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Benefits of the Mediterranean Diet: Insights From the PREDIMED Study

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ABSTRACT

The PREDIMED (PREvención con DIeta MEDiterránea) multicenter, randomized, primary prevention trial assessed the long-term effects of the Mediterranean diet (MeDiet) on clinical events of cardiovascular disease (CVD). We randomized 7447 men and women at high CVD risk into three diets: MeDiet supplemented with extra-virgin olive oil (EVOO), MeDiet supplemented with nuts, and control diet (advice on a low-fat diet). No energy restriction and no special intervention on physical activity were applied. We observed 288 CVD events (a composite of myocardial infarction, stroke or CVD death) during a median time of 4.8 years; hazard ratios were 0.70 (95% CI, 0.53–0.91) for the MeDiet + EVOO and 0.70 (CI, 0.53–0.94) for the MeDiet + nuts compared to the control group. Respective hazard ratios for incident diabetes (273 cases) among 3541 non-diabetic participants were 0.60 (0.43–0.85) and 0.82 (0.61–1.10) for MeDiet + EVOO and MeDiet + nuts, respectively versus control. Significant improvements in classical and emerging CVD risk factors also supported a favorable effect of both MeDiets on blood pressure, insulin sensitivity, lipid profiles, lipoprotein particles, inflammation, oxidative stress, and carotid atherosclerosis. In nutrigenomic studies beneficial effects of the intervention with MedDiets showed interactions with several genetic variants (TCF7L2, APOA2, MLXIPL, LPL, FTO, M4CR, COX-2, GCKR and SERPINE1) with respect to intermediate and final phenotypes. Thus, the PREDIMED trial provided strong evidence that a vegetable-based MeDiet rich in unsaturated fat and polyphenols can be a sustainable and ideal model for CVD prevention.

Statement of Conflict of Interest: see page 56.

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Abbreviations and Acronyms

AF = atrial fibrillation

CHD = coronary heart disease

CV = cardiovascular

CVD = CV disease

DLP = dyslipidemia

EVOO = extra-virgin olive oil

FFQ = food frequency questionnaire

HTN = hypertension

MeDiet = Mediterranean diet

MetS = metabolic syndrome

PAD = peripheral artery disease

PREDIMED = PREvención con DIeta MEDiterránea

RCT = randomized control trial

T2DM = type 2 diabetes mellitus

Cardiovascular (CV) disease (CVD) is the main cause of worldwide premature mortality. Coronary heart disease (CHD) and stroke ranked first and third, respectively, as the leading global causes of disability-adjusted years according to the global burden of disease estimates for 2010.[1] Furthermore, the projections of mortality from CVD for 2030 are dismal[2,3] and underline the need for preventive strategies as a public health priority. In this context, a high-quality diet and a healthy lifestyle at middle age are the most important factors for CVD prevention.[3–7] Consequently, the diet-heart hypothesis has been a long-standing tenet in CVD prevention and nutritional epidemiology during the last 50 years.[7,8] Recently, the relevance of overall high-quality food patterns, rather than the focus on single nutrients and foods, has emerged as a powerful paradigm to address the inherent complexity of dietary exposures and to assess their potential CVD preventive effects. Food patterns can be described as the amounts, proportions, combinations or varieties for the consumption of different foods and beverages and the frequency with which they are usually consumed. This approach allows the assessment of synergistic interactions and cumulative effects among different foods and nutrients, preempts confounding by alternative dietary exposures, avoids some problems of co-linearity between foods or nutrients and thus provides a strong methodological tool in nutritional epidemiology.[9,10] Even though randomized dietary intervention trials are the hallmarks for acquiring knowledge on the effects of diet on CVD, most research in the field of dietary patterns is observational, with some potential for residual confounding and other possible sources of bias. These limitations have been the subject of ample but probably undue criticism.[10,11]

