Accruing evidence on benefits of adherence to the Mediterranean diet on health: an updated systematic review and meta-analysis

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ABSTRACT
Background: The Mediterranean diet has long been reported to be protective against the occurrence of several different health outcomes.
Objective: We aimed to update our previous meta-analysis of published cohort prospective studies that investigated the effects of adherence to the Mediterranean diet on health status.
Design: We conducted a comprehensive literature search through electronic databases up to June 2010.
Results: The updated review process showed 7 prospective studies published in the past 2 y that were not included in the previous meta-analysis (1 study for overall mortality; 3 studies for cardiovascular incidence or mortality, 1 study for cancer incidence or mortality, and 2 studies for neurodegenerative diseases). These recent studies included 2 health outcomes not previously investigated (ie, mild cognitive impairment and stroke). The meta-analysis for all studies with a random-effects model that was conducted after the inclusion of these recent studies showed that a 2-point increase in adherence to the Mediterranean diet was associated with a significant reduction of overall mortality [relative risk (RR) = 0.92; 95% CI: 0.90, 0.94], cardiovascular incidence or mortality (RR = 0.90; 95% CI: 0.87, 0.93), cancer incidence or mortality (RR = 0.94; 95% CI: 0.92, 0.96), and neurodegenerative diseases (RR = 0.87; 95% CI: 0.81, 0.94). The meta-regression analysis showed that sample size was the most significant contributor to the model because it significantly influenced the estimate of the association for overall mortality.
Conclusion: This updated meta-analysis confirms, in a larger number of subjects and studies, the significant and consistent protection provided by adherence to the Mediterranean diet in relation to the occurrence of major chronic degenerative diseases. Am J Clin Nutr 2010;92:1189–96.

INTRODUCTION
The Mediterranean diet has long been reported to be the optimal diet for preventing noncommunicable diseases and preserving good health (1–3). The Mediterranean-style diet is not a specific diet, but rather a collection of eating habits traditionally followed by people in the different countries bordering the Mediterranean Sea. The diet refers to a dietary profile commonly available in the early 1960s in the Mediterranean regions and characterized by a high consumption of fruit, vegetables, legumes, and complex carbohydrates, with a moderate consumption of fish, and the consumption of olive oil as the main source of fats and a low-to-moderate amount of red wine during meals. A great deal of attention has been given to tools that estimate the adherence of individuals to the Mediterranean diet because of the usefulness of these tools to identify the whole dietary pattern instead of single foods or nutrients (4). Hence, computational scores have been created and used in several large epidemiologic studies to seek whether they could be useful to estimate the risk of disease in the general population (5). Some large epidemiologic studies conducted in different cohorts evidenced an association between a greater adherence to Mediterranean diet, a reduced risk of mortality, and the incidence of major chronic diseases (6–9). In 2008, we performed a meta-analysis that included all cohort prospective studies that investigated this issue, and we observed that a 2-point increase of adherence to Mediterranean diet conferred a significant protection against mortality, the occurrence of cardiovascular diseases, and major chronic degenerative diseases (3). However, the previous meta-analysis is 3 y old, and more prospective cohort studies have since been published.

Therefore, the aim of this study was to update our previous systematic review and meta-analysis conducted in cohort prospective studies that investigated the effects of adherence to Mediterranean diet on several health outcomes and to use these data to conduct a meta-regression analysis to explore a potential source of heterogeneity in studies.

