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The Mediterranean Diet and Physical Activity: Interaction Analysis and Assessment of a Mediterranean Lifestyle Score for the Prevention of Chronic Diseases and Premature Mortality

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Pamplona, Navarra, Spain
October 2021

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Memoria presentada por Dª Maria Soledad Hershey de la Cruz para aspirar al Grado de Doctor por la Universidad de Navarra en el Programa de Doctorado de Medicina Aplicada y Biomedicina.

El presente trabajo ha sido realizado bajo la Dirección de los siguientes Profesores en el Departamento de Medicina Preventiva y Salud Pública y autorizamos su presentación ante el Tribunal que lo ha de juzgar.

Pamplona, 15 de octubre de 2021

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To my grandfather

ACKNOWLEDGEMENTS

I would like to thank all those who made this doctoral dissertation possible and who have been a tremendous support throughout the years. Without each and every one of you this journey would not have been as enlightening and successful as it has been. I give thanks to God for accompanying me each step of the way and placing all you in my life.

To my supervisors: thank you **Dr. Miguel Ángel Martínez González**, the founder and director of this department, co-supervisor of my undergraduate, masters, and doctoral theses, for your abiding support since first offering me the opportunity to join *La Familia Preventiva* in 2016. Thank you **Dr. Miguel Ruiz-Canela López** for instilling in me your passion, commitment, and expertise, as well as your joyful disposition that has been so fundamental; you have helped me dream bigger and achieve more than I could have ever imagined.

To the entire **Department of Preventive Medicine and Public Health in the School of Medicine at the University of Navarra**: thank you to the professors, post-docs, PhD students, nurses, dietitians, collaborators, students, and visiting scientists for their friendship, conversations, collaborations, generous disposition, and good nature, which distinguish this department's positive atmosphere and strong sense of family. These include both those who have worked by my side in *edificio de Castaños* and *edificio de investigación*:

Thank you to those who I say goodbye to - Maira Bes Rastrollo, Alfredo Gea Sánchez, Estefanía Toledo Atucha, Nerea Martín Calvo, Carmen De la Fuente Arrillaga, Cristina Razquin Burillo, Jokin de Irala Estévez, Cristina López Del Burgo, Silvia Carlos Chilleron, Carmen Sayon Orea, Zenaida Vázquez Ruiz, Beatriz San Julián Aranguren, Leticia Goñi Mateos, Estíbaliz Goñi Ochandorena, Mónica Arias Colinas, Maite Sesma, Victor de la O, Inmaculada Aguilera, María Barbería Latasa, María José Cobo Díez, Octavio Pano Espinola, Carmen Lacarra Martín, Mario Gil Conesa, Matthias Henn, Rafa Pérez Araluce, Anacristina Rovayo, Vanessa Bullon Vela, Maria Vasilj, Arantxa Mata Fernández, Alejandro Fernández Montero, and María Jesús Tejedor Fernández, among others.

Thank you to those who have already said their goodbye - Ana Sánchez Tainta, Anaïs Rico, Pau Monzon, Irene Barrientos Ruiz, Maria Canales, Cristina Sánchez Quesada, Barbara Rocha, Elena Lorenzo, Marco Antonio Bernal, Christian Martinez Serrano, Liz Ruiz Estigarribia, Ismael Álvarez Álvarez, Gladys Ruth Morales, Ligia Domínguez, Mariana Muñoz García, Elisa Mercano, and Jose Luis Santos Martin, among others.

And a very special thank you to - Andrea Romanos Nanclares, Nacho Fernández Lázaro, Clara Gómez Donoso, Alice Radaelli, and Irene Talavera Rodríguez for their close friendships both in and out of the office.

To **Dr. Stefanos Kales**: thank you for hosting me at the Harvard TH Chan School of Public Health, Boston, Massachusetts to complete my predoctoral research stay and the invitation to return as a post-doc to continue this line of research within the Department of Environmental Health. I am very much looking forward to this next step in my career.