A weakness of the diet-heart hypothesis is that most of the available experimental research in the field has not used hard clinical outcomes, but only intermediate risk biomarkers. The existence of multiple pathways leading from diet to CVD speaks against the simplistic approach of giving a high value to changes in any single biomarker. Moreover, the induction period can vary for the different pathways in which diverse biomarkers are involved, thus limiting the possibility of assessing multiple biomarker combinations at any time point. Furthermore, other lesser-known pathways could account for a substantial proportion of clinical CVD events. The most sensible approach, therefore, in order to investigate the diet-heart hypothesis is to use hard clinical CVD events as end-points of randomized controlled trials (RCTs). Most feeding trials, however, are usually short term and rarely include clinical end-points such as CVD events or death.[11] The PREDIMED trial was designed to overcome both the problem of the single-nutrient approach and the limitations of assessing only intermediate risk markers. Indeed, the PREDIMED randomized trial used an overall food pattern as the intervention and assessed hard CVD events as end-points[12] providing a high level of scientific evidence.

Scientific evidence of the cardio-metabolic benefits of the Mediterranean diet

The abundant and consistent observational evidence that was available to support the benefits of the Mediterranean diet (MeDiet) and, specifically, of tree nuts and olive oil, on CV health prompted us to choose this traditional dietary model enriched with olive oil or nuts as the intervention.[13–44] Table 1 summarizes the results of meta-analyses and systematic reviews assessing the effects of MeDiet on different cardiometabolic outcomes.

The MeDiet is defined as the traditional dietary pattern found in the early 1960s in Greece, Southern Italy, Spain and other olive-growing countries of the Mediterranean basin. It is a frugal diet that uses generous amounts of olive oil as main culinary fat and has a high consumption of plant-derived foods (fruit, vegetables, legumes, nuts and seeds, and whole grain cereals); frequent but moderate intake of wine (especially red wine), usually with meals; moderate consumption of seafood and dairy products (especially yogurt and cheese, but not whole milk, butter or cream), poultry and eggs; and low consumption of sweet desserts, red and processed meats. In comparison with other healthy patterns, such as the DASH diet, the healthy US dietary pattern or the Alternative Healthy Eating Index, the consumption of fruit and fish is usually higher in the MeDiet, while the consumption of dairy products tends to be lower. In healthy vegetarian food patterns, meat and seafood are not consumed, but eggs and dairy are most frequently included. Legumes, nuts/seeds, and processed soy are all higher in a healthy vegetarian food pattern than in the healthy U.S.-style or Mediterranean-style patterns.

A considerable scientific advantage of the MeDiet over other healthy dietary patterns was the availability of a previous randomized trial, the Lyon Diet Heart study, conducted in myocardial infarction survivors (i.e., it was a secondary prevention trial). It showed that a MeDiet enriched with alpha-linolenic acid, but not olive oil, provided a strong protection against recurrent CHD.[45]

Our hypothesis when designing the PREDIMED trial was that the MeDiet would be superior to a low-fat diet for primary CVD prevention. This hypothesis had never been tested previously using a RCT design.

Design and methods of the PREDIMED study

The PREDIMED study was a primary prevention trial which tested the long-term effects of the MeDiet on incident CVD in men and women at high CVD risk aged 55–75 years (men) or

Table 1 – Scientific evidence on the Mediterranean diet.
Systematic reviews assessing the association between adherence to the Mediterranean diet and cardio-metabolic outcomes.