METHODS

Literature search
According to the statement of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (10), we systematically searched published cohort prospective studies that investigated the association between adherence to the Mediterranean diet and health outcomes in electronic databases: MEDLINE (source: PubMed, 1966 to June 2010; www.pubmed.com), EMBASE (1980 to June 2010; www.embase.com), Web of Science (isieweb ofknowledge.com), The Cochrane Library (source: The Cochrane...
Central Register of Controlled Trials, 2010, issue 2; www.the cochranelibrary.com/), clinicaltrials.org, and Google Scholar (scholar.google.com). Relevant keywords relating to the Mediterranean diet in combination as MeSH terms and text words (“Mediterranean diet,” or “diet” or “dietary pattern” “Mediterranean,” or “adherence” or “score” and their variants) were used in combination with words relating to health status (“health,” or “mortality” or “morbidity,” or “cardiovascular diseases,” or “neoplastic diseases,” or “cancer,” or “neoplasm,” or “degenerative diseases,” or “Alzheimer’s disease,” or “Parkinson’s disease,” or “cerebrovascular disease,” or “dementia,” or “cognition disorder,” or “stroke,” or “outcome,” or “prospective,” or “follow-up,” or “cohort” and their variants). The search strategy had no language restrictions. References from the extracted articles and reviews were also consulted to complete the data bank. When multiple articles for a single study were present, we used the latest publication and supplemented it, if necessary, with data from the most complete or updated publication.

Study selection

We identified studies that prospectively evaluated the association of an a priori score used for assessing adherence to Mediterranean diet and adverse clinical outcomes. We excluded studies if they had a cross-sectional or case-control design, if they analyzed adherence to a nonspecific dietary pattern or to a recommended dietary guideline and not to Mediterranean diet, if they evaluated cohort of patients who suffered from a prior clinical event (ie, in a secondary prevention), if they did not adjust for potential confounders, and if they did not report an adequate statistical analysis.

Data extraction

Two investigators (FS and AC) assessed potentially relevant articles for eligibility. The decision to include or exclude studies was hierarchical and initially made on the basis of the study title, then of the study abstract, and finally of the complete study manuscript. Two researchers independently completed searches, identification of studies, data abstraction, and tabulation, and discordances were resolved by discussions. The following baseline characteristics were extracted from the original articles by using a standardized data extraction form and included in the meta-analysis: lead author, year of publication, cohort name, country of origin of the cohort, sample size of the cohort, number of outcomes, duration of follow-up, age at entry, sex, outcomes, components of the Mediterranean diet adherence score, and variables that entered into the multivariable model as potential confounders (Table 1).

Outcomes of interests were overall mortality, mortality and/or incidence from cardio- and cerebrovascular diseases, mortality and/or incidence from cancer, and incidence of neurodegenerative diseases.

Quality assessment

Study quality was assessed according to the following criteria: 1) number of study participants, 2) duration of follow-up, and 3) adjustment for potential confounders. Studies with a high number of participants, long duration of follow-up, and adjustment for confounders that include demographic, anthropometric, and traditional risk factors were considered of high quality.

Statistical analyses

We used the Review Manager program (RevMan, version 5.0.18 for Macintosh, 2008; The Cochrane Collaboration, Copenhagen, Denmark) and the Statistical Package for Social Sciences software (SPSS, version 18.0 for Macintosh; SPSS, Chicago, IL) to pool and analyze results from the individual studies. The methods and results of all recently identified cohort prospective studies were added to the previous table, and data were formally combined. Pooled results were reported as relative risks (RRs) and presented with 95% CIs with 2-sided P values by using a random-effects model (DerSimonian and Laird method) and the general variance-based method. P < 0.05 was considered statistically significant. When available, we used the results of the original studies from multivariate models with the most complete adjustment for potential confounders; the confounding variables included in the analyses are shown in Table 1.

Statistical heterogeneity was evaluated by using the I² statistic, which assessed the appropriateness of pooling the individual study results. The I² value provided an estimate of the amount of variance across studies because of heterogeneity rather than chance. Where I² was >50%, the heterogeneity was considered substantial. A small study bias or publication bias was appraised by visual inspection of the funnel plot of effect size against the SE and, analytically, by the Egger’s test. Moreover, to investigate possible sources of heterogeneity across studies, we performed a meta-regression analysis to investigate the effects of various characteristics of studies on the study estimates of RRs.