To **Dr. Mercedes Sotos Prieto**: thank you for your contribution to this dissertation by first inspiring this line of research on the Mediterranean lifestyle, providing the MEDLIFE index, and for offering thoughtful feedback on my work as well as professional insight. I could not be more excited to be working with you more closely in my post-doc.

To **Dr. J Alfredo Martínez Hernández** and the rest of the **Department of Food Sciences and Physiology in the School of Pharmacy and Nutrition at the University of Navarra**: thank you for providing me with a Degree in Human Nutrition and Dietetics and a European Master's Degree in Food Science, Nutrition and Metabolism (E-MENU). My distinguished nutrition background has and will continue to contribute a valuable perspective to the field of public health.

To the **SUN cohort participants, research team, and advisors** for their ongoing participation and collaboration with this ever-growing research endeavor.

To the **Feeding America's Bravest's participants and research team** for their incredible work to improve the health of US career firefighters through this intervention study.

To the **Asociación de Amigos Universidad de Navarra** for providing me with the financial support to carry out this doctoral thesis and the **Government of Navarra** for financing my research stay.

To **my family**: thank you Mom and Dad, for supporting my dream to pursue my undergraduate studies abroad, which led me to the University of Navarra. I take with me many experiences and much personal growth acquired in the past 8 and a half years from this decision. I am very thankful for my parents', brothers', and sister's support, time dedicated to help, and care for me. A special thank you to my sister who has lived with and accompanied me during these last years of my PhD. To my *abuela* and family in Madrid who provided me with much love and renewed energy through family encounters. And to my family in the US for all their support from a distance through phone calls and prayers.

And finally, to all of my professors and educators at all the academic institutions from which I have had the privilege of receiving an extraordinary education.

I want to express my deepest gratitude to all of you, and many more who I have not mentioned here, for helping me achieve this great accomplishment. It truly is a dream come true.

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ABBREVIATIONS

AHEI: Alternative Healthy Eating Index

AP: Attributable Proportion

BMI: Body mass index

CI: Confidence intervals

CVD: Cardiovascular disease

DALYs: Disability-adjusted life-years

DAMB: Data advisory monitory board

FAB: Feeding America's Bravest

FFQ: Food frequency questionnaire

GBD: The Global Burden of Diseases, Injuries, and Risk Factors Study

HALE: Healthy life expectancy

HDL: High-density lipoprotein

HEI: Healthy eating index

hs-CRP: High sensitivity C-reactive protein

HR: Hazard ratios

ICD: International Classification of Diseases

IFD: Indianapolis Fire Department

IRB: Institutional review board

LDL: Low-density lipoprotein

MADP: Mediterranean alcohol drinking pattern

MDS: Mediterranean diet score

MDNI: Mediterranean Diet Nutritional Intervention

MedDiet: Mediterranean dietary pattern

MEDAS: Mediterranean diet adherence screener

MEDLIFE: Mediterranean lifestyle

mMDS: Modified Mediterranean diet score

NSI: Spanish National Statistics Institute

OR: Odds ratio

PA: Physical activity

PSM: Public Safety Medical

pts: points

Q: Quartile

R^2: the coefficient of determination (R-squared)

