

ULRR

Randomization to 6-month Mediterranean diet compared with a low-fat diet leads to improvement in dietary inflammatory index scores in patients with coronary heart disease: the AUSMED Heart Trial

Item Type	Article
Authors	Mayr, Hannah L.;Thomas, Colleen J.;Tierney, Audrey;Kucianski, Teagan;George, Elena S.;Ruiz-Canela, Miguel;Hebert, James R.;Shivappa, Nitin;Itsiopoulos, Catherine
Citation	Nutrition Research;55, pp. 97-107
Publisher	Elsevier
Download date	2026-03-14 09:14:10
Item License	https://creativecommons.org/licenses/by-nc-sa/1.0/
Link to Item	https://hdl.handle.net/10344/6921

Accepted Manuscript

Randomization to 6-month Mediterranean diet compared with a low-fat diet leads to improvement in Dietary Inflammatory Index scores in patients with coronary heart disease: the AUSMED Heart Trial

Hannah L Mayr, Colleen J Thomas, Audrey C Tierney, Teagan Kucianski, Elena S George, Miguel Ruiz-Canela, James R. Hebert, Nitin Shivappa, Catherine Itsiopoulos

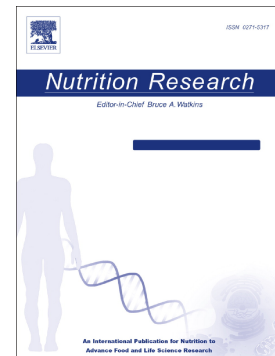
PII: S0271-5317(17)31105-3
DOI: [doi:10.1016/j.nutres.2018.04.006](https://doi.org/10.1016/j.nutres.2018.04.006)
Reference: NTR 7879

To appear in:

Received date: 30 November 2017
Revised date: 20 March 2018
Accepted date: 10 April 2018

Please cite this article as: Hannah L Mayr, Colleen J Thomas, Audrey C Tierney, Teagan Kucianski, Elena S George, Miguel Ruiz-Canela, James R. Hebert, Nitin Shivappa, Catherine Itsiopoulos, Randomization to 6-month Mediterranean diet compared with a low-fat diet leads to improvement in Dietary Inflammatory Index scores in patients with coronary heart disease: the AUSMED Heart Trial. The address for the corresponding author was captured as affiliation for all authors. Please check if appropriate. Ntr(2018), doi:[10.1016/j.nutres.2018.04.006](https://doi.org/10.1016/j.nutres.2018.04.006)

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.



Randomization to 6-month Mediterranean diet compared with a low-fat diet leads to improvement in Dietary Inflammatory Index scores in patients with coronary heart disease: the AUSMED Heart Trial

Hannah L Mayr^{a,b}, Colleen J Thomas^c, Audrey C Tierney^{a,d}, Teagan Kucianski^a, Elena S George^{a,e}, Miguel Ruiz-Canela^{f,g}, James R. Hebert^{h,i}, Nitin Shivappa^{h,i}, Catherine Itsiopoulos^a.

^aDepartment of Rehabilitation, Nutrition and Sport, School of Allied Health, La Trobe University, Melbourne, Victoria, Australia, 3086. ^bDepartment of Nutrition and Dietetics, Northern Health, Melbourne, Victoria, Australia, 3076. ^cDepartment of Physiology, Anatomy and Microbiology, School of Life Sciences, La Trobe University, Melbourne, Victoria, Australia, 3086. ^dDepartment of Clinical Therapies, University of Limerick, Limerick, V94 T9PX, Ireland. ^eSchool of Exercise and Nutrition Sciences, Deakin University, Geelong, Victoria, Australia. ^fDepartment of Preventive Medicine and Public Health, University of Navarra, Pamplona, Spain. ^gCIBER Fisiopatología de la Obesidad y Nutrición (CIBERobn), Madrid, Spain. ^hCancer Prevention and Control Program and Department of Epidemiology and Biostatistics, Arnold School of Public Health, University of South Carolina, Columbia, SC 29208 USA. ⁱConnecting Health Innovations LLC, Columbia, SC 29201 USA.

Email contacts: HLM: h.mayr@latrobe.edu.au, CJT: colleen.thomas@latrobe.edu.au, ACT: A.Tierney@latrobe.edu.au, TK: T.Kucianski@latrobe.edu.au; EG: elena.george@deakin.edu.au; MRC: mcanela@unav.es, JRH: jhebert@mailbox.sc.edu; NS: shivappa@mailbox.sc.edu, CI: C.Itsiopoulos@latrobe.edu.au.

Corresponding author: colleen.thomas@latrobe.edu.au

Department of Physiology, Anatomy and Microbiology, School of Life Sciences, La Trobe University, Melbourne, Victoria, Australia, 3086

Abbreviations:

DII; dietary inflammatory index
CHD; coronary heart disease
MedDiet; Mediterranean diet
CRP; C-reactive protein
TNF- α ; Tumor necrosis factor- α
IL; interleukin
CONSORT; Consolidated Standards of Reporting Trials
AMI; acute myocardial infarction
MUFA; monounsaturated fatty acids
PUFA; polyunsaturated fatty acids
EVOO; extra virgin olive oil
MVPA; moderate-to-vigorous physical activity
MEDAS; Mediterranean Diet Adherence Screener
Hs; high sensitivity
ELISA; enzyme-linked immunosorbent assay
SD; standard deviation
IQR; interquartile range
ANCOVA; Analysis of covariance
CI; confidence interval
RE; retinol equivalents

Abstract

A higher dietary inflammatory index (DII[®]) score is associated with inflammation and incidence of coronary heart disease (CHD). We hypothesized that a Mediterranean diet (MedDiet) intervention would reduce DII score. We assessed dietary data from a randomized controlled trial comparing 6-month MedDiet versus low-fat diet intervention, in patients with CHD. We aimed to determine the DII scores of the prescribed diets' model meal plans, followed by whether dietary intervention led to lower (i.e., more anti-inflammatory) DII scores and consequently lower high sensitivity C-reactive protein (hs-CRP) and interleukin-6 (hs-IL-6). DII scores were calculated from 7-day food diaries. The MedDiet meal plan had a markedly lower DII score than the low-fat diet meal plan (-4.55 vs. -0.33, respectively). In 56 participants who completed the trial (84% male, mean age 62±9 years), the MedDiet group significantly reduced DII scores at 6-months (n=27; -0.40±3.14 to -1.74±2.81, p=0.008) and the low-fat diet group did not change (n=29; -0.17±2.27 to 0.05±1.89, p=0.65). There was a significant post-intervention adjusted difference in DII score between groups (compared to low-fat, MedDiet decreased by -1.69 DII points; p=0.004). When compared to the low-fat diet, the MedDiet non-significantly reduced hs-IL-6 (-0.32pg/mL, p=0.29) and increased hs-CRP (+0.09mg/L, p=0.84). These findings demonstrated that MedDiet intervention significantly reduced DII scores compared to a low-fat diet. However, in this small cohort of patients with CHD this did not translate to a significant improvement in measured inflammatory markers. The effect of improvement in DII with MedDiet should be tested in larger intervention trials and observational cohorts.

Keywords: Dietary inflammatory index; Mediterranean diet; low-fat diet, inflammation; coronary disease; randomized controlled trial.

1. Introduction

Chronic, low-grade systemic inflammation is a recognized risk factor for development of chronic diseases, including coronary heart disease (CHD)[1]. Dietary intakes have been shown to affect inflammation via both pro-inflammatory and anti-inflammatory mechanisms[2]. Healthy dietary patterns, which focus on foods rather than single nutrients, have been shown to be anti-inflammatory[3, 4]. Among these, the Mediterranean diet (MedDiet) has the strongest evidence for improvement in markers of inflammation[5, 6]. However, the effect of the MedDiet on biomarkers of inflammation is unclear in patients with CHD[7].

The Dietary Inflammatory Index (DII[®]) is a literature-derived dietary score which incorporates 45 nutritional factors that are known to modulate inflammatory markers, in either a pro- or anti-inflammatory manner[8]. This method of characterizing diet differs from other dietary pattern adherence scores as it was formulated based on findings evident in the literature relating diet to inflammatory cytokine signaling pathways. This is fundamentally different than patterns of food intake that are associated with a particular set of dietary recommendations (e.g., the Alternative Healthy Eating Index in relation to the American Food Pyramid)[9] or a culinary tradition (e.g., the MedDiet)[10]. Since its development, the DII has been applied to diet intake data in a variety of study cohorts to indicate associations with cardiovascular risk factors and CHD[11]. A higher (more pro-inflammatory) DII score has been associated with the inflammatory markers C-reactive protein (CRP)[12], tumor necrosis factor-alpha (TNF- α)[13], and interleukin (IL)-6[14], incidence of metabolic syndrome, blood pressure and triglycerides[15], obesity[16], and greater incidence of

cardiovascular events[17-19]. The DII offers an attractive alternative assessment tool to laboratory-based measurement of inflammatory cytokines, which are obtained through invasive means, tend to be costly, and are not routinely available in existing studies.

The DII also provides a method by which established dietary patterns can be compared for their probable anti-inflammatory effect. For instance, a one-day meal plan of a traditional MedDiet had a strong anti-inflammatory DII score[20] and greater MedDiet adherence has been negatively associated with DII scores[16, 21]. The DII also has been demonstrated to be inversely associated with healthy eating scores based on American dietary guidelines[9].

An improvement in DII score can therefore be achieved through healthy diet intervention, which in turn could reduce inflammation and other cardiometabolic risk markers, as well as prevent CHD. To our knowledge only two studies have assessed the impact of diet intervention on the DII. One of these trials showed short (2-month), but not longer-term (6-month), improvement in DII with vegetarian compared to omnivorous diets in a small cohort (n=63) of overweight adults[22]. The other study of a large cohort (n=14, 339) of post-menopausal women reported modest improvement in DII after 6-years on a low-fat diet[23]. Neither of these studies, however, assessed whether the reduction in DII score achieved with improved diet led to a change in inflammatory markers.