RESULTS

The updated search from recent years resulted in the identification of 7 additional prospective studies (11–17) published up to June 2010 that were not identified and included previously. Characteristics of these recent studies are displayed in Table 1. Of these, 1 study presented the overall mortality as a clinical outcome (14), 3 studies presented the incidence and/or mortality from cardiovascular diseases (13, 15, 16), 1 study presented the incidence and/or mortality from neoplastic diseases (17), and 2 studies presented the incidence of neurodegenerative diseases (11, 12). Notably, 2 of the 7 studies reported clinical outcomes not previously investigated, such as mild cognitive impairment and stroke (12, 13). On the other hand, one study was an updated analysis of a study already reported in the previous meta-analysis for the overall mortality outcome; therefore, only the most updated study was added to this updated final analysis (14). Taken altogether, a total of 18 cohort prospective studies were included and entered into the final analysis. As a whole, the updated analysis determined a study population that reached a total of 2,190,627 subjects analyzed. Included samples ranged in size from 161 to 485,044, with a follow-up time that ranged from 4 to 20 y.

Considerable variations among studies were observed in the number of participants, sex and age of participants, length of follow-up, and dietary components used to measure the adherence score to the Mediterranean diet. Studies conducted in
Table 1
Study characteristics of the recent prospective studies investigating adherence to the Mediterranean diet and health outcomes

<table>
<thead>
<tr>
<th>Author, year (cohort)</th>
<th>Country</th>
<th>Sample size/total cohort</th>
<th>Outcome</th>
<th>Follow-up</th>
<th>Age</th>
<th>Sex</th>
<th>Components of the adherence score</th>
<th>Adjustment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feart et al, 2009</td>
<td>France</td>
<td>50/1410</td>
<td>Alzheimer disease</td>
<td>5</td>
<td>68–95^2</td>
<td>M/F</td>
<td>1) High legume intake 2) High whole-grain products intake 3) High fruit intake 4) High vegetable intake 5) High fish intake 6) High MUFA:SFA ratio 7) Moderate alcohol consumption 8) Low red and processed meat intake 9) Low dairy product intake</td>
<td>Age, sex, BMI, hypertension, hypercholesterolemia, smoking habit, education, marital status, total energy, physical activity, medications, depression, apoE genotype, and stroke</td>
</tr>
<tr>
<td>Scarmeas et al, 2009</td>
<td>United States</td>
<td>241/1199 (only in those who were cognitively normal at baseline)</td>
<td>Mild cognitive impairment</td>
<td>4.5</td>
<td>76.7 M/F</td>
<td>M/F</td>
<td>1) High legume intake 2) High cereal intake 3) High fruit intake 4) High vegetable intake 5) High fish intake 6) High MUFA:SFA ratio 7) Moderate alcohol consumption 8) Low meat intake 9) Low dairy product intake</td>
<td>Age, sex, ethnicity, education, apoE genotype, total energy, and BMI</td>
</tr>
<tr>
<td>Fung et al, 2009</td>
<td>United States</td>
<td>2391/74,886</td>
<td>CHD, Stroke</td>
<td>20</td>
<td>38–63</td>
<td>F</td>
<td>1) High legume intake 2) High whole-grain products intake 3) High fruit intake 4) High vegetable intake 5) High fish intake 6) High MUFA:SFA ratio 7) Moderate alcohol consumption 8) Low red and processed meat intake</td>
<td>Age, smoking habit, BMI, menopausal status, hormone use, total energy, multivitamin intake, alcohol, family history, physical activity, and aspirin use</td>
</tr>
<tr>
<td>Trichopoulou et al, 2009</td>
<td>Greece</td>
<td>1075/23,349</td>
<td>Overall mortality</td>
<td>8.5</td>
<td>20-86 M/F</td>
<td>M/F</td>
<td>1) High legume intake 2) High cereal intake 3) High fruits/nuts intake 4) High vegetable intake 5) High fish intake 6) High MUFA:SFA ratio 7) Moderate alcohol consumption 8) Low meat and poultry intake 9) Low dairy product intake</td>
<td>Age, sex, education, smoking habit, waist:hip ratio, MET score, total energy, and BMI</td>
</tr>
</tbody>
</table>