RCT: Randomized controlled trial

RERI: Relative excess risk due to interaction

SD: Standard deviation

SDG: Sustainable Development Goals

SI: Synergy Index

S-NDI: Spanish National Death Index

SUN: Seguimiento Universidad de Navarra

T2D: Type 2 Diabetes

TG: Triglycerides

US: United States of America

USDA: United States Department of Agriculture

VO2max: Maximum rate of oxygen consumption

WHO: World Health Organization

YLDs: Years of life lived with disability

YLLs: Years of life lost

INTRODUCTION

Epidemiology is the science that studies disease occurrence and health states in human populations.[1] Epidemiology represents a multifaceted and interdisciplinary field of medicine which assesses the effect of exposures, including causal inference, on any health-related outcome that impacts the overall health of a given population.[1–3] In recent decades, epidemiological studies have sought to better understand the causality of some detrimental exposures (i.e. poor nutrition, lack of exercise, pollution, inadequate sleep, smoking) and have proposed public health strategies for mitigating the world's current non-communicable or chronic disease (NCDs) epidemic.[1,4] A variety of highly prevalent NCDs are largely responsible for both morbidity and mortality worldwide. Whereas morbidity is the state of being symptomatic or unhealthy for a disease or condition, mortality is related to the number of deaths caused by the health outcome, presented either as an absolute number or "per 1000" rate of the given population.[4] Additionally, in public health it is important to capture fatal outcomes in a summary measure of average levels of population health, such as life expectancy.[5]

At the population-level, in addition to the attributable mortality, mortality rates, and life expectancy, other health metrics estimate the population impact of disease and disease risk factors, not only fatalities. These metrics include years of life lost due to premature mortality (YLLs), years of healthy life lost due to disability (YLDs), and disability-adjusted life-years (DALYs).[6] These measures help understand the burden that a health outcome may place on a population and offer stakeholders some direction as to how health outcomes can be more effectively prioritized to design and proactively implement prevention strategies, and properly allocate the pertinent resources to each alternative strategy.[4] Furthermore, the population impact of morbidity and cause-specific mortality have been recently evidenced using these estimated measures within The Global Burden of Diseases, Injuries, and Risk Factors Study (GBD).[7]

Global Burden of Disease

Life Expectancy

The GBD annually applies a systematic effort to quantify the comparative magnitude of health loss due to diseases, injuries, and risk factors by age, sex, and geography over time. In 2019, that study observed 31 (86%) of 36 high-income countries had an observed male life expectancy at birth higher than expected. Moreover, Spain, Portugal, Israel, Italy, Singapore, and Malta had estimated more than 5 years longer male life expectancy at birth than expected. However, despite this positive measure of population health, Spain was one of the seven high-income countries to experience declines in total population, alongside Greece, Greenland, Andorra, Japan, Portugal, and Italy.[8] Similarly, 80.6% (29 out of 36 countries) of the high-income GBD region observed a higher female life expectancy at birth than expected. Yet, on the contrary, five countries in this high-income region; Brunei, Greenland, Monaco, the United States of America (US), and Germany, had shorter than expected male life expectancy at birth. The US in particular observed a difference of −1.1 years between the observed and the expected male life expectancy in 2019, in line with previous analyses on the increasing harmful effect of obesity and cardiometabolic multimorbidity on longevity.[8–10] The study of both global and region-specific health metrics offer opportunities to improve and emulate countries that are performing well.[7] In particular, this information can provide insights towards effective public health policies and how to achieve global health goals, such as the United Nations' Sustainable Development Goals (SDG) and the World Health Organization's (WHO) Global Action Plan for the Prevention and Control of Noncommunicable Diseases.[11,12]