Current recommendations in Australia promote a low-fat diet for the prevention of CHD[24], for which there is some evidence to suggest this type of diet is associated with reduced DII scores[9]. A MedDiet, by nature of its composition, is likely to lead to a more anti-inflammatory DII score. The impact of adherence to a MedDiet on DII scores is of interest both because of the popularity of Mediterranean cuisine and concern about chronic, low-

grade, systemic inflammation. Therefore, our objectives were to determine (1) how the DII score of a MedDiet compared with that of a low-fat diet, (2) the impact of randomization to an intervention with these two diets for 6-months on change in DII score and CRP and IL-6 (two inflammatory biomarkers on which the DII development was based)[8], and (3) the association between improvement in MedDiet adherence score and DII score, in an adult population with CHD. We hypothesized that the prescribed MedDiet model would have a lower DII score (i.e., greater dietary anti-inflammatory potential) than that of the low-fat diet, and that this would translate to greater improvement in DII score and inflammation in participants randomized to the MedDiet intervention. We also hypothesized an association between improvement in MedDiet adherence and DII scores.

2. Methods and Materials

2.1. Study design

The AUStralian MEDiterranean Diet Heart Trial (AUSMED Heart Trial) is a multicenter, parallel design, randomized controlled trial (RCT) for the secondary prevention of CHD in a multi-ethnic Australian population (Australia and New Zealand Clinical Trials Register: ACTRN12616000156482, <http://www.anzctr.org.au/>). The trial involves 6-month intervention with a MedDiet versus low-fat diet and a 12-month follow up to assess the primary outcome of aggregate cardiovascular events. The present study investigated the 6-month effect of two dietary interventions (MedDiet and low-fat diet) and the impact that MedDiet adherence had on DII scores (i.e., inflammatory potential of the diet), as well as the individual effects of the diets on CRP and IL-6 in a pilot cohort of patients with CHD.

2.2. Participants and recruitment

Between 2014 to 2016, patients were recruited from two teaching hospitals in Melbourne, Australia. The study is being conducted in accordance with the CONSORT guidelines[25] and the guidelines laid down in the Declaration of Helsinki[26]. All procedures involving patients were approved by the Human Research Ethics Committees of La Trobe University, the Northern Hospital and St Vincent's Hospital Melbourne, with written informed consent obtained from all enrolled participants.

Eligible patients were adults with CHD, able to read and write in English, and who had experienced at least one of the following: acute myocardial infarction (AMI); angina pectoris with documented coronary artery disease on imaging; coronary artery bypass grafting; or percutaneous coronary intervention. Exclusion criteria included: malignant tumor, symptomatic chronic heart failure (New York Heart Association Functional Classification II, III & IV[27]), chronic inflammatory disease requiring immunosuppressant or non-steroidal anti-inflammatory drugs, chronic kidney disease stage 3 or above[28], decompensated liver disease (or taking medications that cause hepatosteatosis), pregnancy or breastfeeding, history of allergy to olive oil or nuts, or current participation in a lifestyle program, drug or supplement trial. Patients were approached at outpatient, rehabilitation or inpatient cardiac settings. Eligible and interested patients attended a pre-baseline appointment where the study was explained in detail, forms and equipment were provided, consent was obtained and randomization was conducted.

2.3. Randomization and interventions

Enrolled participants were randomly assigned in a 1:1 ratio to the MedDiet group or the low-fat diet group. Randomization tables were developed by the trial statistician using a computer generated stratified approach based on sex, age (<55, 55 to 65 and >65 years) and history of AMI (yes/no). Baseline, 3-month and 6-month face-to-face appointments were conducted to obtain anthropometry, biomarkers and dietary data and for counselling with the dietitian. Five short phone reviews for follow-up dietary counselling with the dietitian also occurred across the 6-months, at weeks 3, 6, and 9 and months 4 and 5. Telephone-delivered lifestyle advice previously has been demonstrated to be effective for reducing coronary risk factors in the Australian setting and is a low-cost intervention method[29]. Different dietitians worked with the two different diet study groups to prevent contamination. Consultation frequency and data collection time points were consistent across the two intervention groups. All participants continued to receive standard medical care provided at their respective hospital or primary care settings; however, they were instructed not to engage in any nutritional interventions, including cardiac rehabilitation or other research trials for the duration of this study.

2.3.1. Mediterranean diet

The rationale and development of the MedDiet intervention has been explained in detail elsewhere (George, et al., 2018, manuscript under preparation). It was designed based on the principles of the traditional Cretan MedDiet[30], including information from seminal intervention trials conducted in Mediterranean populations[31, 32], trials conducted in Australia where MedDiet foods and meals were provided[33, 34] and the Hellenic dietary guidelines[35]. The diet was modelled via a 2-week meal plan incorporating key dietary components of a MedDiet and a mix of traditional and modified recipes considered to be realistic options for multi-ethnic Australians. In terms of contribution to total energy

consumption, target macronutrient intakes were 42% total fat (of which at least 50% was from monounsaturated fatty acids [MUFA] and 25% from polyunsaturated fatty acids [PUFA]), <10% saturated fatty acids, 35% carbohydrate, 15% protein, and $\leq 5\%$ alcohol.

Participants were provided with the 2-week model meal plan (Table S1 in Supplemental Materials provides an example of one day), the MedDiet resource kit which is to be published in George, et al., 2018 (manuscript under preparation), an associated recipe book, The Mediterranean Diet by Itsiopoulos (2013) (ISBN 9781742610825), and shopping list. Other resources provided included a food pyramid, weekly food intake checklist and label reading information. Food group recommendations included: daily intake of extra virgin olive oil (EVOO), nuts, vegetables, fruit and wholegrain cereals, regular intake of legumes, fish and yoghurt, and limited intake of commercial sweets or pastries and red or processed meat. Poultry, eggs and feta cheese were recommended in moderation. For participants choosing to consume alcohol, red wine was suggested to be consumed in moderation (1-2 standard glasses) with meals. To facilitate dietary compliance and to encourage intake of staple Mediterranean foods less familiar to this population, a hamper was provided to participants at baseline and 3-months. Each hamper included 6L EVOO (to achieve 60-80mL/day) and 1.2kg nuts (almonds, walnuts and hazelnuts to achieve 30g/day) as well as samples of tinned tuna and salmon, canned legumes and Greek yoghurt.

2.3.2. *Low-fat diet*

Participants in the low-fat diet group were instructed to follow the standard diet recommendations provided to cardiac patients in Australia at the time this study was developed (in 2014), with a matched counselling frequency and set-up to that of the MedDiet

group. Recommendations from the Australian National Heart Foundation as well as the Australian Dietary Guidelines and Nutrient Reference Values were consulted for design of the low-fat diet[24, 36, 37]. In terms of contribution to total energy consumption, target macronutrient intakes were <30% total fat, <7% saturated fat, 45-65% carbohydrate, 15-25% protein and \leq 5% alcohol. Food group recommendations included daily intake of grains and cereals (mostly whole grains, 5-7 serves/day), vegetables (5-6 serves/day), fruit (2 serves/day), protein foods (2-3 serves/day) and low-fat dairy foods (2 serves/day)[36]. Based on macronutrient and food group targets, a 1-week meal plan was created to model a comparative nutrient profile for this diet and to generate a resource for participants (Table S1 in Supplemental Materials provides an example of one day). Resources for label reading, low-fat cooking and recommended daily food group serves also were provided. To aid compliance and encourage their continuation in the trial, participants were provided with a supermarket voucher at their three face-to-face appointments.

In both study groups the dietary advice was tailored to each individual through client-centered counselling and goal setting with the dietitian[38, 39]. Both diets were prescribed *ad libitum* to achieve weight maintenance with no specific recommendations on energy restriction. There was some cross-over in the general food group recommendations between the low-fat diet and the MedDiet, including daily intake of vegetables, fruit and wholegrain cereals and limited processed foods. However, the MedDiet specifically included promotion of EVOO, nuts, leafy greens, tomatoes, onion, garlic, legumes, oily fish and fermented dairy foods and more significantly reduced intake of red meat, which were not key components of the low-fat diet[40].

2.4. Measurements

This study reports only on baseline and 6-month data measurements. Medical conditions related to eligibility were collected in the screening process using hospital records and by questionnaire at the pre-baseline appointment. Sociodemographic and clinical characteristics were collected through a self-report survey completed at baseline, which included details on type, dose and frequency of medication and supplement use. Exercise was not a target of this intervention, however physical activity levels were assessed by triaxial Actigraph accelerometer (WGT3X-BT; Actigraph Corp, Florida, United States) worn by participants 7 days prior to each face-to-face appointment. Time spent in moderate-to-vigorous physical activity (MVPA) minutes per week was determined based on established criteria[41]. Weight and height were measured, after an 8-h fast, using international standards for anthropometric assessment[42] by trained research personnel. Body weight was measured to the nearest 0.1kg using calibrated digital scales, without shoes and after removal of heavy jewelry, outer layers of clothing and pocket contents. Height was measured to the nearest 0.1cm, while barefoot using a wall-mounted stadiometer. Body mass index was calculated as weight (kg) / height (m²) and overweight was classified as >25 kg/m²[43]. A fasting blood sample was taken from the antecubital vein using standard venous puncture techniques. All blood samples were processed immediately and aliquots were stored at -80°C until assay. Serum high sensitivity (hs)-CRP levels were measured at a commercial laboratory (Dorevitch Pathology Pty Ltd, Heidelberg, Australia) by chemical analyzer (Cobas Integra 400, Roche). Serum hs-IL-6 levels were measured by enzyme-linked immunosorbent assay (ELISA) (Abcam, #ab46042, detection sensitivity <0.81 pg/mL) in duplicate.

