
5. Preedy VR. The Importance of Folate in Health. In: Preedy VR, editor. B Vitamins and Folate Chemistry, Analysis, Function and Effects. The Royal Society of Chemistry; 2013. p. 734–53.
6. Allen LH. Causes of vitamin B12 and folate deficiency. In: Food and Nutrition Bulletin. 2008.
7. FSANZ. Monitoring the Australian population's intake of dietary folic acid before and after mandatory fortification [Internet]. 2016 [cited 2016 Aug 7]. Available from: o http://www.foodstandards.gov.au/consumer/nutrition/folicmandatory/Pages/default.aspx
8. NHMRC. Nutrient Reference Values for Australia and New Zealand - Folate [Internet]. 2014 [cited 2016 Jul 27]. Available from:

https://www.nrv.gov.au/nutrients/folate

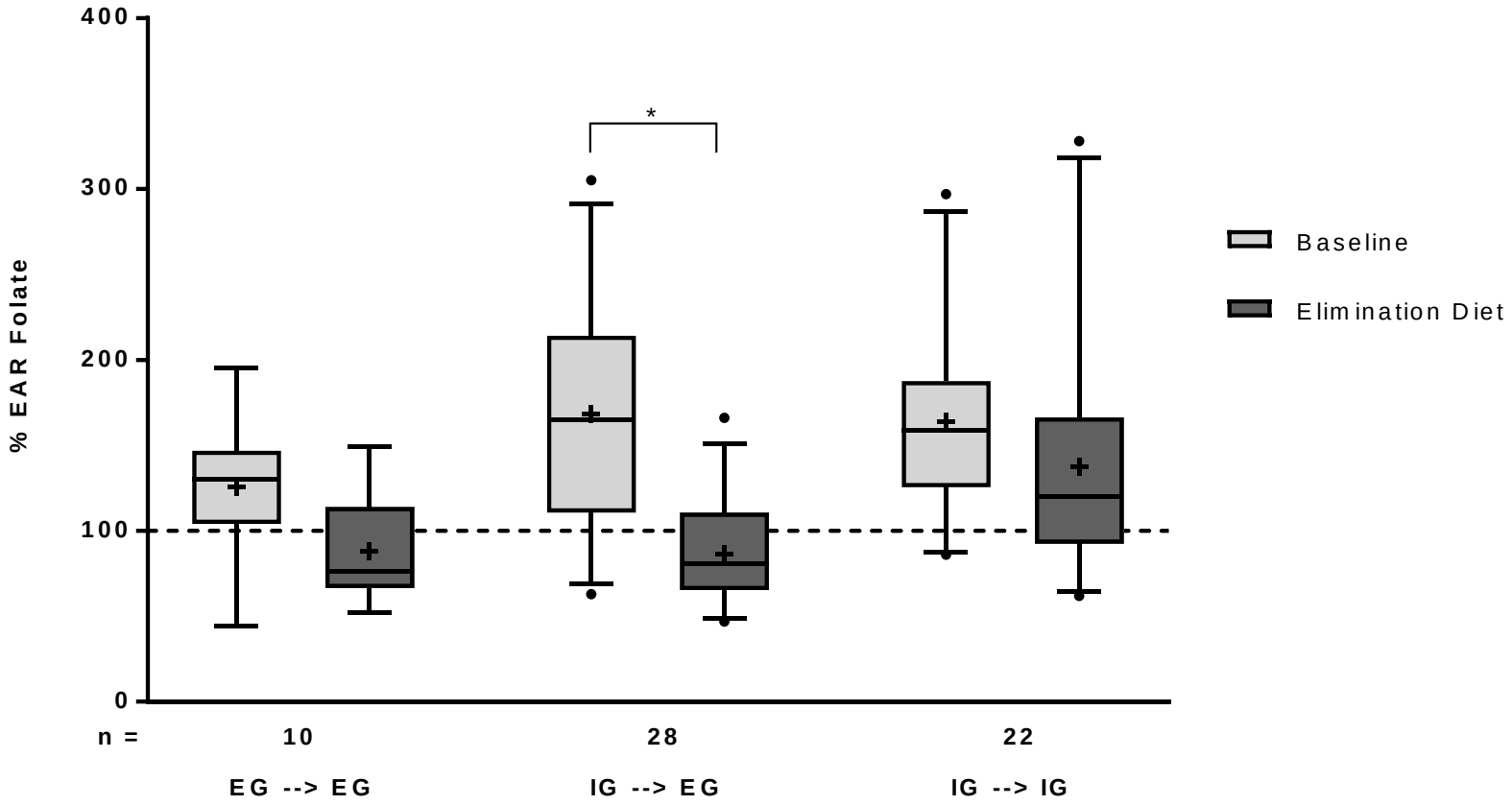
9. ABS. 4363.0.55.001 - Australian Health Survey: Users' Guide, 2011-13 - Discretionary Foods [Internet]. Australian Bureau of Statistics; 2015 [cited 2016 Aug 5]. Available from: http://www.abs.gov.au/AUSSTATS/abs@.nsf/Latestproducts/BA1526F0D19FA21DCA257CD2001CA166?opendocument
10. Roy R, Hebden L, Rangan A, Allman-Farinelli M. The development, application, and validation of a Healthy eating index for Australian Adults (HEIFA-2013). Nutrition [Internet]. Elsevier Inc.; 2016;32(4):432–40. Available from: http://dx.doi.org/10.1016/j.nut.2015.10.006
11. ABS. 4364.0.55.008 - Australian Health Survey: Usual Nutrient Intakes, 2011-12 - Table 3: Vitamins. ABS; 2015.
12. Swain A, Soutter VL, Loblay R. RPAH Food Intolerance Handbook Volume II: Challenges, liberalising & management. Allergy Unit, Royal Prince Alfred Hospital; 2016.
13. Gibson RS. Principles of Nutritional Assessment. 2nd ed. New York: Oxford University Press; 2005.
14. FSANZ. Folic Acid Fortification [Internet]. 2016 [cited 2016 Aug 7]. Available from: http://www.foodstandards.gov.au/consumer/nutrition/folicmandatory/Pages/default.aspx
15. Blackmores. Sustained Release Multi + Antioxidants [Internet]. 2014 [cited 2016 Sep 20]. Available from: https://www.blackmores.com.au/products/sustained-release-multi?gclid=CL-p3LOpndACFZUsvQodMc8G5A