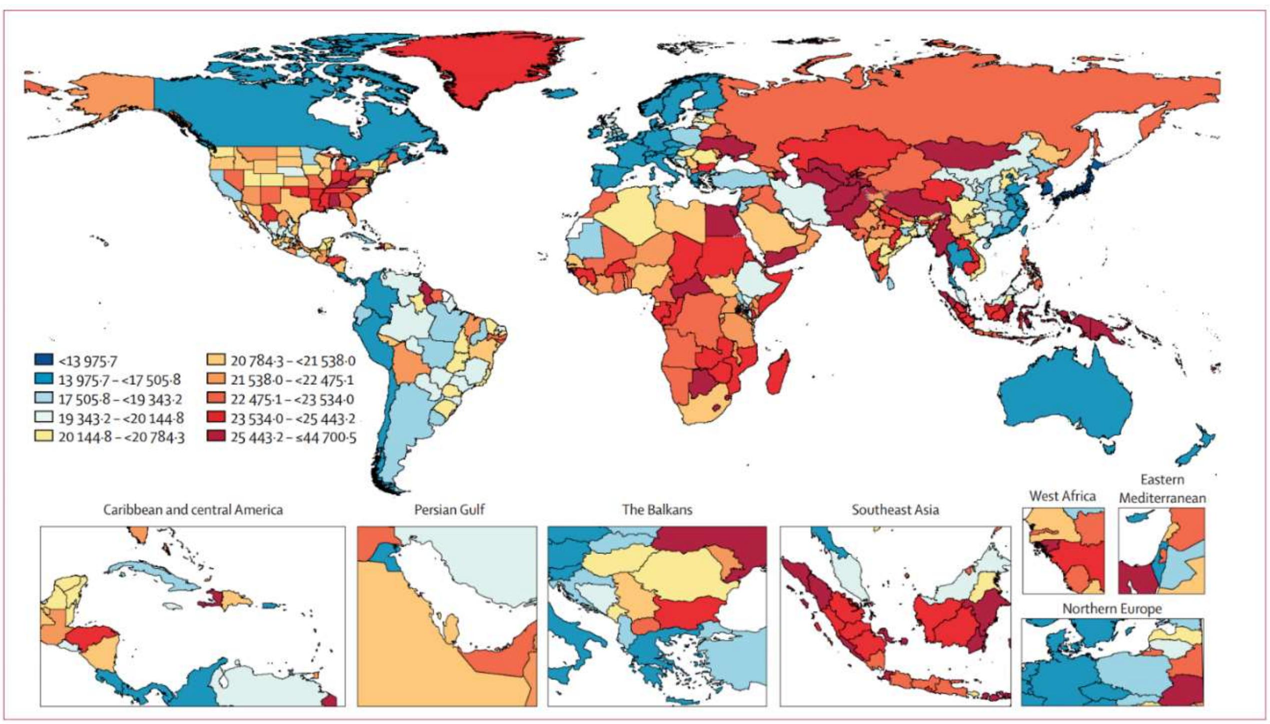
As evidenced by the most recent GBD findings, population demographic trends point towards rising mean age due to dropping fertility and birth rates as life expectancy simultaneously increases. The skyrocketing aging of Western populations will be paralleled by insurmountable challenges due to the burden of chronic disease and disability (i.e. diabetes, stroke, or myocardial infarction) and will place the sustainability of most health systems in severe compromise, and ultimately reduce life expectancy.[10,13] This situation is particularly severe in Spain after several decades of maintaining a synthetic index of fecundity below 1.5.

This demographic and epidemiologic situation has driven strong concerns regarding the quality of life of individuals at later stages of life as chronic disease risk increases.[7,8] Two hundred and four GBD countries and territories have all seen numbers of years spent in poor health increase, even though both life expectancy at birth and healthy life expectancy (HALE) at birth have improved at the same time.[8] HALE is the average number of years that a person can expect to live in "full health" by taking into account years lived in less than full health due to disease or injury, which is calculated as all-cause YLD rate per capita, adjusted for independent comorbidity, by age, sex and country.[5] This means that while age-standardized DALY rates have improved, the population growth and aging has increased the burden of DALYS at older ages, maintaining absolute DALYs worldwide constant.[7] These aging population trends have real policy issues to consider, in addition to individuals' quality of life, such as the effect on economic growth as the size of workforces decline and retired populations grow, as well as the overall strain on health systems. Such trends require public health strategies to promote overall health and wellbeing of these populations.[8]

Noncommunicable Diseases

Since 1990, YLDs from NCDs and injuries have become increasingly greater contributors of burden worldwide.[7] In 2019, NCDs were estimated to be responsible for 1.62 billion DALYs (95% uncertainty intervals (UI): 1.43 – 1.82), an increase from 43.2% (40.4 – 45.7) of total DALYs in 1990 to 63.8% (61.4 – 66.0) of total DALYs in 2019, varying by demographic region (Figure 1).[7] These DALYs from NCDs considered death and disability resulting from cardiovascular disease (CVD), musculoskeletal and mental disorders, and cancer. In particular, DALYs from diabetes increased by more than 80% between 2000 and 2019, indicative of the rapidly increasing prevalence of type 2 diabetes (T2D) in association with the obesity pandemic.[7,14] These diseases have become a key priority for healthcare systems since they significantly increase healthcare costs and reduce life expectancy, while being largely preventable with lifestyle.[13,15]

Figure 1. Age-standardized DALY rates (per 100,000) from NCDs by location, both sexes, 2019.