Systematic Review	N (Studies)	Exposure	Outcome	Effects of Increased Adherence to MeDiet [a]	Comment
Esposito 2011[31]	16	MeDiet (randomized trials)	Weight loss	−1.75 kg (−2.86; −0.64)	Greater weight loss with energy restriction and longer follow-up
Buckland 2008[38]	21	MeDiet (randomized trials)	Weight loss	Beneficial (13 studies); no evidence (8 studies)	Qualitative systematic review
Nordmann 2011[28]	6	MeDiet (randomized trials)	Risk factors	Beneficial	Reductions in BMI, BP, glucose and CRP
Grosso 2014[24]	58	MeDiet	Risk factors	Beneficial	Qualitative systematic review
Schwingshackl 2014[18]	17	MeDiet	Flow-mediated dilatation	WMD 1.86% (0.23–3.48)	Adiponectin levels also increased
Schwingshackl 2014[18]	17	MeDiet	High-sensitivity CRP	WMD = −1 mg/l (−1.5; −0.5)	Similar favorable changes in IL-6 and ICAM-1
Kastorini 2011[30]	50	MeDiet (randomized trials)	Metabolic syndrome	RR = 0.50 (0.29–0.85) [b]	Significant beneficial effects were found for each of the metabolic syndrome criteria
Esposito 2013[25]	14	MeDiet	Metabolic syndrome	Beneficial	Qualitative systematic review
Schwingshackl 2014[15]	9	MeDiet	Type-2 diabetes	RR = 0.81 (0.73–0.90)	Quantitative meta-analysis: long-term studies were fairly homogenous (I^2 = 0%) and showed a stronger risk reduction RR = 0.75 (0.68–0.83)
Koloverou 2014[16]	17	MeDiet	Type-2 diabetes	RR = 0.77 (0.66–0.89)	Quantitative meta-analysis: subgroups by region, health status, and degree of confounding control rendered similar results.
Esposito 2010[33]	9	MeDiet	Type 2 diabetes and glycemic control	Beneficial	Qualitative systematic review
Esposito 2014[23]	5	MeDiet (observational)	Type-2 diabetes	Beneficial	Qualitative systematic review
Esposito 2014[23]	5	MeDiet (randomized trials)	Glycemic control	Beneficial	Qualitative systematic review
Georgoulis 2014[20]	17	MeDiet	Type-2 diabetes and other outcomes	Beneficial	Qualitative systematic review
Roman 2008[39]	20	MeDiet	CVD and risk factors	Beneficial	Qualitative systematic review
Widmer 2014[14]	Not stated	MeDiet and its components	CVD	RR = 0.95 (0.83–0.97)	Qualitative systematic review: favorably compared with pharmacologic interventions
Ros 2014[17]	Not stated	MeDiet	CVD	Beneficial	Qualitative systematic review: the results of the PREDIMED trial are presented in the context of their consistency with observational results.
Whayne 2014[19]	Not stated	MeDiet	CVD	Beneficial	Qualitative systematic review
Sofi 2008[37]	4	MeDiet (+2/9 points)	CVD	RR = 0.91 (0.87–0.95)	This meta-analysis was subsequently updated
Sofi 2010[32]	8	MeDiet (+2/9 points)	CVD	RR = 0.90 (0.87–0.93)	Quantitative meta-analysis: I^2 = 35%
Sofi 2014[21]	20	MeDiet (+2/9 points)	CVD	RR = 0.90 (0.87–0.92)	Quantitative meta-analysis: I^2 = 38%
Martínez-González 2014[22]	2	MeDiet (randomized trials)	CVD	RR = 0.62 (0.45–0.85)	Quantitative meta-analysis: I^2 = 55%
Martínez-González 2014[22]	16	MeDiet (observational, +2/9 points)	CVD	RR = 0.90 (0.86–0.94)	The heterogeneity disappeared after removing 3 studies assessing only fatal cases
Martinez-Gonzalez 2009[35]	5	MeDiet	CVD	Beneficial	Qualitative systematic review
de Lorgeril 2008[36]	Not stated	MeDiet	CVD	Beneficial	Qualitative systematic review
Rees 2013[26]	11		CVD	No evidence	

Table 1 (*continued*)