(Continued)
<table>
<thead>
<tr>
<th>Author, year (cohort)</th>
<th>Country</th>
<th>Sample size/total cohort</th>
<th>Outcome</th>
<th>Follow-up</th>
<th>Age</th>
<th>Sex</th>
<th>Components of the adherence score</th>
<th>Adjustment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Martínez-González et al, 2010 (16) (SUN Study)</td>
<td>Spain</td>
<td>100/13,609</td>
<td>CVD</td>
<td>4.9</td>
<td>38</td>
<td>M/F</td>
<td>1) High legume intake 2) High cereal intake 3) High fruit/nuts intake 4) High vegetable intake 5) High fish intake 6) High MUFA:SFA ratio 7) Moderate alcohol consumption 8) Low meat and poultry intake 9) Low dairy product intake</td>
<td>Age, sex, family history of CVD, total energy, physical activity, smoking habit, BMI, diabetes, aspirin, hypertension, and hypercholesterolemia</td>
</tr>
<tr>
<td>Buckland et al, 2010 (17) (Spanish cohort of the EPIC-Heart Study)</td>
<td>United Kingdom, France, Denmark, Sweden, Germany, Italy, Spain, Netherlands, Norway, and Greece</td>
<td>449/485,044</td>
<td>Gastric adenocarcinoma</td>
<td>8.9</td>
<td>35–70</td>
<td>M/F</td>
<td>1) High legume intake 2) High cereal intake 3) High fruit/nuts intake 4) High vegetable intake 5) High fish intake 6) High olive oil intake 7) Moderate alcohol consumption 8) Low meat and poultry intake 9) Low dairy product intake</td>
<td>Age, sex, BMI, education, smoking habit, and total energy</td>
</tr>
</tbody>
</table>

1 MUFA, monounsaturated fatty acid; SFA, saturated fatty acid; CHD, coronary heart disease; EPIC, European Prospective Investigation into Cancer and Nutrition; MET, metabolic equivalent tasks; SUN, Seguimiento Universidad de Navarra; CVD, cardiovascular disease.

2 Range (all such values).

3 Mean.
European countries increased to 9 studies (50% of the total number of studies included).

By grouping the studies according to the different clinical outcomes, the overall mortality did not change for the number of studies evaluated (ie, 9 studies that included an updated analysis of the Greek cohort of the EPIC (European Prospective Investigation into Cancer and Nutrition) with a total of 514,118 subjects and 34,322 deaths, whereas a consistent increase of studies and subjects analyzed for the other clinical outcomes has been reported. Indeed, cardiovascular mortality and incidence were consistently augmented by the number of studies and subjects analyzed, with a total of 7 cohorts, 534,064 subjects, and 8739 deaths or incident cases. Similarly, the mortality from and incidence of neoplastic diseases reached 1,006,410 subjects and 11,378 events, whereas the number of studies that evaluated the association between adherence to the Mediterranean diet and neurodegenerative diseases increased from 3 to 5 cohorts, with a total of 136,235 subjects and 1074 cases, including an additional clinical outcome such as mild cognitive impairment.

**Meta-analysis**

Meta-analytic pooling under a random-effects model confirmed the already reported significant association between a 2-point increased adherence to the Mediterranean diet and a reduced risk of mortality from all causes (RR = 0.92; 95% CI: 0.90, 0.94; \( P < 0.00001 \)) (Figure 1) with no evidence of statistical heterogeneity across studies (\( I^2 = 33\%; P = 0.15 \)). When studies that evaluated different clinical outcomes were grouped, we observed that a 2-point increase of adherence to the Mediterranean diet still remained associated with a reduced risk of mortality from and incidence of cardiovascular diseases (RR = 0.90; 95% CI: 0.87, 0.93; \( P < 0.00001 \)) (Figure 2), which showed no significant heterogeneity across studies (\( I^2 = 35\%; P = 0.15 \)). Likewise, in studies that investigated the mortality from and incidence of neoplastic diseases (Figure 3), a greater adherence to the Mediterranean diet still determined a significant amount of protection, with a similar extent than that of previous meta-analysis (RR = 0.94; 95% CI: 0.92, 0.96; \( P < 0.00001 \)) and again no significant heterogeneity (\( I^2 = 6\%; P = 0.38 \)). The results of the previous meta-analysis were also confirmed for the occurrence of neurodegenerative diseases (Figure 4), which extended to mild cognitive impairment, with a 13% reduced risk of individuals who most adhered to the Mediterranean diet (RR = 0.87; 95% CI: 0.81, 0.94; \( P < 0.00001 \)) and no significant heterogeneity across studies (\( I^2 = 0\%; P = 0.73 \)).