Source: http://www.healthdata.org/results/gbd_summaries/2019/non-communicable-diseases-level-1-cause

Established in 2015 by the United Nations General Assembly, the 2030 SDGs aim to reduce premature mortality from NCDs by one-third relative to 2015 levels and to promote mental health and wellbeing. In light of the foreseen shortage to meet this goal by most countries, a NCD Countdown 2030 is currently holding stakeholders, including *The Lancet*, public health scientists, civil society leaders, and the WHO, accountable for monitoring and reviewing the progress made towards the prevention, treatment, and controlling of NCDs.[16] In addition, the WHO has established an indicator across countries of the existence of at least one operational multisectoral national policy, strategy or action plan that integrates several NCDs and shared risk factors. Other indicators monitored by the WHO for the prevention and control of NCDs include measures to reduce tobacco demand, harmful use of alcohol and unhealthy diets, and promote physical activity (PA).[17] Nonetheless, the GBD has shown that there has been no real progress in reducing exposure to modifiable behavioral risk factors, meanwhile metabolic risks are, on average, increasing every year.

Causes of Morbidity and Mortality

NCDs are the leading cause of death globally, primarily due to the four main NCDs; CVD, diabetes, cancer, and respiratory disease, and their main risk factors; tobacco use, unhealthy diet, physical inactivity, excess body weight, and harmful use of alcohol.[17] More specifically, the global leading causes of death in 2019 included ischemic heart disease, stroke, chronic obstructive pulmonary disease, lower respiratory infections, neonatal conditions, cancers, neurological disorders, diarrheal diseases, diabetes, and kidney diseases. In 2019, these seven NCDs accounted for 44% of all deaths, or, 80% of the top 10 causes of death. Ischemic heart disease was responsible for the largest increase in leading causes of death when considering the global absolute change between the years 2000 and 2019.[18] Furthermore, all NCDs together accounted for 74% of deaths.

In addition to geographical location or country, leading causes of death may vary by sociodemographic characteristics, such as age, ethnicity, sex, and socioeconomic status. Among NCD deaths, millions occur before the age of 70 years, defined as "premature" deaths, primarily from cancer and CVD, and disproportionately affecting low- and middle-income countries.[8] In Spain, the leading causes of death in 2019 varied between sexes, identifying cancer as the leading cause among males, whereas CVD was the leading cause of death in females (Figure 2).[19] Hence, despite population differences, mortality from NCDs continues to increase worldwide.

Figure 2. Leading causes of mortality in Spain by sex, comparison between 1990 and 2019.