Systematic Review	N (Studies)	Exposure	Outcome	Effects of Increased Adherence to MeDiet [a]	Comment
		MeDiet (?)-only trials			The selection of trials apparently had little connection with the concept of MeDiet.
Panagiotakos 2004[42]	6	MeDiet	CHD	8%–45% relative risk reduction	Qualitative systematic review
Psaltoupolou 2013[27]	22	MeDiet	Stroke	RR = 0.71 (0.5–0.89)	Quantitative meta-analysis: meta-regression suggested stronger protection among males.
Tyrovolas 2010[34]	9	MeDiet	CVD and cancer	Beneficial	Qualitative systematic review
Martinez-Gonzalez 2004[41]	14	Feeding randomized trials	CVD and cancer	Unknown	The authors support the case for the urgent need of a trial such as PREDIMED
Sofi 2008[37]	8	MeDiet (+2/9 points)	All-cause mortality	RR = 0.91 (0.89–0.94)	This meta-analysis was subsequently updated
Sofi 2010[32]	9	MeDiet (+2/9 points)	All-cause mortality	RR = 0.92 (0.90–0.94)	Quantitative meta-analysis: I^2 = 33%
Sofi 2014[21]	18	MeDiet (+2/9 points)	All-cause mortality	RR = 0.92 (0.91–0.93)	Quantitative meta-analysis: I^2 = 47%
Trichopoulou 2000[43]	3	MeDiet	All-cause mortality	Longer survival	Qualitative systematic review
Serra-Majem 2006[40]	35	MeDiet	A variety of outcomes	Beneficial	Qualitative systematic review
Trichopoulou 2014[44]	Not stated	MeDiet	A variety of effects	Beneficial	Opinion of experts around the world
Maderuelo-Fernández 2014[13]	14	Interventions to promote MeDiet	Adherence to MeDiet	Beneficial	Qualitative systematic review: hard end-points were not assessed

(+2/9 points): effects associated with increasing 2 points in a 0–9 score of adherence to the MeDiet.
I^2: index to quantify heterogeneity in meta-analyses, please check Higgins et al. BMJ 2003;327:557–60.
Abbreviations: MeDiet: Mediterranean diet; CVD: cardiovascular disease; CHD: coronary heart disease; RR: relative risk (95% confidence intervals); WMD: weighted mean difference; CRP: C-reactive protein; IL-6: interleukin 6; ICAM-1: intercellular adhesion molecule; BMI: body mass index; BP: blood pressure.
[a] Risk ratios in meta-analyses of epidemiologic studies, usually adjusted for multiple confounders, compared the highest versus the lowest category of adherence to the MeDiet. Outcome changes describe the mean changes for the MeDiet versus comparator diets in meta-analyses of RCTs; only statistically significant changes are shown. Values between brackets are 95% confidence intervals.
[b] An apparent erratum was corrected. The authors presented the log of hazard ratio (95% CI) as −0.69 (−2.16 to −1.16), but this is impossible, the correct upper limit should probably be −0.16 (as we have assumed).

60–80 years (women). PREDIMED was a multicenter, nutritional intervention RCT carried out in Spain from 2003 to 2011. The study was funded by the official Spanish agency for scientific research, Instituto de Salud Carlos III.[12] A Web site (www.predimed.es) and the supplemental material published together with the final results[46] provide full details of the study protocol. We selected participants from >200 primary care facilities affiliated with 11 recruiting sites. All participants were at high risk for CVD, but had no history of previous CVD episodes at enrollment. Criteria for recruitment were the presence of either type 2 diabetes mellitus (T2DM) or ≥3 risk factors (smoking, overweight or obesity, hypertension/HTN, dyslipidemia/DLP, and family history of early-onset CVD). Participants were randomized into one of three diets: 1) MeDiet supplemented with extra-virgin olive oil (EVOO); 2) MeDiet supplemented with nuts; and 3) control diet (advice on a low-fat diet).