2.4.1. Dietary assessment

Participants completed a 7-day food diary in household measures the week prior to the baseline and 6-month appointments. The diary included type, brand, quantity and cooking methods for consumed foods, with missing details clarified by the dietitian at the appointment. All food diaries were entered into FoodWorks8[®] (Xyris software Australia Pty Ltd), using the most up-to-date databases *AUSNUT 2013*, *AusBrands 2015* and *AusFoods 2015* for nutrient intake analyses. Any day recorded in the food diary that was considered to represent a highly abnormal intake for the participant was excluded from entry into FoodWorks8. This exclusion was applied to only two participants for reasons of religious fasting (three days) and nil intake due to acute illness (one day). The 2-week MedDiet and 1-week low-fat diet meal plans were designed against the desirable macronutrient profile using FoodWorks8 analysis. The 14-point Mediterranean Diet Adherence Screener (MEDAS), generated and validated for the PREDIMED study[44], was measured at baseline and 6-months for each participant. Dietitians assisted participants to complete the MEDAS during consults for the MedDiet group and calculated the score retrospectively for the low-fat diet group using 7-day food diary data in order to avoid contamination with the MedDiet principles. A higher score is reflective of better adherence to a traditional MedDiet pattern, with a score of 9 or above out of 14 considered an acceptable adherence criterion[45].

2.4.2. *Dietary Inflammatory Index (DII[®])*

The development[8] and validation[12] of the DII has been described elsewhere. Through evaluation of peer-reviewed literature published between 1950 and 2010, the DII score is based on 1943 articles that examined the association between inflammation and 45 individual nutrient, food or flavonoid intake parameters (listed in Table 2). Points were assigned to each of these parameters according to whether: 1) they increased (+1), decreased (-1) or had no (0)

effect on the four established pro-inflammatory biomarkers; IL-1 β , IL-6, TNF- α and CRP or 2) decreased (+1), increased (-1) or had no (0) effect on the two established anti-inflammatory biomarkers; IL-4 and IL-10. The score for each of the food parameters was weighted according to the study design employed and total number of research articles. Overall parameter-specific inflammatory effect scores were then calculated based on the ratio of the total weighted number of articles to the weighted pro- and anti-inflammatory articles for each parameter followed by subtracting the anti- from the pro-inflammatory fraction. Parameters which had a robust pool of literature, i.e. greater than the median number of 236 weighted articles, were assigned the full value of that score. Parameters with the total article weight less than 236 were adjusted by dividing it by 236 and then multiplying this fraction by the previously defined inflammatory effect score.

In this study, the DII was computed twice in the participants, at baseline and after 6-months of intervention. Our assessment of the DII included all 45 of its parameters. Dietary intake data were adjusted against a reference global daily mean and standard deviation intake for each parameter to obtain a Z-score. The global intake data was based on consumption data from 11 countries around the world. To further reduce the effective skewness, each Z score was converted to a proportion (i.e., values ranging from 0 to 1) and this value was centered on zero by multiplying by 2 and subtracting 1. This centered proportion score for each intake parameter was multiplied by its respective parameter-specific inflammatory effect score to obtain the parameter-specific DII score. Each of these 45 scores were then summed to obtain an overall DII score for each individual and meal plan. Finally, the DII score for participant data was re-calculated with the inclusion of nutrient supplement intake (DII_{diet+supplements}).

The intake values for most of the DII parameters (energy, protein, carbohydrate, total fat, MUFA, PUFA, omega-3, omega-6, saturated fatty acids, trans fat, cholesterol, fiber, alcohol, caffeine, folate, beta carotene, vitamin A, vitamin B6, vitamin B12, vitamin C, vitamin E, iron, magnesium, niacin, riboflavin, selenium, thiamin and zinc) were obtained from FoodWorks8 nutrient analyses of the food diaries or meal plans. For the omega-6 component only intake of linoleic acid was available. Isolated food components in the DII (green/black tea, garlic, ginger, onion, pepper, rosemary, saffron, turmeric and thyme/oregano) were extracted from the food diaries and meal plans and the total daily intake in grams calculated. Vitamin D was calculated from the 7-day food diaries and meal plans using an electronic Australian nutrient table database[46]. Intake of eugenol was calculated based on the recognized content in cloves in *Phenol Explorer*[47]. For calculation of flavonoids (flavan-3-ol, flavones, flavonols, flavonones, anthocyanidins and isoflavones) the USDA Databases for the Flavonoid Content of Selected Foods (Release 3.2, November 2015) and Isoflavone Content of Selected Foods (Release 2.0, September 2008) were used as these databases had a wide variety of foods, including data on raw, cooked or processed options.

2.5. Statistical analyses

Participants who did not complete the intervention were excluded from analyses. As this study represented an exploratory analysis of a pilot cohort, a power calculation was not performed[48]; however, literature was consulted to determine if a significant effect on DII could be expected in the current sample. Because randomization to a vegetarian diet compared to a healthy omnivorous diet (total n=26) resulted in a significant improvement in DII (mean change of -1.4 , $p < 0.05$) after 2-months[22], it was therefore expected that our MedDiet derived from traditional Cretan (Greek Mediterranean) principles would be

associated with even lower DII scores[20] and an even greater reduction in DII than a low-fat diet in our participant sample (total n=56) after 6-months.

Data are presented as means \pm standard deviation (SD) or standard error (SEM), medians (interquartile range [IQR]) or n (%) as appropriate. The Kolmogorov–Smirnov test was applied to assess the normality of continuous variables. According to this, an Independent Student’s T-test or non-parametric Mann-Whitney U test was used to compare continuous variables. Categorical variables were compared using the *Chi-square* test. McNemar’s test assessed the differences in percentage adherence to each component of the MEDAS between baseline and 6-months within each study group. Analysis of covariance (ANCOVA) with baseline values as a covariate were performed to assess differences for change in MEDAS, DII scores and inflammatory biomarkers between diet study groups, participant tertiles of change in MEDAS and adherence to MEDAS components at 6-months. Changes in biomarkers also included baseline DII score as a covariate in the ANCOVA models. For all analyses based on hs-CRP, participants with serum levels $>10\text{mg/L}$ were excluded, as these higher concentrations reflect acute rather than chronic inflammation[49]. Pearson or Spearman rho correlations were conducted between the MEDAS scores (independent variable) and DII scores (dependent variable) at baseline, 6-months and for change variables with adjustment for diet intervention group. *R*-values were classified as weak (0.10 to 0.29), moderate (0.30 to 0.49) or strong (0.50 to 1.00) correlations[50]. Statistical significance was set at $p<0.05$, except when multiple *post-hoc* comparisons were performed within or between study groups, in which case a Bonferroni correction was applied. For between- and within-group comparisons, 95% confidence intervals (CIs) were calculated. All statistical analyses were conducted in SPSS[®] statistical package version 23 (IBM Corp, Released 2015).

3. Results

3.1. Participants

Figure 1 demonstrates the randomization to diet study groups and completion of study appointments. Of the 35 participants randomized to the low-fat diet group, 31 started and 29 completed the intervention. Of the 37 participants randomized to the MedDiet group, 35 started and 27 completed the intervention. Participants dropped out for medical or family related issues or were lost to follow up. The drop-outs had a significantly higher pro-inflammatory DII score, with a mean baseline DII score of 1.76 ± 2.3 compared to -0.28 ± 2.7 in the completers at baseline ($p=0.047$). The drop-outs also had a significantly lower median intake of fiber at baseline with 19.5 (3.4) vs. 24.9 (15.5) g/day ($p=0.01$). For all other measured variables the drop-outs were similar to those who completed the study.

Sociodemographic, medical and lifestyle characteristics between the diet study groups at baseline are reported in Table 1. Overall, the participants represented a middle to older-aged adult (mean age 62 ± 9 years), mostly male (84%) group, who were from a variety of ethnic backgrounds (55% Australian, 18% Asian, 17% European, 5% British, 4% African and 3% North American). More than 80% were overweight, 29% had diagnosed type 2 diabetes mellitus and 70% had experienced an AMI. The median time since recent cardiac event was less than 6-months. All participants were taking medications, of which the most common were anti-platelets (91%), statins (89%) and a range of anti-hypertensives. Less than half the cohort were taking nutrient supplements, of which vitamin D (20%), omega-3 (16%) and multivitamins (14%) were the most common. Few participants (13%) were current smokers, the majority had attended a cardiac rehabilitation program (80%) and more than one third had

previously seen a dietitian. There were no significant differences between the study groups for any of these characteristics at baseline. For intake of energy, contribution to energy from macronutrients, and micronutrients there also were no significant differences between the study groups. The low-fat diet participants did, however, have a significantly higher median intake of fruit compared to the MedDiet group, with 1.9 (1.2) versus 0.9 (1.2) serves per day ($p=0.006$), with no other differences in food group intake at baseline observed.

3.2. DII of model meal plans

Supplementary Table S1 provides an example of one day from each of the intervention diet meal plans and Table 2 reports on the mean daily intake for each of the 45 DII parameters and DII score for each meal plan. Both meal plans had anti-inflammatory (-) DII scores, but the meal plan for the MedDiet was markedly more anti-inflammatory than that of the low-fat diet (-4.55 vs. -0.33, respectively). The meal plans comparatively, contained some key differences for intake of DII parameters. The MedDiet was higher in the pro-inflammatory nutrient parameter of total fat content, but also higher in numerous anti-inflammatory parameters of the index (MUFA, PUFA, omega-3 and -6, fiber, alcohol, vitamin A, D and E, flavonoids, garlic, onion, rosemary and thyme/oregano).

3.3. Adherence to MedDiet

MEDAS score significantly increased in the MedDiet group from 5.52 ± 2.2 at baseline to 10.89 ± 1.6 at 6-months (5.37, $p<0.001$, 95%CI 4.56, 6.18). The low-fat diet group also significantly improved their score from 4.76 ± 1.9 at baseline to 6.00 ± 2.0 at 6-months (1.24, $p=0.002$, 95%CI 0.46, 2.02). The change in the MedDiet group was significantly greater than

that in the low-fat diet group, as assessed by ANCOVA, with baseline values as a covariate (adjusted difference 5.61, $p < 0.001$, 95% CI 3.11, 8.12). Baseline and 6-month adherence to each individual MEDAS component[44] within the two study groups is presented in the Supplemental Materials Table S2. Within the MedDiet group, the proportion of participants adhering to each of the score components significantly improved ($p < 0.05$), except for olive oil quantity ($p = 0.13$), sugar-sweetened beverages ($p = 0.25$) and wine ($p = 0.38$). Within the low-fat diet group the only components in the MEDAS score which had significantly improved adherence rates were intake of vegetables and reduced use of dairy spreads/cream ($p < 0.05$).