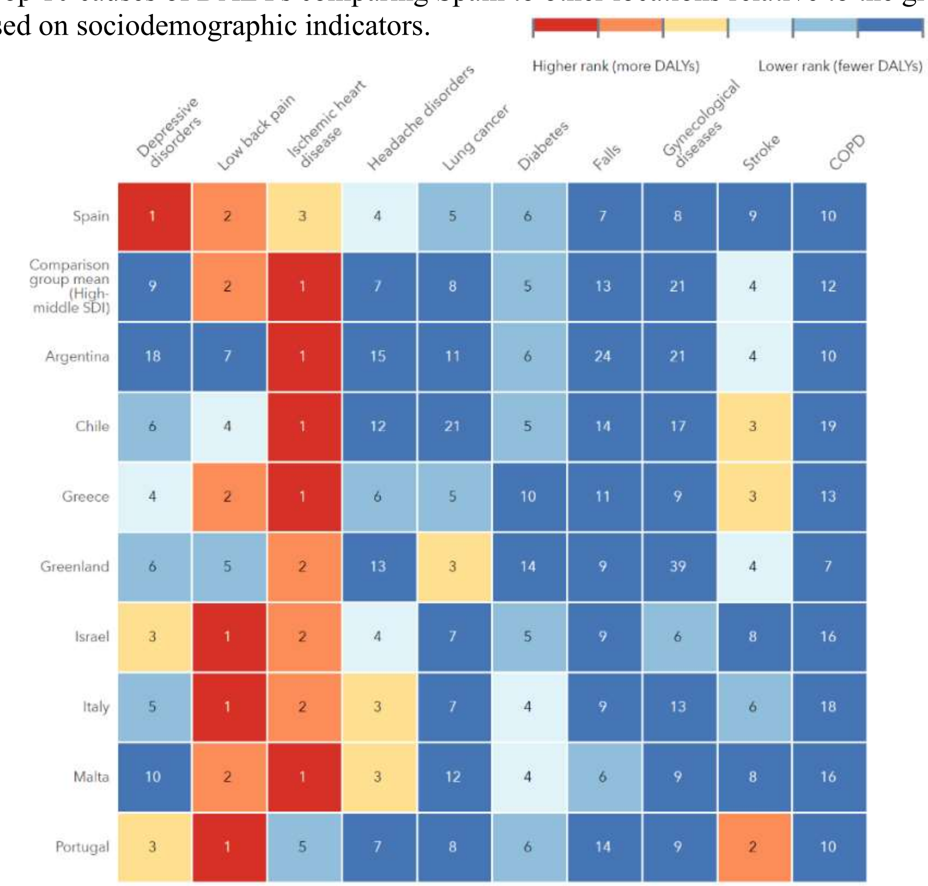
Source: self-constructed at https://vizhub.healthdata.org/gbd-compare/

The GBD identified ischemic heart disease, stroke, and diabetes as major contributors to the global burden of disease, first emerging in the 25-to-49-year age group and even more so in the older age groups.[7] In particular, ischemic heart disease and stroke were the leading causes of DALYs in ages 50 years and older. Additionally, the rising prevalence of diabetes, which is linked to increases in body mass index (BMI) and has the second largest negative total effect on reducing HALE worldwide, has hindered the pathway to reducing the burden of CVD.[7,20] Chronic kidney disease is also strongly related to CVD, diabetes, and obesity, sharing common risk factors and intervention strategies.[7]

The largest contributors to disability, including mental disorders, are associated with few deaths. Nonetheless, depression has been linked to severely impaired quality of life and increases in mortality among US adults between the ages of 20 and 50 years, described as deaths of despair resulting from suicide, accidental poisonings (including opioids), and chronic liver disease or cirrhosis, with a similar pattern for both males and females.[21] The particular subgroup of American whites aged 45 to 55 years in rural counties were most likely to die prematurely due to these deaths of despair, as well as observe increases in death from chronic diseases, such as cancer and CVD, which was not the same case for older populations aged 55 to 65 years in the same setting.[21] Hence, age-specific mortality rates can also provide important evidence on where new diseases are emerging or adverse risk factor trends are creating an impact.[8]

Depression in particular is a leading cause of morbidity, estimated to affect over 4.4% of the world's population in 2015, increasing by 18.4% between 2005 and 2015, and ranked as the third leading contributor to global disability (DALYs due to depressive disorders increased by 14.3% from 2007 to 2017) for both males and females combined.[22,23] Meanwhile, according to the most recent data from the 2020 European Health Survey in Spain (EESE), the prevalence of depression among the Spanish population above the age of 15 years was 5.3%.[24] Depression was ranked a more important cause of death and disability in Spain when compared to other countries comparable in sociodemographic indicators, ranked as the first cause for DALYs (Figure 3).[7] Although depressive disorders maintained second in ranking among level three causes of YLDs (GBD classifies causes in a hierarchy of four levels; Level 3 includes specific causes such as tuberculosis, stroke, and road injury), depression was responsible for a 28% change in total YLD in Spain from 1990 to 2019.[19] Therefore, as disability becomes an increasingly large component of disease burden and increased healthcare costs, further research is needed to identify new, and more effective, intervention strategies to reduce the emergence of such contributors.