16. Cenovis. Once Daily Women's Multi [Internet]. 2016 [cited 2016 Sep 20]. Available from: https://cenovis.com.au/once-daily-womens-multi/

17. FSANZ. Thiamin Fortification [Internet]. 2015 [cited 2016 Nov 3]. Available from: http://www.foodstandards.gov.au/consumer/nutrition/Pages/Thiamin-fortification-.aspx

18. Swain AR, Soutter VL, Loblay RH. Friendly Food. Murdoch Books; 2014.

Appendices



Appendix I. Paired comparisons of folate intake at baseline and on the Elimination Diet with different changes in gluten intake.
"+" indicates the mean.
Whiskers plotted to the 5th and 95th percentile. EG – excluding gluten, IG – including gluten.
* $p < 0.0001$

Appendix II. Mean (SD) diet quality scores of participants at baseline and on the Elimination Diet based on the Healthy Eating Index for Australian Adults – 2013 (HEIFA)

Food Group	Maximum Score	Baseline			Elimination Diet		
		Total (n = 102)	Excluding Gluten (n = 18)	Including Gluten (n = 84)	Total (n = 61)	Excluding Gluten (n = 38)	Including Gluten (n = 22)
Total HEIFA	100	53.0 (8.1)	56.9 (7.7)	52.2 (8.0)	57.9 (8.7)	56.5 (8.7)	60.2 (8.3)
Total Vegetable HEIFA	10	4.7 (2.5)	6.8 (1.8)	4.3 (2.4)	4.9 (2.5)	4.8 (2.6)	5.1 (2.3)
Vegetable (serves)	5	3.1 (1.5)	4.3 (1.0)	2.9 (1.4)	3.1 (1.6)	3.1 (1.6)	3.2 (1.6)
Vegetable (variety)	5	1.6 (1.1)	2.6 (0.9)	1.4 (1.1)	1.8 (1.0)	1.7 (1.1)	1.9 (0.9)
Total Fruit HEIFA	10	2.6 (2.6)	3.3 (3.0)	2.5 (2.5)	1.3 (1.5)	1.4 (1.5)	1.2 (1.6)
Fruit (serves)	5	2.3 (1.8)	2.8 (1.9)	2.1 (1.7)	1.3 (1.5)	1.4 (1.5)	1.2 (1.6)
Fruit (variety)	5	0.4 (1.4)	0.6 (1.6)	0.4 (1.3)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
Total Grain HEIFA	10	4.4 (2.5)	4.0 (3.0)	4.5 (2.3)	4.2 (2.7)	3.8 (2.5)	4.9 (2.8)
Grains (serves)	5	2.9 (1.4)	2.4 (1.5)	3.0 (1.4)	2.9 (1.4)	2.8 (1.5)	3.0 (1.3)
Wholegrain (serves)	5	1.5 (1.7)	1.6 (1.8)	1.5 (1.6)	1.4 (1.7)	1.0 (1.5)	1.9 (1.9)
Meat alternatives	10	8.5 (2.4)	9.1 (1.9)	8.4 (2.5)	8.4 (2.5)	8.6 (2.4)	8.0 (2.7)
Dairy alternatives	10	4.7 (3.4)	4.2 (3.6)	4.9 (3.3)	3.3 (3.6)	2.2 (3.4)	5.1 (3.4)
Unsaturated Fat	5	2.2 (1.8)	2.8 (1.9)	2.1 (1.8)	3.1 (2.1)	3.4 (2.0)	2.6 (2.2)
Fluid	5	1.2 (1.3)	1.2 (1.2)	1.2 (1.3)	1.1 (1.4)	0.9 (1.3)	1.4 (1.6)
Non-core components ^	40	24.6 (2.0)	25.4 (1.9)	24.4 (2.1)	31.6 (1.9)	31.6 (1.9)	31.7 (1.9)

^ Non-core components is the sum of scores from non-core foods, alcohol, sodium, saturated fat and added sugar

Appendix III. Sample menu plan to meet folate Estimated Average Requirement (EAR) for a gluten-excluding participant aged 19 years or over

Dietary Folate Equivalent EAR = 320 µg[8]			
Food	**Quantity**	**Dietary Folate Equivalents (µg)**	**% EAR**
Breakfast			
Gluten-Free Weet-Bix	2 biscuits	127	40
Rice milk	250 mL	0	0
Canned pear halves in syrup	2 halves	1	0
Morning Tea			
Celery sticks	75g	10	3
Chickpea dip (Friendly Food recipe[18])	40g	12	4
Lunch			
Red kidney beans	0.5 cup	38	12
Steamed potato	1 small potato (100 g)	35	11
Chicken, cooked (skinless)	80 g	18	6
Quinoa, cooked	0.5 cup	35	11
Afternoon Tea			
Rice milk	250 mL	0	0
Boiled egg	1 egg	39	12
Dinner			
Brussels sprout, cooked	75 g	43	13
Brown lentils, cooked	0.5 cup	25	8
Brown rice	0.5 cup	13	4
Lamb, cooked	65 g	11	3
Coffee baked pear (Friendly Food recipe[18])	1 pear	14	4
Total		**407**	**131**

Figure Legends

Figure 1. a) Folate adequacy of all participants at baseline and on the Elimination Diet. b) Folate adequacy of gluten-excluding and gluten-including participants at baseline and on the Elimination Diet.

"+" indicates the mean.

* $p < 0.0001$, ** $p = 0.003$. Whiskers plotted to the 5^{th} and 95^{th} percentiles.

Figure 2. Effect of folic acid-containing supplements on folate intake.

"+" indicates the mean.

* $p = 0.02$, ** $p = 0.001$, *** $p < 0.0001$. Whiskers plotted to the 5^{th} and 95^{th} percentile.