Full-time registered dietitians delivered the intervention. Throughout the study, participants attended quarterly individual visits and group sessions in which they were instructed to follow the allocated diets. Participants also attended quarterly group sessions where they received written material with information on key Mediterranean foods and seasonal shopping lists, menus and specific recipes for a typical week. This material was discussed in detail with the dietitians. Allotments of EVOO (1 L per week, including a minimum of 50 mL/day for participants and the rest for family needs) or mixed nuts (30 g/day: 15 g walnuts, 7.5 g almonds and 7.5 g hazelnuts plus extra allocations for the family) were supplied at no cost to each participant randomly assigned to the MeDiet groups on a quarterly basis during the group sessions with dietitians. Participants in the control diet group attended similar quarterly sessions with explanations and written material on the low-fat diet and they received non-food gifts in these sessions. The three diets were energy-unrestricted and no intervention on physical activity was conducted.

A validated 14-point MeDiet screener[47] was used by dietitians as a tool to both assess actual adherence to the MeDiet and enhance future adherence. These 14 items were:

1. Use of olive oil as the main culinary fat
2. Consumption of ≥4 tablespoons/d of olive oil (including oil used for frying, salads, out-of-house meals, etc.)

3. Consumption of ≥2 servings/d of vegetables
4. Consumption of ≥3 servings/d of fruits
5. Consumption of <1 serving/d of red meat, hamburger or meat products (ham, sausage, etc.)
6. Consumption of <1 serving/d of butter, margarine, or cream
7. Consumption of <1 serving/d of sweetened and/or carbonated beverages
8. Consumption of ≥1 serving/d of wine
9. Consumption of ≥3 servings/week of legumes
10. Consumption of ≥3 servings/week of fish or shellfish
11. Consumption of <3 servings/week of commercial sweets or pastries (not homemade), such as cakes, cookies, biscuits or custard
12. Consumption of ≥3 servings/week of nuts (including peanuts)
13. Preferential consumption of chicken, turkey or rabbit meat instead of veal, pork, hamburger or sausage
14. Consumption of ≥2 servings/week of sofrito, a sauce made with tomato and onion, leek or garlic and simmered with olive oil.

Validated food frequency questionnaires covering 137 foods were collected yearly by the dietitians. This repeated collection of dietary data allowed us to use the PREDIMED trial as a unique setting for subsequent cohort studies analyzed as a prospective observational follow-up study with repeated measurements of diet, thus improving the quality of our dietary assessment.[10]

Fasting blood and spot urine were obtained and serum, plasma and DNA samples were saved. Objective biomarkers of adherence to the supplemental foods (urinary hydroxytirosol as marker of EVOO consumption and plasma α-linolenic acid as marker of walnut consumption) were determined in random sub-samples.

The pre-specified primary end-point of the trial was incident CVD (a composite of non-fatal myocardial infarction/MI, non-fatal stroke or CVD death). This composite event occurred in 288 participants during a median follow-up of 4.8 years. The trial was neither powered nor designed to independently assess each of the three components of the combined end-point. Secondary outcomes included total mortality, T2DM, metabolic syndrome (MetS), peripheral arterial disease (PAD), atrial fibrillation (AF), neurodegenerative diseases and major cancers. An event adjudication committee, whose members were blinded to group allocation, was responsible for event ascertainment. All participants provided written informed consent and the protocol was approved by the institutional review boards of all participating centers.

Main results of the PREDIMED trial

We randomized 7447 participants into the three PREDIMED intervention groups. The groups were well-balanced with respect to their baseline characteristics and pharmacologic treatments. Though small, between-group differences in some baseline characteristics were observed, but they were not clinically meaningful. Furthermore, we adjusted all risk estimates for these variables. The mean age of participants was 67 years, 57% were women and the mean body mass index was 30 kg/m^2. The baseline prevalence of diabetes was nearly 50% and the prevalence of DLP and HTN was higher than 70% and 80%, respectively.