Figure 3. Top 10 causes of DALYs comparing Spain to other locations relative to the group average based on sociodemographic indicators.



Age-standardized DALY rate per 100,000, 2019 COPD: chronic obstructive pulmonary disease

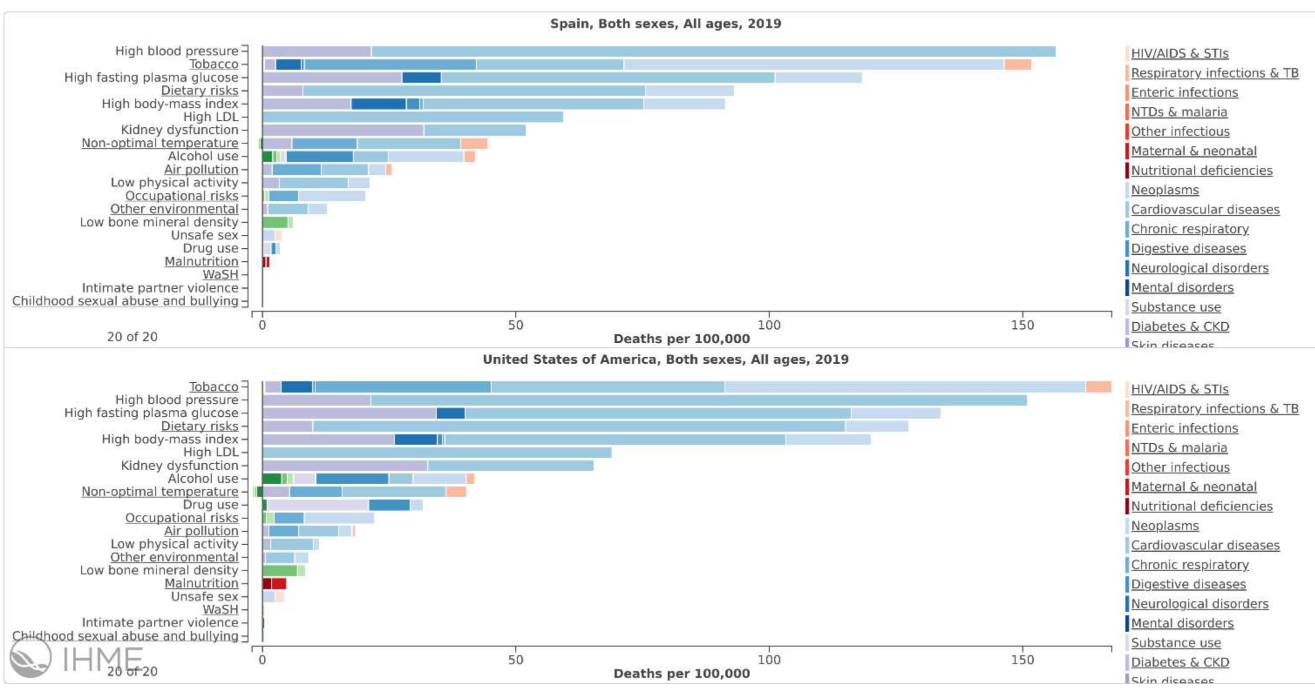
Source: http://www.healthdata.org/spain

Risk Factors for Morbidity & Mortality

Behavioral Risk Factors

According to the GBD, the change in leading risk factors by DALYs between 1990 and 2019 indicated an increase in numerous metabolic and behavioral risk factors. The largest increases in risk exposures within this time period were from ambient air pollution, drug use, high fasting plasma glucose, and high BMI.[6] Globally, metabolic risks (high BMI) and behavioral factors (inappropriate diet, smoking, and physical inactivity) contributed the most to the attributable death and DALYs from leading NCDs.[20] In particular, the leading risk factor for attributable deaths globally was high systolic blood pressure, which accounted for 10.8 million deaths (95% UI: 9.51 – 12.1), equivalent to 19.2% (16.9 – 21.3) of all deaths in 2019. This was followed by tobacco (smoked, second-hand, and chewing), accounting for 8.71 million deaths (8.12 – 9.31) or 15.4% (14.6 – 16.2) of all deaths, proceeded by high fasting plasma glucose, high BMI, and high low-density lipoprotein (LDL) cholesterol. Although the order varied slightly between Spain and the US, the leading risk factors for mortality in these countries in 2019 were high blood pressure, tobacco, high fasting plasma glucose, dietary risks, and high BMI (Figure 4). The globalization of unhealthy lifestyle behaviors, particularly those related to diet quality, caloric intake, smoking, and PA, are determinants of many metabolic risks and thus a current priority for public health authorities.[6]

Figure 4. Behavioral and metabolic risk factors for all-cause mortality comparing Spain and the United States of America, both sexes, 2019.



Source: self-constructed at https://vizhub.healthdata.org/gbd-compare/

Cardiometabolic Factors

In the clinical setting, in order to address ideal cardiovascular health, a cluster of metabolic factors has been employed to clinically identify those persons at high risk of CVD and T2D. Although various diagnostic criteria have been proposed over the past decades, all definitions of metabolic syndrome fit the definition of a syndrome; a clustering of factors that frequently occur together, more often than by chance alone, and for which the cause is often uncertain.[25] In this regard, metabolic syndrome, according to the harmonized definition established in 2009 by the International Diabetes Federation, American Heart Association, National Institute of Health, International Atherosclerosis Society, World Heart Federation and International Association for the Study of Obesity, is a condition of clustered cardio-metabolic CVD and T2D risk factors, which requires at least three out of five criteria:[26]