Table 1. Mean (SD) of paired Healthy Eating Index for Australian Adults – 2013 (HEIFA) sub-scores of participants at baseline and on the Elimination Diet (ED) with different changes in gluten intake

		Baseline	ED	Baseline	ED	Baseline	ED
	Gluten Intake [(b)]	X	X	✓	X	✓	✓
Food Group	Maximum Score						
Total HEIFA	100	54.9 (5.7)	56.1 (6.8)	49.5 (6.9)	56.7 (9.4)*	56.3 (8.0)	60.0 (8.4)
Total Vegetable HEIFA	10	6.2 (1.9)	4.2 (2.9)	4.3 (2.0)	4.9 (2.4)	4.3 (2.7)	5.0 (2.4)
Vegetable (serves)	5	3.8 (1.1)	2.7 (1.9)	2.9 (1.2)	3.1 (1.5)	2.9 (1.7)	3.2 (1.6)
Vegetable (variety)	5	2.1 (0.9)	1.5 (1.1)	1.3 (0.9)	1.8 (1.0)	1.4 (1.1)	1.9 (0.9)
Total Fruit HEIFA	10	2.5 (3.0)	1.4 (2.0)	2.4 (2.0)	1.4 (1.3)	2.7 (2.8)	1.3 (1.6)
Fruit (serves)	5	2.1 (1.8)	1.4 (2.0)	2.2 (1.7)	1.4 (1.3)	2.3 (1.8)	1.3 (1.6)
Fruit (variety)	5	0.6 (1.6)	0.0 (0.0)	0.2 (0.9)	0.0 (0.0)	0.5 (1.5)	0.0 (0.0)
Total Grain HEIFA	10	4.3 (3.3)	3.9 (2.9)	3.7 (2.3)	3.7 (2.5)	5.9 (2.0)	5.0 (2.8)
Grains (serves)	5	2.5 (1.5)	2.9 (1.6)	2.5 (1.5)	2.7 (1.5)	3.8 (1.2)	3.0 (1.3)
Wholegrain (serves)	5	1.9 (2.1)	1.0 (1.5)	1.2 (1.4)	1.0 (1.5)	2.1 (1.6)	2.0 (1.9)
Meat and alternatives	10	9.2 (1.7)	8.8 (2.2)	8.3 (2.5)	8.4 (2.5)	8.3 (2.9)	8.0 (2.7)
Dairy and alternatives	10	4.6 (4.0)	1.6 (3.5)	4.5 (2.9)	2.4 (3.4)*	6.1 (3.7)	5.1 (3.4)
Unsaturated Fat	5	3.3 (1.9)	3.3 (2.4)	2.1 (1.9)	3.4 (1.9)	2.0 (1.8)	2.6 (2.2)
Fluid	5	1.4 (1.3)	1.2 (1.5)	1.0 (1.3)	0.9 (1.3)	1.6 (1.3)	1.5 (1.6)
Non-core components [(a)]	40	23.5 (6.8)	31.8 (5.0)*	23.3 (6.3)	31.5 (7.3)**	25.5 (7.6)	31.5 (5.4)*

[(a)] Non-core components is the sum of scores from non-core foods, alcohol, sodium, saturated fat and added sugar

[(b)] Gluten intake: X = excluding gluten, ✓ = including gluten

* $p < 0.05$, ** $p < 0.0001$

Table 2. Contribution of core and non-core food and ingredients to mean folate intake (%)

	Baseline (n = 102)	**Elimination Diet (n = 61)**
Mean Folate Intake (% EAR (SD))	168 (59)	108 (54)
Core Foods	89	90
Core Ingredients of Core Foods	85	90
Non-Core Ingredients of Core Foods	4	0
Non-Core Foods	11	10
Core Ingredients of Non-Core Foods	7	5
Non-Core Ingredients of Non-Core Foods	4	5

Table 3. Food group contributions to folate intake from core ingredients of core foods in gluten-excluding and gluten-including participants

Food Group	Gluten-Excluding				Gluten-Including			
	n	Average Serves /person /day	Average DFE (μg) /person /day	% EAR	n	Average Serves /person /day	Average DFE (μg) /person /day	% EAR
Baseline								
	n = 18				**n = 84**			
Grains								
Gluten-containing grains	0	-	-	-	83	2.5	230	72
Gluten-free grains	16	2.3	55	17	65	1.5	25	8
Vegetables	18	5.5	148	46	84	3.4	90	28
Meat and alternatives	18	1.8	48	15	82	1.5	43	13
Dairy and alternatives	16	1.2	40	13	81	1.3	45	14
Fruit	17	1.4	65	20	79	0.9	36	11
Elimination Diet								
	n = 38				**n = 23**			
Grains								
Gluten-containing grains	0	-	-	-	22	2	160	50
Gluten-free grains	38	3.3	83	26	21	1.8	58	18
Vegetables	38	3.6	82	26	23	3.8	94	29
Meat and alternatives	37	1.8	58	18	23	1.7	51	16
Dairy and alternatives	11	1.3	67	21	21	1.7	60	19
Fruit	24	0.8	5	2	17	0.9	7	2