**Publication bias**

The funnel plots of effect sizes compared with SEs that were performed to investigate possible publication biases were symmetric for all different clinical outcomes, which suggested the absence of possible publication biases.

**Meta-regression analysis**

Finally, to investigate the effects of various study characteristics on the study estimates of the RR, we conducted a meta-regression analysis by grouping studies according to some characteristics, such as the number of subjects studied, number of cases observed, country of origin of the patients, age of the patients, and length of follow-up. The sample size of the included studies was the only contributor to the protection compared with the overall mortality (\( \beta = 0.059 \pm 0.018; P = 0.013 \)) in the present model. By plotting the estimates of associations for overall mortality with the number of subjects analyzed for each study, it was shown that the average protection compared with the overall mortality was reached with studies that investigated >2000 subjects (Figure 5).

![FIGURE 1. Forest plot of the association between a 2-point increase of adherence score to the Mediterranean diet and the risk of all-cause mortality. The center of each square indicates the relative risk of the study, and the horizontal lines indicate 95% CIs. The area of the square is proportional to the amount of information from the study. The diamond indicates pooled estimates.](https://academic.oup.com/ajcn/article-abstract/92/5/1189/4597540)
In this updated meta-analysis of prospective studies that investigated the association between adherence to the Mediterranean diet and health status, findings were pooled from >2 million subjects and 50,000 deaths or incident cases from any causes and/or from any cardiovascular, neoplastic, and neurodegenerative diseases. We confirmed the results of our previously published meta-analysis, which suggested a significant protection against major chronic degenerative diseases for subjects who reported a greater adherence to the Mediterranean diet (3). Indeed, a 2-point increase of adherence to the Mediterranean diet significantly determined a 8% reduction of death from any causes, a 10% reduction from death and/or incidence of cardio- and cerebrovascular diseases, a 6% reduction from death and/or the incidence of neoplastic diseases, and a 13% reduction of the incidence of neurodegenerative diseases. Furthermore, in this updated meta-analysis, which included 7 articles published in the past 3 y that could not be considered in the previous meta-analysis, some other clinical outcomes, such as stroke and mild cognitive impairment, were included, and a meta-regression analysis to investigate the possible role of confounding factors was also conducted.

Several studies over the past decades showed the Mediterranean diet to be significantly associated with different health outcomes, such as a favorable health status, better biochemical profile, and quality of life (1–3, 26, 27). Accordingly, overwhelming evidence on the health effects of such a dietary profile has been reported in many different study cohorts by stimulating both scientific and lay communities to promote this dietary pattern as the ideal dietary pattern for preventing non-communicable diseases (3, 27).

### FIGURE 2

Forest plot of the association between a 2-point increase of adherence score to the Mediterranean diet and the risk of mortality from or incidence of cardiovascular diseases. The center of each square indicates the relative risk of the study, and the horizontal lines indicate 95% CIs. The area of the square is proportional to the amount of information from the study. The diamond indicates pooled estimates. CHD, coronary heart disease.

### FIGURE 3

Forest plot of the association between a 2-point increase of adherence score to the Mediterranean diet and the risk of mortality from or incidence of neoplastic diseases The center of each square indicates the relative risk of the study, and the horizontal lines indicate 95% CIs. The area of the square is proportional to the amount of the information from the study. The diamond indicates pooled estimates.
In this context, with the aim of identifying a practical tool to estimate how populations follow the guidelines of Mediterranean diet, adherence scores were computed (4). Many different scores were created and tested in different study cohorts, but the most important was definitely that created in 1995 and developed in its final form in 2003 by Trichopoulou et al (6, 18). In their studies, which investigated the Greek cohort of the EPIC study, Trichopoulou et al showed, for the first time to our knowledge, a significant association between an increase of such an adherence score and the reduction of overall mortality. Subsequently, many studies that used this kind of epidemiologic approach have been released by extending the evidence to many different health outcomes (3).