3.4. Impact of diet interventions on DII and inflammatory markers

Baseline and 6-month DII scores for the MedDiet and low-fat diet groups are presented in Figure 2. Within the MedDiet group there was a significant decrease from -0.40 ± 3.14 to -1.74 ± 2.8 at 6-months for DII (-1.34 , $p = 0.008$, 95% CI -2.33 , -0.36), with a similar magnitude of change for DII_{diet+supplements} (-1.41 , $p = 0.006$, 95% CI -3.19 , -0.56). Within the low-fat diet group the change in DII score from baseline (-0.17 ± 2.27) to 6-months (0.05 ± 1.89) was not significant (0.22 , $p = 0.65$, 95% CI -0.73 , 1.17); with a similar result for change in DII_{diet+supplements} (0.15 , $p = 0.75$, 95% CI -0.79 , 1.10). The between-group change as assessed by ANCOVA, with baseline scores as covariates, demonstrated a significantly greater reduction in the MedDiet group compared to low-fat diet group in mean score of DII (adjusted difference -1.69 , $p = 0.004$, 95% CI -2.82 , -0.56 .) and DII_{diet+supplements} (adjusted difference -1.75 , $p = 0.004$, 95% CI -2.90 , -0.59).

For mean daily intake of nutrient and food parameters of the index at 6-months (Table 2) there was a significantly higher intake of total fat, MUFA, omega-6, alcohol, vitamin E, flavones, flavonols and garlic and lower intake of trans fat in the MedDiet group compared to low-fat diet group.

From baseline to 6-months there was a non-significant decrease in hs-CRP within the MedDiet group (-0.28 mg/L, $p=0.38$, 95% CI -0.91, 0.36) and the low-fat diet group (-0.16 mg/L, $p=0.73$, 95% CI -1.13, 0.80). For hs-IL-6, there was also a non-significant decrease within the MedDiet group (-0.48 pg/mL, $p=0.17$, 95% CI -1.18, 0.21), but virtually no change within the low-fat diet group (-0.03 pg/mL, $p=0.91$, 95% CI -0.59, 0.53). The between-group changes as assessed by ANCOVA, with baseline marker levels and DII scores as covariates, demonstrated no significant difference between the MedDiet and low-fat diet group for hs-CRP (adjusted difference 0.09 mg/L, $p=0.84$, 95% CI -0.80, 0.98) or hs-IL-6 (adjusted difference -0.32 pg/mL, $p=0.29$, 95% CI -0.93, 0.28). The results did not differ when the covariate of baseline DII scores was removed from the models.

3.5. DII and MedDiet adherence

In all participants, MEDAS score had a moderate, negative correlation with DII at baseline ($r= -0.44$, $p=0.001$) and a strong, negative correlation with DII at 6-months ($r= -0.54$, $p<0.001$), which was strengthened when we adjusted for diet study group ($r= -0.70$, $p=0.001$). The 6-month change variable for MEDAS score had a weak negative correlation with change in DII score ($r= -0.29$, $p=0.03$), however this correlation did not remain when study group was controlled for ($r= -0.18$, $p=0.34$). At 6-months, half ($n=28$) of the pooled cohort of participants had high adherence to a traditional MedDiet (MEDAS ≥ 9)[45] and they

had a significantly lower DII score compared to those with a MEDAS score <9 when adjusted for baseline DII score (-1.58 ± 2.70 vs. -0.04 ± 2.10 , $p=0.04$, 95% CI 0.07, 2.40).

Mean 6-month changes in DII scores, adjusted for baseline DII values, were 0.17 (95% CI -0.708, 1.04), -1.17 (95% CI -2.24) and -1.29 (95% CI -2.47, -0.109) in tertiles 1 (-2 to 2 point change), 2 (3 to 5 point increase) and 3 (6 to 9 point increase) for change in MEDAS score, respectively. The mean adjusted changes across tertiles for DII_{diet+supplements} were 0.17 (95% CI -0.708, 1.04), -1.17 (95% CI -2.24, -0.094) and -1.29 (95% CI -2.47, -0.109), respectively. Table 3 presents the corresponding adjusted differences in mean values of change in DII score across tertiles of change in MEDAS, with T3 as the referent tertile. There were no significant differences across tertiles.

Mean change in DII score (without supplements) between adherers and non-adherers to each of the MEDAS score components at 6-months are presented in Table 4. Participants who adhered to the recommendations for intake of olive oil as the main culinary fat and quantity, vegetables, legumes, nuts, white meat preferred to red/processed meat and soffritto sauce at 6-months had a significantly greater reduction in DII score from baseline to 6-months than those who did not adhere to those components at 6-months when adjusted for baseline DII scores.

4. Discussion

This study assessed both the theoretical and intervention effect on DII scores of a MedDiet versus low-fat diet in an Australian setting. Through analysis of our model meal plans, we demonstrated that the MedDiet had high anti-inflammatory potential whereas a low-fat diet

had modest anti-inflammatory potential. In our pilot cohort of adult patients with CHD the MedDiet group adhered to the prescribed diet pattern and significantly reduced mean DII score after 6-months. By contrast, the low-fat diet did not influence DII score. After adjusting for baseline values, the MedDiet group significantly decreased mean DII score in comparison to the low-fat diet group. These results support our hypothesis that a MedDiet would improve DII score more significantly than a low-fat diet. Furthermore, when data for both intervention groups were pooled, greater MedDiet adherence scores significantly correlated with lower DII scores at baseline and 6-months. However, our results demonstrated that the MedDiet did not significantly reduce levels of inflammatory biomarkers hs-CRP and hs-IL-6, which was unexpected given the magnitude of improvement in DII score achieved with that diet.

The high anti-inflammatory potential of a MedDiet model was previously demonstrated in a one-day meal plan based on traditional Cretan MedDiet principles[20]. The analysis in that study also included the full 45 parameters of the index and produced a highly anti-inflammatory DII score of -3.96, which is comparable to the total score of -4.55 for our 2-week MedDiet meal plan. The slightly lower DII score of our MedDiet could be explained by the incorporation of additional herbs and spices, its higher isoflavone content due to inclusion of soy and linseed bread, which also has a high content of omega-3 PUFA, and a 2-week period with increased capacity to capture greater food and nutrient variety.

Despite both interventions representing healthy dietary patterns, the meal plan for the MedDiet had much greater anti-inflammatory potential than that of the low-fat diet. For the MedDiet model there were higher concentrations of nutrient components which are anti-inflammatory in the index, including MUFA, PUFA, fiber, alcohol, vitamins and flavonoids. This is reflective of the higher content of EVOO, nuts, vegetables, fruits, herbs, oily fish and

wine in the MedDiet. The low-fat meal plan had a higher content of carbohydrates, protein and cholesterol, which are each pro-inflammatory parameters of the index. Previously, higher participant adherence to healthy low-fat diet patterns has been demonstrated to be associated with lower DII scores[9]. However, our results are the first to show the theoretical DII score for a low-fat diet model.

Adherence to the MedDiet in our multi-ethnic cohort was inversely related to DII at baseline and 6-months. Our experimental data also demonstrated increasing MedDiet adherence score correlated with a lowering of DII; however, this was not significant when diet study group was accounted for. The association between higher adherence to a traditional MedDiet pattern and a lower DII score has previously been demonstrated in cross-sectional analyses. In over 7,000 Spanish participants enrolled in the PREDIMED trial, adherence to the MedDiet was higher in the lowest quintile of DII scores[16]. In a cohort of over 40,000 Australian, Italian or Greek-born Australians, there was an inverse correlation between the DII and MedDiet score[21].

Despite clear evidence to support the anti-inflammatory DII of the traditional MedDiet pattern, it had not previously been shown how a MedDiet intervention can change dietary intake and subsequently DII scores. This was important to consider in our non-Mediterranean cohort, where adherence to a MedDiet was low at baseline and a low-fat diet is still considered the standard recommendation. Our results demonstrated that a MedDiet intervention can be adhered to and leads to a significant reduction in DII score in a multi-ethnic cohort with chronic cardiovascular disease. Comparatively, prescription of the low-fat diet did not significantly change DII. We must recognize that in this cohort of patients with diagnosed CHD, the majority had received some type of low-fat dietary advice prior to

entering this study. Nonetheless, the theoretical and actual scope for improvement in DII with a low-fat diet was much lower than the MedDiet. At the end of the interventions, significant dietary intake differences between the groups mirrored many of the differences in the two model meal plans (such as dietary fats, alcohol, vitamin E and flavonoids).

Our findings add to the limited available literature testing the effect of healthy diet interventions on the DII. Randomization to plant-based diets (total n=63) demonstrated a vegan, vegetarian and pesco-vegetarian diet each reduced mean DII significantly (by -1.5, -1.4 and -1.6, respectively) after 2 months in a predominately female cohort[22]. However, these improvements in DII scores were not maintained at 6-months. Furthermore, there was limited improvement in that trial's control groups prescribed healthy semi-vegetarian and omnivorous[22] diets, which had similar dietary prescription to our low-fat diet group. In a large cohort of postmenopausal women low-fat diet intervention achieved a modest reduction in DII score (by -0.16) between 1-year and 6-years follow-up[23]. Both of these previous trials assessed only a subset of the 45 DII parameters, 27 and 32 respectively, therefore their findings may not truly reflect the diets' inflammatory potential.