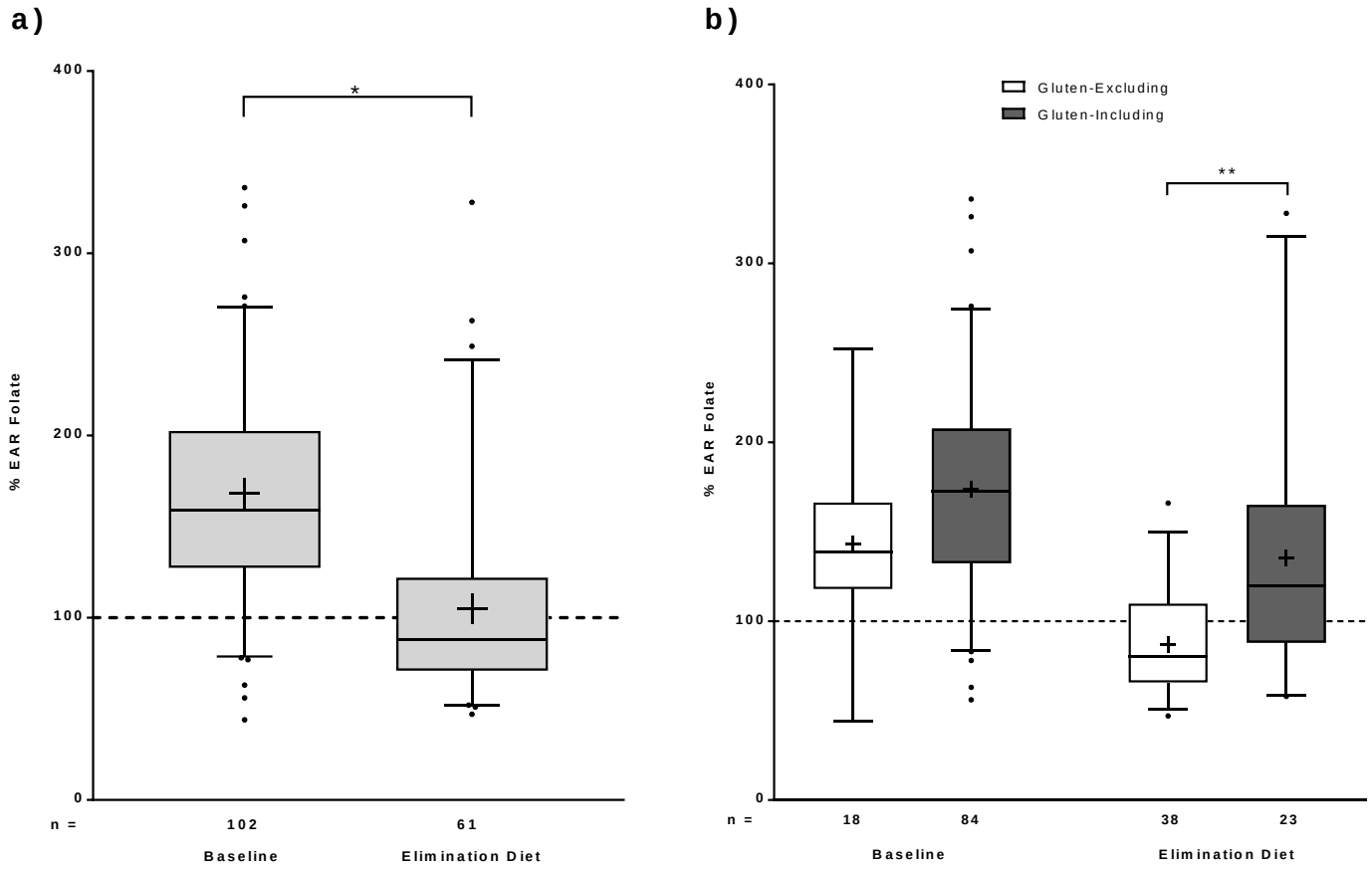


Figure 1. a) Folate adequacy of all participants at baseline and on the Elimination Diet. b) Folate adequacy of gluten-excluding and gluten-including participants at baseline and on the Elimination Diet.