Compliance with the intervention in the two MeDiet groups was adequate.[48] Our tracking of objective biomarkers in random participant subsamples also indicated compliance with the intended dietary intervention. However, the achieved absolute difference in adherence to the MeDiet (according to the 14-item screener) between the intervention group and the control group was modest, amounting to a maintained difference of 2 points out of 14. There were no between-group differences in physical activity during the study. No diet-related adverse effects occurred.

We assessed the effect of baseline adherence to the 14-point score with respect to the subsequent incidence of the primary CVD end-point during follow-up.[49] As shown in Fig 1, the effect was remarkable. The multivariable-adjusted hazard ratio for participants with a baseline 14-item screener in the 2nd–3rd quintile who scored between 8 and 9 points was 0.72 (95% confidence interval [CI]: 0.55–0.94), and for those with the highest adherence (two upper quintiles, scoring 10–14 points) it was 0.47 (CI: 0.35–0.65).

The observed rates per 1000 person-years for the primary end point were 8.1, 8.0, and 11.2 in the MeDiet + EVOO, MeDiet + nuts, and control groups, respectively. The unadjusted hazard ratios were 0.70 (CI, 0.53–0.91) for the MeD + EVOO and 0.70 (CI, 0.53–0.94) for the MeDiet + nuts. The relative risk reductions, absolute risk reductions and number needed to treat are shown in Table 2 after multivariable adjustment for sex, age, adiposity variables, and baseline CVD risk factors. No effect on all-cause mortality was apparent. Significant disease risk reductions were also observed for incident T2DM (in the subset of participants initially free of T2DM)[50,51] and for other CVD outcomes, such as PAD[52] and AF.[53] Hence, the PREDIMED study showed with an RCT design for the first time that a MeDiet supplemented with either EVOO or nuts is useful in the primary prevention of CVD, PAD, AF, and T2DM in individuals at high risk.

A beneficial effect of the intervention on MetS status was also observed in the PREDIMED trial.[54,55] In comparison with the control group, participants randomized to either MeDiet were more likely to show reversion of MetS, with HR 1.35 (CI 1.15–1.58) for the MeDiet + EVOO, and HR 1.28 (CI 1.08–1.51) for the MeDiet + nuts. Similarly, the PREDIMED MeDiet interventions were shown to reduce blood pressure and the risk of HTN[56,57] and to slow the progression of subclinical atherosclerosis, as determined by changes in ultrasound-assessed carotid intima-media thickness and plaque.[58,59]

Number of events hypothetically prevented with the Mediterranean diet

Table 3 shows the number of hard clinical CVD events that could be prevented in a hypothetical cohort of 1000 persons undergoing the nutritional intervention with the MeDiet used in the PREDIMED trial. These results suggest that even a modest intervention with the MeDiet has the potential to

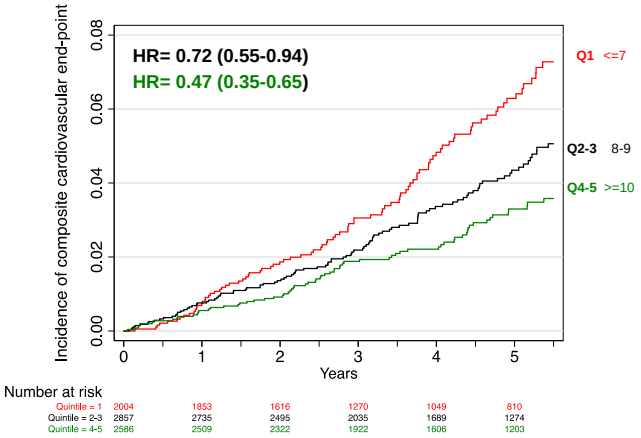

Fig 1 – Baseline adherence to the Mediterranean diet (14-point PREDIMED score) and incidence of the primary end-point in the PREDIMED trial (a composite of myocardial infarction, stroke or cardiovascular death). Q1-Q5: quintiles.

account for a sizable reduction in the number of clinical events in a relatively short period of time.