We hypothesized that improvement in DII with our MedDiet intervention would affect inflammatory markers. However, our results showed that in this pilot cohort of patients with CHD the MedDiet did not significantly improve hs-CRP or hs-IL-6. When adjusting for baseline DII score, a promising trend was observed regarding a greater reduction in hs-IL-6 in the MedDiet, compared to low-fat diet participants. A significant effect of the MedDiet compared to control diets in intervention trials on both CRP (14 studies) and IL-6 (6 studies) has previously been confirmed[6]. However, a recent systematic review[7] which explored the effect of the MedDiet on inflammation in studies only recruiting patients with CHD,

found that there was no significant effect of Mediterranean-type diets compared to low-fat diets on CRP (4 studies)[51-54], and three of those studies also had small sample sizes ($n < 100$). Current treatment regimens for patients with diagnosed CHD include intensive pharmacotherapy[55]. In particular, both aspirin and statin medication have pleiotropic anti-inflammatory effects, which have been proposed to provide additional explanation for the impact which these medications have on reducing secondary CHD events[56, 57]. For hs-CRP, mean baseline levels in this AUSMED cohort were well within the normal range ($< 3 \text{ mg/L}$)[49]; hence, they may have had limited scope for improvement in this marker.

Observational studies have demonstrated that a more pro-inflammatory DII is associated with higher circulating levels of inflammatory markers CRP[12] and IL-6[14]. However, the effect of prospective changes in DII scores on these markers has not previously been tested and the existing studies were not conducted in populations on intensive medications, such as our CHD cohort. The association between DII and inflammatory markers has not been validated in patients with CHD. In the current AUSMED cohort, we have demonstrated (Mayr, et al., 2017, Companion Paper 2) that higher DII score had a nonsignificant association with an increased odds of elevated hs-CRP ($> 3 \text{ mg/L}$) at baseline (OR=1.16, 95%CI 0.89, 1.51).

Assessment of all 45 intake parameters of the DII strengthens our results. The MedDiet differs from other healthy dietary patterns through culinary aspects such as use of herbs and spices and other plant-foods high in flavonoids. When these parameters are not included in the DII calculation, the anti-inflammatory potential of the MedDiet, particularly in comparison to the low-fat diet which does not necessarily include these components, could be underestimated. Our participants' nutrient and food intake data were collected via 7-day food diaries and data was verified with the participant by a dietitian. This method would likely

result in more accurate DII scores than with use of food frequency questionnaires or 24-h dietary records, which are the most common methods used in other published analyses[11]. Our DII calculation also adjusted for supplement use.

Alongside our study's strengths and its novel findings, we must acknowledge some limitations. We had a small sample size, nine drop-outs were not included in our analysis and these participants had higher (more pro-inflammatory) DII scores at baseline. The majority of included participants were male, mean DII for both study groups was anti-inflammatory at baseline and mean hs-CRP levels were within the normal range. Our cohort is representative of patients with CHD on intensive pharmacotherapy, and who are potentially motivated and health conscious. Therefore, our results were affected by inadequate statistical power and are not necessarily applicable to healthy subjects, other disease populations or women.

Participants in the MedDiet group in this study were given food hampers to facilitate compliance, which is not reflective of a real-world setting. Our assessment for flavonoid intake was estimated using databases from the United States which may not accurately reflect Australian food composition. Our assessment of omega-6 was limited to linoleic acid.

Finally, our analyses were not based on intention-to-treat as the study was designed to test changes in DII score in participants who completed a dietary intervention. Future research should test whether the reduction in DII achieved by the MedDiet leads to an improvement in markers of inflammation (including markers in addition to CRP and IL-6) in a larger sample, or other cohorts including healthy subjects and individuals with other comorbidities.

We have demonstrated that a model MedDiet based on traditional principles of the diet is theoretically more anti-inflammatory than a low-fat diet. Furthermore, 6-month intervention with this MedDiet in a multi-ethnic, mostly male population with CHD was adhered to well

and lead to a significant improvement in DII scores. A low-fat diet did not improve DII scores in this patient group. The reduction in DII scores from consumption of this MedDiet tended to improve the inflammatory marker hs-IL-6 and this could be related to a reduction in risk of CHD. However, future studies in larger, well-powered samples and other population groups are needed to confirm whether reduction in DII scores through MedDiet intervention leads to reduced inflammation and associated risk of cardiometabolic diseases.

Acknowledgment

The authors are very grateful to all the participants of the study for their enthusiastic involvement and to the personnel of the affiliated hospital sites. We thank Cassandra Bendall for her assistance with data collection and entry and Jessica Radcliffe for her support during the data collection and entry process. This work was supported by La Trobe University (Understanding Disease RFA Start-Up Grant, 2013. HLM was supported by an Australian Government Research Training Program Scholarship and a Northern Health PhD Scholarship. JRH and NS were supported by the United States National Institute for Diabetes, Digestive and Kidney Diseases (grant no. R44DK103377).

The roles of the sponsors are as follows: the supplemental foods used in the study were generously donated by Cobram Estate of Boundary Bend Limited (extra virgin olive oil); the Almond Board of Australia (almonds); Jalna Dairy Foods Pty Ltd (Greek yoghurt); Simplot Australia Pty Ltd (canned fish and legumes); HJ Heinz Company Australia (canned fish and legumes); and Carman's (muesli bars). However, the sponsors had no role in the design, collection, analysis or writing of this article. The authors have no relevant interests to declare. However, in the spirit of full disclosure, JRH and NS make the following statement regarding their other work on DII-derived products. JRH owns controlling interest in Connecting

Health Innovations LLC (CHI), a company planning to license the right to his invention of the dietary inflammatory index (DII) from the University of South Carolina in order to develop computer and smart phone applications for patient counselling and dietary intervention in clinical settings. NS is an employee of CHI. The subject matter of this paper will not have any direct bearing on that work, nor has that activity exerted any influence on this project. HLM collected the presented data and analyzed the dietary intake and DII data (with support from CI, CJT and AT) and wrote the manuscript. All co-authors critically reviewed and edited the manuscript. EG and TK created the model meal plans and the Mediterranean diet and low-fat diet interventions (with support from CI and ACT). TK designed the trial protocol used herein (with support from CI and ACT). MRC assisted with design of the analyses for the manuscript. NS and JRH performed calculation of the DII from dietary intake data and provided important input in reviewing and editing drafts of the manuscript. However, they were not involved in performing statistical analyses on which the actual results are based.

Figure Captions

Figure 1. Study flow diagram of AUSMED participant appointments from randomization to diet study groups to end of intervention, 2014-2016. MedDiet, Mediterranean diet; AMI, acute myocardial infarction.

Figure 2. DII scores at baseline and 6-months in the Low-fat diet and Mediterranean diet study groups. Values are presented as means \pm SEM. DII, dietary inflammatory index; MedDiet, Mediterranean diet. *Significant difference between study groups at that time point, Independent Student's T-test, $p < 0.025$ (Bonferroni correction applied). †Significant difference to baseline within diet intervention group, Paired samples T-test, $p < 0.05$. ‡Significant difference between diet intervention groups for change in DII score, ANCOVA adjusted for baseline scores, $p < 0.05$.

References

- [1] Fuster V, Badimon L, Badimon JJ, Chesebro JH. The Pathogenesis of Coronary Artery Disease and the Acute Coronary Syndromes. *N Engl J Med* 1992;326:310-8.
- [2] Mozaffarian D. Dietary and Policy Priorities for Cardiovascular Disease, Diabetes, and Obesity: A Comprehensive Review. *Circulation* 2016;133:187-225.
- [3] Fung TT, McCullough ML, Newby P, Manson JE, Meigs JB, Rifai N, et al. Diet-quality scores and plasma concentrations of markers of inflammation and endothelial dysfunction. *Am J Clin Nutr* 2005;82:163-73.
- [4] Smidowicz A, Regula J. Effect of Nutritional Status and Dietary Patterns on Human Serum C-Reactive Protein and Interleukin-6 Concentrations. *Adv Nutr* 2015;6:738-47.
- [5] Neale E, Batterham M, Tapsell LC. Consumption of a healthy dietary pattern results in significant reductions in C-reactive protein levels in adults: a meta-analysis. *Nutr Res* 2016;36:391-401.
- [6] Schwingshackl L, Hoffmann G. Mediterranean dietary pattern, inflammation and endothelial function: a systematic review and meta-analysis of intervention trials. *Nutr Metab Cardiovasc Dis* 2014;24:929-39.
- [7] Mayr HL, Tierney AC, Thomas CJ, Ruiz-Canela M, Radcliffe J, Itsiopoulos C. Mediterranean-type diets and inflammatory markers in patients with coronary heart disease: a systematic review and meta-analysis. *Nutr Res* 2018;50:10-24.
- [8] Shivappa N, Steck SE, Hurley TG, Hussey JR, Hébert JR. Designing and developing a literature-derived, population-based dietary inflammatory index. *Public Health Nutr* 2014;17:1689-96.
- [9] Wirth MD, Hébert JR, Shivappa N, Hand GA, Hurley TG, Drenowatz C, et al. Anti-inflammatory Dietary Inflammatory Index scores are associated with healthier scores on other dietary indices. *Nutr Res* 2016;36:214-9.