The results of the first meta-analysis published in 2008, which comprised a total of 12 studies and >1.5 million subjects, were that a 2-point increase of score that estimated adherence to the Mediterranean diet significantly determined protection compared with premature death and various diseases. In the past 3 y, the interest of researchers on this issue rose, and a total of 7 new cohort prospective studies were added to the literature (11–17). On the other hand, the issue has been shown to be of extreme interest for the general population as well as for health administrators and governments for better tailoring their prevention strategies and policies.

The present updated meta-analysis has some advantages with respect to the previous one: first, it present, to our knowledge, the most comprehensive and updated assessment on this issue, with inclusion of new studies and an update of previously reported studies; second, the association between the Mediterranean diet and health outcomes was extended to other relevant disease states that were not previously investigated, such as stroke and mild cognitive impairment. These clinical outcomes are of clinical relevance for the ever-increasing age of the general population. Third, it reinforces the findings that a slight increase of the adherence score to the Mediterranean diet, as was observed by an increase of 2 points of the adherence score, is significantly

**FIGURE 4.** Forest plot of the association between a 2-point increase of adherence score to the Mediterranean diet and the risk of incidence of neurodegenerative diseases. The center of each square indicates the relative risk of the study, and the horizontal lines indicate 95% CIs. The area of the square is proportional to the amount of the information from the study. The diamond indicates pooled estimates.

**FIGURE 5.** Meta-regression analysis of the relation between sample sizes of the studies and all-cause mortality. The area of the circle is proportional to the number of subjects studied in the study; the dashed line indicates the overall estimate of the association for studies that investigated all-cause mortality.
associated with a reduced risk of mortality and the incidence of main chronic degenerative diseases. Moreover, in the present updated meta-analysis, we performed a meta-regression analysis to give deeper insight into the nature of the association between the adherence score and health status. The meta-regression analysis showed that studies with larger study populations were more significantly associated with a lesser degree of heterogeneity and a higher estimate of association. In particular, studies with populations of >2000 subjects seemed to be more significantly related to the mean outcome observed in the meta-analysis. This finding could be easily explained by the increased power of the studies related to the higher number of subjects analyzed.

In contrast, although we believe that this updated meta-analysis provides useful information, the findings must be interpreted with caution because of some weaknesses. First, the short interval of time that passed from the first meta-analysis and the present meta-analysis, as well as the not so high number of new studies, did not allow us to be confident that these updated data can be used for all studied outcomes. Second, the use of meta-analysis technique to evaluate the protection of a dietary pattern, such as the Mediterranean diet, compared with the occurrence of several and different diseases can lead to an overestimation of the results. Third, the transferability of such an adherence score to the general population and the day-to-day clinical practice is not free of practical problems. The adherence score’s ease of use is low because the method of calculation implies the analysis of raw data that comes from a cohort study, and the real amount of each food category to consume is not established. Some relevant differences among the studies are present, and the transferability to the clinical practice is difficult to understand. However, with the use of such meta-analyses, we are able to understand the real estimate of the association between the adherence score to the Mediterranean diet and health status and to stimulate the interest of researchers to make uniform and standardize, if possible, the score and its components.

In conclusion, we updated the results of our previous meta-analysis and were able to confirm the estimates of the association of adhering to the Mediterranean diet in terms of protection against overall mortality and incidences of various chronic degenerative diseases, with an extension of the findings to some other health outcomes such as stroke and mild cognitive impairment. Moreover, the results of the meta-regression analysis indicated that the number of subjects enrolled in a population study is the most important contributor to the statistical model of association. These results will likely help researchers and health administrators to better tailor forthcoming studies in the field of nutrition and public health.

The authors’ responsibilities were as follows—FS and AC: conception and design of the study and drafting of the manuscript; FS, RA, GFG, and AC: analysis and interpretation of the data; FS; RA and GFG: critical revision of the article for important intellectual content; FS: statistical expertise and guarantor of the manuscript; and all authors: final approval of the manuscript. None of the authors had a conflict of interest.

REFERENCES