Observational studies and trials in the context of the PREDIMED trial

Although some authors have suggested that RCTs with hard clinical events as end-points are the only solution to circumventing the problems of measurement error inherent to observational designs in nutritional epidemiology, trials are far from perfect, and also present considerable limitations.[10] These problems include the frequent suboptimal compliance, losses to follow-up, and the ethical need to prematurely halt the trial when there is sufficient evidence of benefit, even when the number of observed events is lower than anticipated. In addition, some degree of contamination of the control group with aspects of the intervention intended only to the active intervention arms of the trial is unavoidable. Moreover some exposures or outcomes cannot be assessed with RCTs.

In this context, the symbiosis between properly designed RCTs and large cohort studies with appropriate and careful control of potentially confounding variables, and due precautions to improve dietary measurements, are currently the best possible option to ascertain the health effects of dietary exposures. Adequately designed and tested food frequency questionnaires (FFQs) have been shown to have acceptable validity when compared to reference measures. In addition, adjustment for total energy intake, usually applying the residual method along with the use of repeated FFQs in long-term prospective cohort studies, further improves their validity estimates.[9,10]

The availability of validated FFQs of each participant with yearly repeated measurements is a unique strength of the PREDIMED trial.[60] Furthermore, biomarker analyses have corroborated the validity of our dietary assessment tools.[61] Most follow-up studies have collected measurements of dietary intake only at baseline and this is a limitation in nutritional epidemiology because diet may change during follow-up. The PREDIMED study has provided a large body of evidence on the associations between diet and diverse health outcomes taking advantage of the validated FFQs.[62–83,47,84–98]

Mechanisms of protection by the Mediterranean diet

CVD protection by the MeDiet can be explained by a beneficial effect on classical and emergent CV risk factors.[56,99–101] Although the underlying mechanisms of protection against CVD by the MeDiet are not fully understood, the richness of this dietary pattern in antioxidant[101] and anti-inflammatory molecules[97] is likely to be relevant. On one hand, this can be due to their anti-oxidant capacity, such as cell redox state modulating enzyme systems. On the other hand, nutrients have the capacity of modulating gene and protein expression and, subsequently, metabolite production. Previous nutrigenomic studies have revealed that the MeDiet has a protective effect on the expression of several proatherogenic genes involved in vascular inflammation, foam cell formation, and thrombosis.[102,103]

Genomics and the Mediterranean diet

We investigated whether the effects of the MeDiet or its components might differ depending on genetic variants. We found several gene–diet interactions in determining both intermediate and CVD phenotypes.[104–108] Suffice it to say that we observed that the association of the MC4R rs17782313 or the FTO rs9939609 polymorphisms with T2DM was modulated by the MedDiet.[106] When adherence to the MedDiet was low (<9 out of 14 points), carriers of the variant alleles had higher T2DM risk than wild-type subjects. However, when adherence to the MedDiet was high (≥9 points), these associations disappeared. These gene–diet interactions remained after adjustment for BMI. Adherence to the MedDiet was found to interact with the TCF7L2-rs7903146 (C>T) polymorphism in relation to fasting glucose, total cholesterol, low-density lipoprotein cholesterol and triglycerides.[107] When adherence to the MedDiet was low, participants with the TT genotype had higher fasting glucose concentrations and lipids than CC + CT individuals but when adherence was high, these differences were not apparent. Moreover, TT subjects had a higher stroke incidence in the control group compared with CC, whereas the dietary intervention with MedDiet was associated with reduced stroke incidence in TT homozygotes but not CC homozygotes.[107] Both genetic and epigenetic effects on microRNA target site polymorphisms were also analyzed. A gain-of-function microRNA-410 target site polymorphism (rs13702T>C) in the lipoprotein lipase gene, interacted with the MedDiet intervention in the association with triglyceride levels and stroke incidence.[109] The interplay between genetic and epigenetic factors may contribute to better understand some biological mechanisms underlying CVD progression. Overall these results highlight the relevance of the multi-level omics approaches to a more comprehensive investigation of the mechanisms accounting for the MedDiet protective effects.