- [10] Cuenca-García M, Artero EG, Sui X, Lee D-c, Hebert JR, Blair SN. Dietary indices, cardiovascular risk factors and mortality in middle-aged adults: findings from the Aerobics Center Longitudinal Study. *Ann Epidemiol* 2014;24:297-303. e2.
- [11] Ruiz-Canela M, Bes-Rastrollo M, Martínez-González MA. The role of dietary inflammatory index in cardiovascular disease, metabolic syndrome and mortality. *Int J Mol Sci* 2016;17:1265.
- [12] Shivappa N, Steck SE, Hurley TG, Hussey JR, Ma Y, Ockene IS, et al. A population-based dietary inflammatory index predicts levels of C-reactive protein in the Seasonal Variation of Blood Cholesterol Study (SEASONS). *Public Health Nutr* 2014;17:1825-33.
- [13] Tabung FK, Steck SE, Zhang J, Ma Y, Liese AD, Agalliu I, et al. Construct validation of the dietary inflammatory index among postmenopausal women. *Ann Epidemiol* 2015;25:398-405.
- [14] Shivappa N, Hébert JR, Rietzschel ER, De Buyzere ML, Langlois M, Debruyne E, et al. Associations between dietary inflammatory index and inflammatory markers in the Asklepios Study. *Br J Nutr* 2015;113:665-71.
- [15] Neufcourt L, Assmann K, Fezeu L, Touvier M, Graffouillère L, Shivappa N, et al. Prospective association between the dietary inflammatory index and metabolic syndrome: Findings from the SU. VI. MAX study. *Nutr Metab Cardiovasc Dis* 2015;25:988-96.
- [16] Ruiz-Canela M, Zazpe I, Shivappa N, Hébert J, Sánchez-Tainta A, Corella D, et al. Dietary inflammatory index and anthropometric measures of obesity in a population sample at high cardiovascular risk from the PREDIMED (PREvencion con DIeta MEDiterranea) trial. *Br J Nutr* 2015;113:984-95.
- [17] Ramallal R, Toledo E, Martínez-González MA, Hernández-Hernández A, García-Arellano A, Shivappa N, et al. Dietary Inflammatory Index and Incidence of Cardiovascular Disease in the SUN Cohort. *PLoS One* 2015;10:e0135221.

- [18] O'Neil A, Shivappa N, Jacka FN, Kotowicz MA, Kibbey K, Hebert JR, et al. Pro-inflammatory dietary intake as a risk factor for CVD in men: a 5-year longitudinal study. *Br J Nutr* 2015;114:2074-82.
- [19] Garcia-Arellano A, Ramallal R, Ruiz-Canela M, Salas-Salvadó J, Corella D, Shivappa N, et al. Dietary Inflammatory Index and Incidence of Cardiovascular Disease in the PREDIMED Study. *Nutrients* 2015;7:4124-38.
- [20] Steck S, Shivappa N, Tabung F, Harmon B, Wirth M, Hurley T, et al. The dietary inflammatory index: a new tool for assessing diet quality based on inflammatory potential. *Digest* 2014;49:1-9.
- [21] Hodge A, Bassett J, Shivappa N, Hébert J, English D, Giles G, et al. Dietary inflammatory index, Mediterranean diet score, and lung cancer: a prospective study. *Cancer Causes Control* 2016;27:907-17.
- [22] Turner-McGrievy GM, Wirth MD, Shivappa N, Wingard EE, Fayad R, Wilcox S, et al. Randomization to plant-based dietary approaches leads to larger short-term improvements in Dietary Inflammatory Index scores and macronutrient intake compared with diets that contain meat. *Nutr Res* 2015;35:97-106.
- [23] Tabung F, Steck S, Zhang J, Ma Y, Liese A, Tylavsky F, et al. Longitudinal changes in the dietary inflammatory index: an assessment of the inflammatory potential of diet over time in postmenopausal women. *Eur J Clin Nutr* 2016;70:1374-80.
- [24] NHF. Reducing risk in heart disease: an expert guide to clinical practice for secondary prevention of coronary heart disease. Melbourne: National Heart Foundation of Australia and the Cardiac Society of Australia and New Zealand; 2012.
- [25] Schulz KF, Altman DG, Moher D. CONSORT 2010 statement: updated guidelines for reporting parallel group randomised trials. *BMC Med* 2010;8:18.

- [26] World Medical Association. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. <https://www.wma.net/what-we-do/medical-ethics/declaration-of-helsinki/>. 2008 [Accessed 14 February 2018].
- [27] The Criteria Committee of the New York Heart Association. Nomenclature and criteria for diagnosis of diseases of the heart and blood vessels. Boston: Little Brown; 1964.
- [28] Levey AS, Eckardt K-U, Tsukamoto Y, Levin A, Coresh J, Rossert J, et al. Definition and classification of chronic kidney disease: a position statement from Kidney Disease: Improving Global Outcomes (KDIGO). *Kidney Int* 2005;67:2089-100.
- [29] Jelinek M, Vale MJ, Liew D, Grigg L, Dart A, Hare DL, et al. The COACH program produces sustained improvements in cardiovascular risk factors and adherence to recommended medications—two years follow-up. *Heart Lung Circ* 2009;18:388-92.
- [30] Keys A, Mienotti A, Karvonen MJ, Aravanis C, Blackburn H, Buzina R, et al. The diet and 15-year death rate in the seven countries study. *Am J Epidemiol* 1986;124:903-15.
- [31] Estruch R, Ros E, Salas-Salvadó J, Covas M-I, Corella D, Arós F, et al. Primary prevention of cardiovascular disease with a Mediterranean diet. *N Engl J Med* 2013;368:1279-90.
- [32] Vincent S, Gerber M, Bernard M, Defoort C, Loundou A, Portugal H, et al. The Medi-RIVAGE study (Mediterranean Diet, Cardiovascular Risks and Gene Polymorphisms): rationale, recruitment, design, dietary intervention and baseline characteristics of participants. *Public Health Nutr* 2004;7:531-42.
- [33] Itsiopoulos C, Brazionis L, Kaimakamis M, Cameron M, Best JD, O’Dea K, et al. Can the Mediterranean diet lower HbA1c in type 2 diabetes? Results from a randomized cross-over study. *Nutr Metab Cardiovasc Dis* 2011;21:740-7.

- [34] Ryan MC, Itsiopoulos C, Thodis T, Ward G, Trost N, Hofferberth S, et al. The Mediterranean diet improves hepatic steatosis and insulin sensitivity in individuals with non-alcoholic fatty liver disease. *J Hepatol* 2013;59:138-43.
- [35] Ministry of Health and Welfare SSHC. Dietary guidelines for adults in Greece. *Archives of Hellenic Medicine* 1999;16:516-24.
- [36] NHMRC. Australian Dietary Guidelines. Canberra: National Health and Medical Research Council; 2013.
- [37] NHMRC. Nutrient Reference Values for Australia and New Zealand; Including Recommended Dietary Intakes. Canberra: Commonwealth of Australia; 2006.
- [38] Samdal GB, Eide GE, Barth T, Williams G, Meland E. Effective behaviour change techniques for physical activity and healthy eating in overweight and obese adults; systematic review and meta-regression analyses. *Int J Behav Nutr Phys Act* 2017;14:42.
- [39] Shilts MK, Horowitz M, Townsend MS. Goal setting as a strategy for dietary and physical activity behavior change: a review of the literature. *Am J Health Promot* 2004;19:81-93.
- [40] Kouris-Blazos A, Itsiopoulos C. Low all-cause mortality despite high cardiovascular risk in elderly Greek-born Australians: attenuating potential of diet? *Asia Pac J Clin Nutr* 2014;23:532-44.
- [41] Freedson PS, Melanson E, Sirard J. Calibration of the Computer Science and Applications, Inc. accelerometer. *Med Sci Sports Exerc* 1998;30:777-81.
- [42] ISAK. International Standards for Anthropometric Assessment. South Australia: International Society for the Advancement of Kinanthropometry; 2001.
- [43] WHO. Physical status: The use of and interpretation of anthropometry, Report of a WHO Expert Committee. Geneva: World Health Organisation; 1995.

- [44] Schröder H, Fitó M, Estruch R, Martínez-González MA, Corella D, Salas-Salvadó J, et al. A Short Screener Is Valid for Assessing Mediterranean Diet Adherence among Older Spanish Men and Women. *J Nutr* 2011;141:1140-5.
- [45] Ferrer JT, Riera IS, Solórzano MC, Cabré MG, Tapias MG, Masoliver CS. Adherence to the Mediterranean Diet in Patients With Coronary Artery Disease. *Rev Esp Cardiol* 2015;68:73-5.
- [46] FSANZ. NUTTAB 2010 - Vitamin D File. <http://www.foodstandards.gov.au/science/monitoringnutrients/nutrientables/pages/default.aspx>. 2010 [accessed 01 June 2017].
- [47] Neveu V, Perez-Jiménez J, Vos F, Crespy V, du Chaffaut L, Mennen L, et al. Phenol-Explorer: an online comprehensive database on polyphenol contents in foods. *Database (Oxford)* 2010;2010:bap024.
- [48] Eldridge SM, Chan CL, Campbell MJ, Bond CM, Hopewell S, Thabane L, et al. CONSORT 2010 statement: extension to randomised pilot and feasibility trials. *Pilot and feasibility studies* 2016;2:64.
- [49] Pearson TA, Mensah GA, Alexander RW, Anderson JL, Cannon RO, Criqui M, et al. Markers of Inflammation and Cardiovascular Disease. Application to Clinical and Public Health Practice: A Statement for Healthcare Professionals From the Centers for Disease Control and Prevention and the American Heart Association 2003;107:499-511.
- [50] Cohen J. *Statistical power analysis for the behavioral sciences* Lawrence Earlbaum Associates. Hillsdale, NJ 1988:20-6.
- [51] Gomez-Delgado F, Garcia-Rios A, Alcalá-Díaz JF, Rangel-Zuniga O, Delgado-Lista J, Yubero-Serrano EM, et al. Chronic consumption of a low-fat diet improves cardiometabolic risk factors according to the CLOCK gene in patients with coronary heart disease. *Mol Nutr Food Res* 2015;59:2556-64.

- [52] Chen CYO, Holbrook M, Duess MA, Dohadwala MM, Hamburg NM, Asztalos BF, et al. Effect of almond consumption on vascular function in patients with coronary artery disease: a randomized, controlled, cross-over trial. *Nutr J* 2015;14:1-11.
- [53] Thomazella MCD, Góes MF, Andrade CR, Debbas V, Barbeiro DF, Correia RL, et al. Effects of high adherence to mediterranean or low-fat diets in medicated secondary prevention patients. *Am J Cardiol* 2011;108:1523-9.
- [54] Tuttle KR, Shuler LA, Packard DP, Milton JE, Daratha KB, Bibus DM, et al. Comparison of low-fat versus Mediterranean-style dietary intervention after first myocardial infarction (from The Heart Institute of Spokane Diet Intervention and Evaluation Trial). *Am J Cardiol* 2008;101:1523-30.
- [55] Chew DP, Scott IA, Cullen L, French JK, Briffa TG, Tideman PA, et al. National Heart Foundation of Australia and Cardiac Society of Australia and New Zealand: Australian clinical guidelines for the management of acute coronary syndromes 2016. *Med J Aust* 2016;205:128-33.
- [56] Ridker PM, Cannon CP, Morrow D, Rifai N, Rose LM, McCabe CH, et al. C-reactive protein levels and outcomes after statin therapy. *N Engl J Med* 2005;352:20-8.
- [57] Amann R, Peskar BA. Anti-inflammatory effects of aspirin and sodium salicylate. *Eur J Pharmacol* 2002;447:1-9.