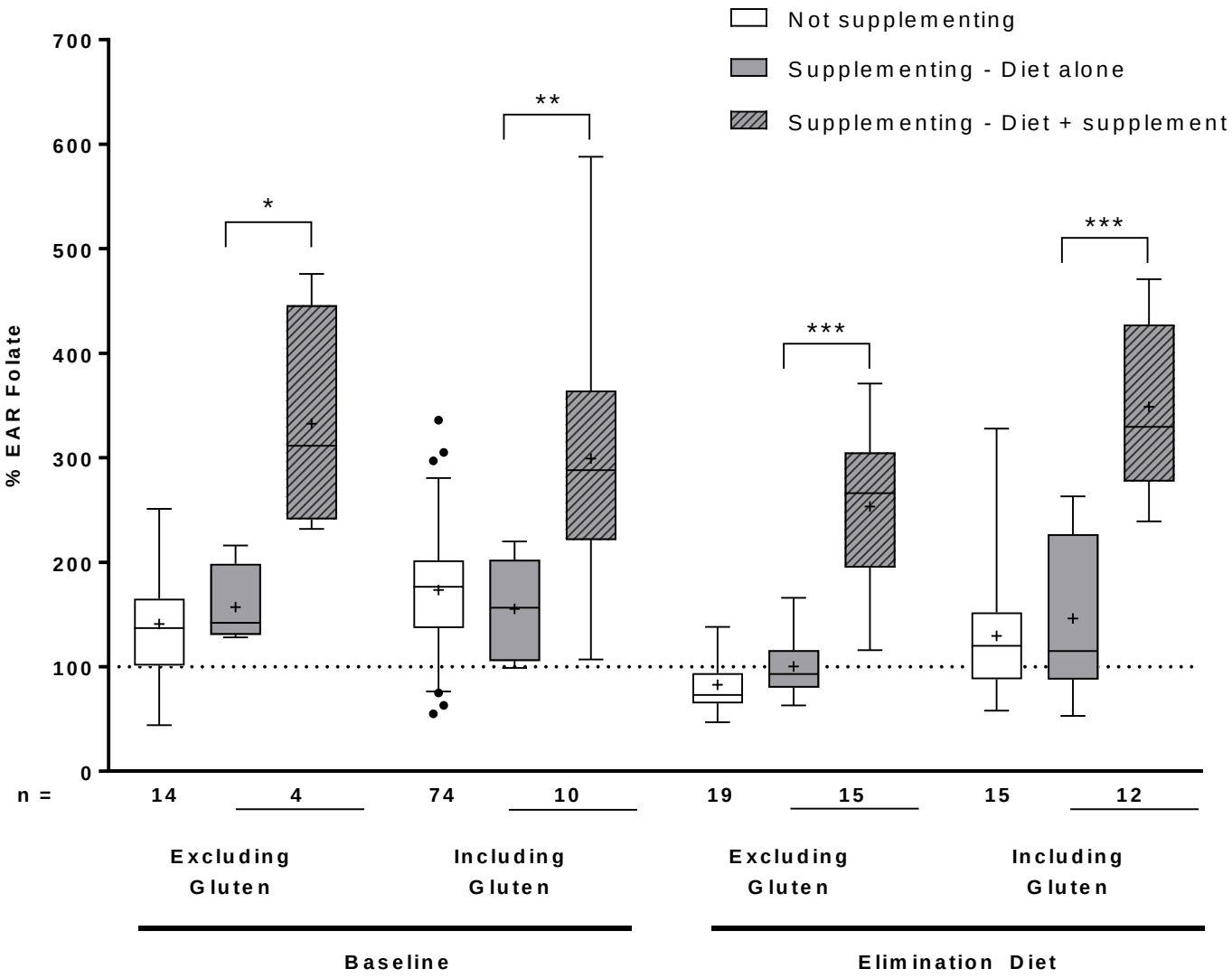


Figure 2. Effect of folic acid-containing supplements on folate intake.