Table 2 – Relative risk reduction, absolute risk reduction and number needed to treat associated with the PREDIMED primary prevention intervention for several hard clinical events (assuming median follow-up = 4.8 years).

	Mediterranean Diet Supplemented With Extra-Virgin Olive Oil			Mediterranean Diet Supplemented With Mixed Nuts		
Clinical Event	Relative Risk Reduction	Absolute Risk Reduction	Number needed to treat	Relative risk Reduction	Absolute Risk Reduction	Number Needed to Treat
Primary CVD end-point	30% (8.0%; 46%)	1.34% (0.36%; 2.05%)	75 (49–281)	28% (4.0%; 46%)	1.25% (0.18%; 2.05%)	80 (49–562)
Type 2 diabetes	40% (15%; 57%)	3.52% (1.32%; 5.02%)	28 (20–76)	18% (–10%; 39%)	1.59% (–0.88%; 3.44%)	-
Peripheral artery disease	64% (35%; 79%)	1.18% (0.64%; 1.45%)	85 (69–155)	46% (8%; 68%)	0.85% (0.15%; 1.25%)	118 (80–679)
Atrial fibrillation	38% (12%; 55%)	1.54% (0.48%; 2.22%)	65 (45–206)	10% (–23%; 34%)	0.40% (–0.93%; 1.37%)	-

Fully adjusted estimates for the hazard ratios from Cox regression models were used to compute the relative risks (RR).
The relative risk reduction (RRR) was computed as RRR = (1 – RR)%.
The absolute risk reduction (ARR) was computed taking into account the baseline incidence of events in the control group (I_0) after a median follow-up of 4.8 years and applying the estimates for the relative risks, i.e. ARR = I_0 (1 – RR).

Conclusions

The findings from the PREDIMED trial, the Lyon Diet-Heart trial, and many large prospective cohorts are fully consistent. These large observational and experimental studies are also supported by mechanistic investigations aimed to assess classical and emergent CVD risk factors and pathophysiological pathways. Anti-inflammatory effects and reduced oxidative stress are very likely explanations for the protection observed in the PREDIMED trial. Taken together, these research findings converge, demonstrating that the traditional MeDiet offers an affordable, attractive, and easily achievable protection against CVD.

Importantly, these findings suggest that an overall dietary pattern that is rich in high-unsaturated fat from natural vegetable sources is preferable for CV health than a low-fat diet. In addition, the MeDiet has been shown to effectively control the residual risk observed after standard pharmacologic treatment of DLP anomalies and HTN in high-risk individuals. Taking into account the advanced age of many participants in the PREDIMED trial and in some of the available cohorts, it can be concluded that it is never too late to improve the food pattern to improve CV health.

Table 3 – Number of expected prevented cases with the PREDIMED primary prevention intervention for several hard clinical events (median follow-up: 4.8 years) in a hypothetical cohort of 1000 subjects. Both Mediterranean diet groups were merged together.

Clinical Event	Number of Prevented Cases (95% CI) per 1000 Hypothetical Participants Receiving the PREDIMED MeDiet-Intervention
Primary CVD end-point	13 (4–20)
Type 2 diabetes	26 (7–41)
Peripheral artery disease	10 (6–13)
Atrial fibrillation	11 (2–18)

Statement of conflict of interest

MA Martinez-Gonzalez received grants for research from the International Nut Council, Reus, Spain; J. Salas-Salvado has received grants for research from the International Nut Council, Reus, Spain and is a non paid member of its Scientific Advisory Committee; Ramon Estruch no conflicts of interest; D. Corella no conflicts of interest; M. Fito no conflicts of interest and E. Ros has received grants for research from the California Walnut Commission, Sacramento, CA, U.S. and is a non paid member of its Scientific Advisory Committee;

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