Table 1. Characteristics of AUSMED participants between diet study groups, 2014-2016^a

Variable	Low-fat diet	MedDiet	P ^b
Age (years)	61.8 ± 9.9	62.7 ± 7.7	0.71
Sex			
Female	3 (10.3)	6 (22.2)	0.40
Male	26 (89.7)	21 (77.8)	
Region of Birth			
Australia	16 (55.2)	15 (55.6)	0.88
Other	13 (44.8)	12 (44.4)	
Europe	5 (17.2)	4 (14.8)	
United Kingdom	2 (6.9)	1 (3.7)	
Asia	4 (13.8)	6 (22.2)	
Africa	1 (3.4)	1 (3.7)	
North America	1 (3.4)		
Education			
Primary School	1 (3.4)	2 (7.4)	0.58
Secondary School	7 (24.1)	4 (14.8)	
Trade/ University	21 (72.4)	21 (77.8)	
Body mass index (kg/m ²)	29.4 ± 5.3	30.4 ± 5.2	0.45
Overweight (>25 kg/m ²)	24 (82.8)	23 (85.2)	1.00
Acute coronary syndrome history			
Acute myocardial infarction	21 (72.4)	18 (66.7)	0.86
Coronary artery bypass grafting	8 (27.6)	5 (18.5)	0.63
Percutaneous coronary intervention	22 (75.9)	21 (77.8)	1.00
Time since event (months) ^c	4.5 (9.0)	5.3 (15.1)	0.65
Type 2 diabetes mellitus	9 (31.0)	7 (25.9)	0.90
Medication use	29 (100)	27 (100)	1.00
Anti-platelet	27 (93.1)	24 (88.9)	0.93
Statin	25 (86.2)	25 (92.6)	0.73
β-blocker	19 (65.5)	17 (63.0)	1.00
ACE-inhibitor	14 (48.3)	14 (51.9)	1.00
Angiotensin 2 receptor blocker	6 (20.7)	5 (18.5)	1.00
Calcium-channel blocker	5 (17.2)	2 (7.4)	0.48
Oral hypoglycaemic agent	7 (24.1)	6 (22.2)	1.00
Insulin	2 (6.9)	2 (7.4)	1.00
Supplement use	12 (41.4)	13 (48.1)	0.81
Omega-3	3 (10.3)	6 (22.2)	0.40
Vitamin D	5 (17.2)	6 (22.2)	0.90
Multivitamin	5 (17.2)	3 (11.1)	0.79
Magnesium	3 (10.3)	3 (11.1)	1.00
Iron	2 (6.9)	0 (0.0)	0.49
Vitamin E	1 (3.4)	0 (0.0)	1.00
Vitamin C	1 (3.4)	0 (0.0)	1.00
Folate	1 (3.4)	0 (0.0)	1.00
Lifestyle			
MVPA (min/week) ^c	118 (203)	194 (287)	0.29
Current smoker	2 (6.9)	5 (18.5)	0.36
Cardiac rehabilitation	25 (86.2)	20 (74.1)	0.42
Dietitian (individual consult)	10 (34.5)	10 (37.0)	1.00
DII ^d	-0.17 ± 2.27	-0.40 ± 3.14	0.75
DII _{diet+supplements} ^d	-0.33 ± 2.29	-0.64 ± 3.19	0.68

Participants were randomized to the Low-fat diet, n=29 or MedDiet, n=27.

Abbreviations: MedDiet, Mediterranean diet; ACE, angiotensin converting enzyme; MVPA, moderate-to-vigorous physical activity; DII, Dietary Inflammatory Index.

^aValues are n (%), means \pm SD or medians (IQR).

^b*Chi square* test of independence, Independent Student's T-test or Mann-Whitney U test.

^cNon-parametric continuous variables, presented as Medians (IQR).

^dNegative number reflects an anti-inflammatory score, while positive number reflects a pro-inflammatory score.

ACCEPTED MANUSCRIPT

Table 2. Parameters and scores of DII for the AUSMED Low-fat diet and Mediterranean diet intervention meal plans and participant intake at 6-months^a

Intake Parameter	Meal Plans		Intake at 6-months		P ^b
	Low-fat diet	MedDiet	Low-fat diet	MedDiet	
Energy (kcal)	2068.23	2240.90	1823.30 ± 551.1	2030.04 ± 486.5	0.08
Protein (g)	99.73	87.32	96.14 ± 25.8	92.08 ± 26.6	0.28
Carbohydrate (g)	277.10	194.00	194.60 ± 67.5	176.61 ± 54.3	0.30
Fat (g)	53.19	105.96	63.93 ± 29.1	91.64 ± 30.8	<0.001*
MUFA (g)	21.28	52.45	24.85 ± 12.4	46.51 ± 18.5	<0.001*
PUFA (g)	10.58	24.76	12.07 ± 7.2	17.25 ± 7.8	0.004*
Omega-3 (g)	1.26	5.87	1.98 ± 0.8	2.59 ± 1.6	0.07
Omega-6 (g)	9.14	18.66	9.85 ± 6.7	14.41 ± 6.6	0.01*
Saturated fat (g)	16.06	20.85	21.02 ± 10.5	20.70 ± 6.4	0.56
Trans fat (g)	0.65	0.58	0.95 ± 0.6	0.65 ± 0.3	0.02*
Cholesterol (mg)	340.95	255.75	307.44 ± 152.6	268.51 ± 128.4	0.22
Fiber (g)	32.88	41.08	28.17 ± 8.9	31.57 ± 11.1	0.21
Alcohol (g)	0.00	11.12	3.32 ± 6.8	8.98 ± 11.3	0.01*
Caffeine (g)	0.08	0.14	0.21 ± 0.3	0.14 ± 0.1	0.56
Folate (µg)	580.38	732.31	536.14 ± 192.6	521.91 ± 167.5	0.94
Beta carotene (µg)	2844.89	3039.02	3297.3 ± 1818.9	4130.52 ± 2521.8	0.25
Vitamin A (RE)	844.20	2161.56	889.38 ± 407.4	1015.70 ± 567.0	0.59
Vitamin B6 (mg)	1.30	2.21	1.85 ± 2.1	1.68 ± 0.7	0.54
Vitamin B12 (µg)	4.40	7.02	5.36 ± 5.1	4.09 ± 2.1	0.10
Vitamin C (mg)	126.21	191.82	127.86 ± 102.8	123.34 ± 80.9	0.75
Vitamin D (µg)	1.45	3.17	2.22 ± 2.8	3.85 ± 4.6	0.14
Vitamin E (mg)	10.57	29.27	11.35 ± 5.9	20.10 ± 6.4	<0.001*
Iron (mg)	13.10	15.27	11.41 ± 3.5	12.20 ± 3.3	0.39
Magnesium (mg)	399.83	462.78	345.61 ± 108.9	375.60 ± 95.9	0.28
Niacin (mg)	23.12	20.22	24.57 ± 8.2	22.39 ± 7.0	0.34
Riboflavin (mg)	1.90	1.85	1.85 ± 0.8	1.72 ± 0.5	0.48
Selenium (µg)	116.81	82.84	89.20 ± 25.0	99.32 ± 36.1	0.34
Thiamin (mg)	1.52	1.31	1.58 ± 0.7	1.33 ± 0.5	0.28
Zinc (mg)	11.36	11.68	10.96 ± 3.0	9.92 ± 2.6	0.14
Flavan-3-ol (mg)	12.02	44.73	288.44 ± 369.8	361.96 ± 359.5	0.47
Flavones (mg)	2.72	14.02	2.97 ± 6.4	3.30 ± 5.3	0.003*
Flavonols (mg)	20.64	36.63	21.78 ± 15.7	33.65 ± 23.6	0.03*
Flavonones (mg)	12.97	13.21	10.51 ± 13.7	13.37 ± 20.6	0.33
Anthocyanidins (mg)	7.01	72.84	23.38 ± 23.8	20.73 ± 13.6	0.75
Isoflavones (mg)	0.26	15.46	0.52 ± 0.9	1.47 ± 2.4	0.59
Eugenol (mg)	0.00	1.53	0.00 ± 0.0	0.22 ± 1.1	0.30
Green/black tea (g)	0.00	0.36	2.65 ± 3.2	3.20 ± 3.1	0.48
Garlic (g)	0.73	1.73	0.21 ± 0.5	0.70 ± 1.0	0.03*
Ginger (g)	0.29	0.00	0.11 ± 0.3	0.12 ± 0.6	0.27
Onion (g)	24.57	58.55	15.92 ± 22.2	24.28 ± 32.9	0.14
Pepper (g)	0.27	0.36	0.02 ± 0.0	0.05 ± 0.1	0.77
Rosemary (mg)	0.09	71.43	0.81 ± 3.2	16.82 ± 67.6	0.32
Saffron (g)	0.00	0.00	0.00 ± 0.0	0.002 ± 0.0	0.30
Turmeric (mg)	0.00	0.00	9.75 ± 38.6	28.04 ± 145.7	0.63
Thyme/oregano (mg)	0.00	540.00	2.17 ± 9.1	22.95 ± 72.3	0.17
DII ^c	-0.33	-4.55	0.05 ± 1.89	-1.74 ± 2.27	0.007*

The 45 intake parameters listed represent all which are included in the DII. The meal plans modeled the diet interventions for the low-fat diet (1-week) and the MedDiet (2-weeks). Dietary intake of the parameters by the low-fat diet (n=29) and MedDiet (n=27) participants at 6-months represents intake from foods only with supplement intake excluded.

Abbreviations: DII, dietary inflammatory index; MedDiet, Mediterranean diet; MUFA, mono-unsaturated fatty acids; PUFA, polyunsaturated fatty acids; RE, retinol equivalents.

^aValues are daily intake for the meal plans and means \pm SD for intake at 6-months.

^bDifferences between intake of the study groups at 6-months, Independent Student's T-test or Mann-Whitney U test.

^cNegative number reflects an anti-inflammatory score, while positive number reflects a pro-inflammatory score.

*Significant difference in intake of that parameter between the MedDiet and low-fat diet group participants at 6-months, $p < 0.05$.

Table 3. Adjusted mean differences in the 6-month change in DII scores between Tertiles of 6-month change in MEDAS^a

DII score	T1 -2 to 2	T2 3 to 5	T3 6 to 9	P- trend^b
Participants (n)	25	17	14	
Change in DII	1.38 -0.08, 2.83	0.06 -1.54, 1.66	0 Reference	0.08
Change in DII _{diet+supplements}	1.46 -0.01, 2.93	0.12 -1.49, 1.73	0 Reference	0.07

Participants were distributed into tertiles of 6-month change in MEDAS score based on having an equal number of participants across three groups using SPSS statistical software. Abbreviations: MEDAS, Mediterranean Diet Adherence Screener; DII, dietary inflammatory index; T, tertile.

^aValues are adjusted differences in mean 6-month change in DII score to T3 with 95% CIs.

^bP-trend between tertiles, ANCOVA adjusted for baseline DII scores.

Table 4. Mean 6-month changes in DII scores between adherers and non-adherers to MEDAS components at 6-months^a

MEDAS component		No at 6-months		Yes at 6-months		Adjusted Difference ^b	P
Olive oil main fat	17	0.65 ± 2.1	39	-1.05 ± 2.7	1.45	0.21, 2.70	0.02*
Olive oil quantity /d	49	-0.27 ± 2.5	7	-2.38 ± 3.3	2.31	0.60, 4.01	0.009*
Vegetable serves /d	21	0.23 ± 2.2	35	-0.99 ± 2.8	1.57	0.40, 2.74	0.01*
Fruit serves /d	36	-0.70 ± 2.6	20	-0.23 ± 2.7	0.81	-0.53, 2.15	0.23
Red/processed meat serves /d	14	0.14 ± 2.4	42	-0.76 ± 2.7	1.24	-0.11, 2.60	0.07
Butter/ marg. /cream serves /d	6	-0.36 ± 2.4	50	-0.55 ± 2.7	0.46	-1.48, 2.40	0.64
Sugar-sweetened drinks /d	4	-0.99 ± 3.0	52	-0.50 ± 2.6	1.28	-1.14, 3.69	0.29
Wine glasses /wk	46	-0.48 ± 2.8	10	-0.76 ± 1.9	1.04	-0.54, 2.61	0.19
Legumes serves /wk	29	0.02 ± 2.1	27	-1.09 ± 3.0	1.48	0.34, 2.62	0.01*
Fish or seafood serves /wk	16	-0.78 ± 2.1	40	-0.44 ± 2.8	0.33	-1.03, 1.68	0.63
Commercial sweets /wk	14	-0.69 ± 2.7	42	-0.48 ± 2.7	0.41	-1.00, 1.81	0.56
Nuts serves /wk	16	-0.31 ± 2.3	40	-0.62 ± 2.8	1.46	0.11, 2.80	0.03*
Poultry preferred to red meats	13	0.82 ± 2.5	43	-0.94 ± 2.6	1.83	0.50, 3.16	0.008*
Use of sofrito sauce /wk	36	-0.26 ± 2.4	20	-1.03 ± 3.0	1.46	0.25, 2.68	0.02*

The MEDAS scoring tool was used to assess Mediterranean diet adherence in all study participants. Each of its 14 components represent a food group based intake recommendation. The data represents changes in DII score according to whether participants adhered to each of those recommendations or not at 6-months.

Abbreviations: DII; dietary inflammatory index; MEDAS, Mediterranean diet adherence screener; marg, margarine.

^aValues are participants (n) and means ± SD.

^bAdjusted mean differences (95% CIs, p-values) between adherers and non-adherers, ANCOVA adjusted for baseline DII scores.

*Significant difference for mean 6-month change in DII score between adherers and non-adherers to that MEDAS component at 6-months, p<0.05.

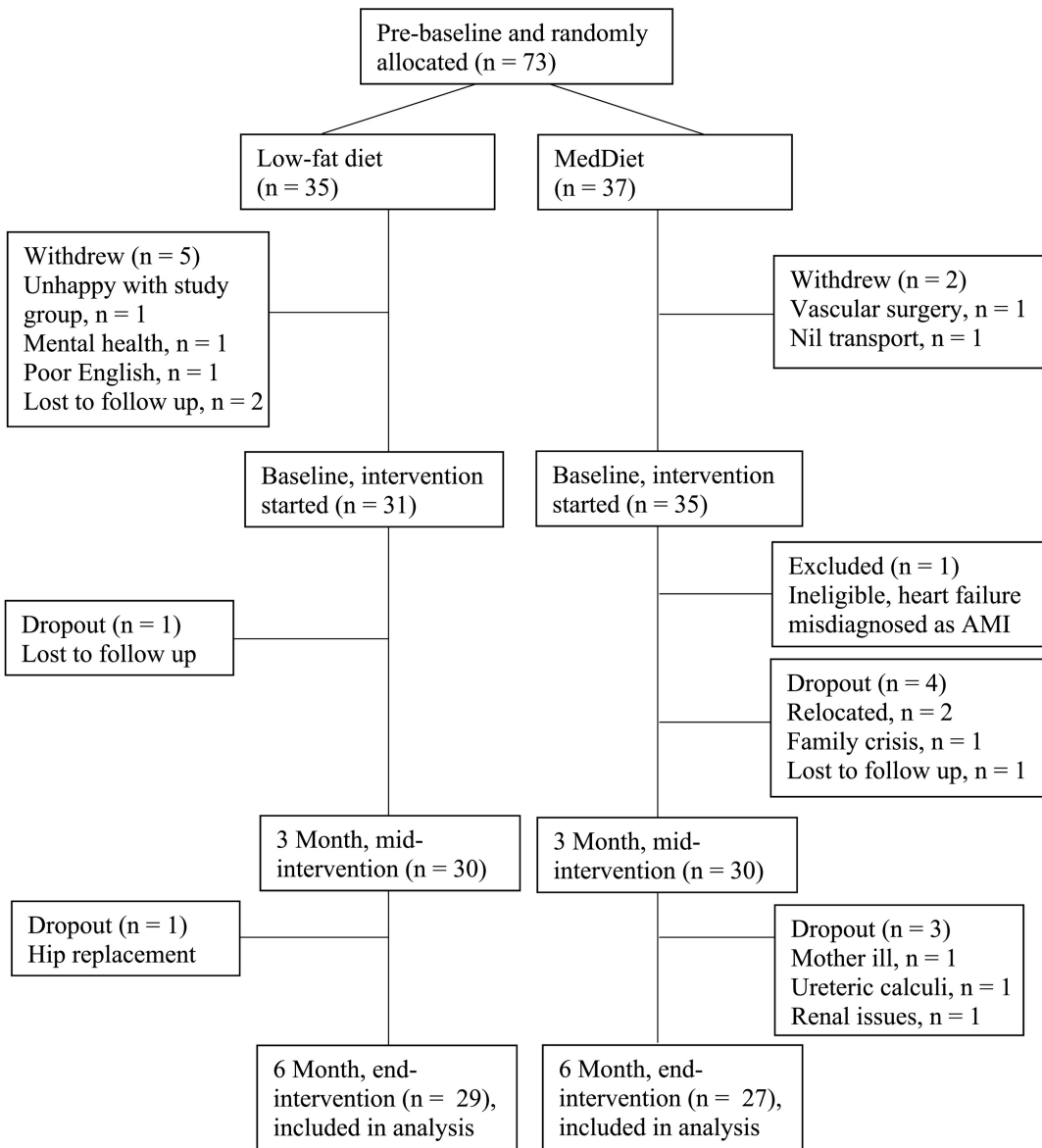


Figure 1

Low-fat diet
MedDiet

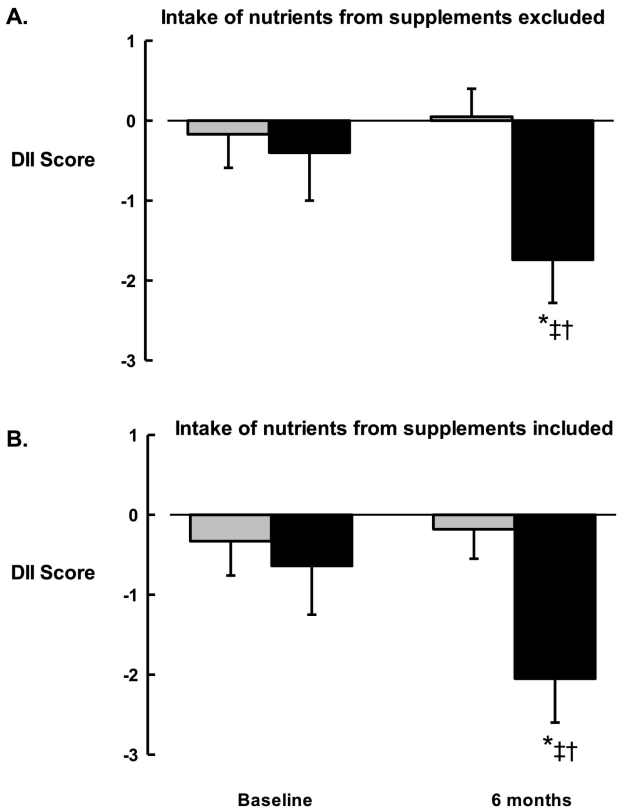


Figure 2