- hyperglycemia; ≥100 mg/dl or treatment with antidiabetic drugs
- raised blood pressure; systolic ≥130 mm Hg or diastolic ≥85 mm Hg, or receiving antihypertensive drugs
- elevated triglyceride levels; ≥150 mg/dl
- low high-density lipoprotein (HDL) cholesterol levels; <40 mg/dl in men or <50 mg/dl in women
- abdominal obesity; waist circumference ≥102 cm in men or ≥88 cm in women (limits vary according to race).

Metabolic syndrome was a better predictor of CVD and total mortality than each of its individual components. Patients with metabolic syndrome are at twice the risk of developing CVD over the next five to ten years than individuals without the syndrome.[26] Furthermore, according to a representative sample of the US population from the National Health and Nutrition Examination Survey (NHANES), the prevalence of metabolic syndrome increased from 32.5% from 2011-2012 to 36.9% in 2015-2016, with significant increases in those aged 20-39 years, women, Asians, and Hispanics.[27] Metabolic syndrome has been largely attributed to an overconsumption of calories, in addition to more sedentary lifestyles worldwide, which has led to increasing obesity rates.[28] Therefore, metabolic syndrome is a useful example of the importance of multiple targets for effective preventive interventions to lower CVD and T2D risk, as well as subsequent healthcare costs and disability.[25–28]

Referred to as Life's Simple 7, ideal cardiovascular health has also been defined by the American Heart Association as the presence of both[29] –

ideal health *behaviors*:

- nonsmoking
- body mass index <25 kg/m^2
- PA at goal levels
- pursuit of a diet consistent with current guideline recommendations

and ideal health *risk factors:*

- untreated total cholesterol <200 mg/dL
- untreated blood pressure <120/<80 mmHg
- fasting blood glucose <100 mg/dL

This definition of cardiovascular health includes both behavioral and metabolic risk factors. Yet, behavioral risk factors, such as tobacco use, unhealthy diet, and physical inactivity, are causes themselves of NCD risk factors, including raised blood pressure, overweight/obesity, raised blood glucose, and raised cholesterol. Given the promotion of lifestyle factors offers an easier public health message for the general population, without the involvement of a clinician, greater emphasis should be placed on prevention strategies addressing modifiable behavioral risk factors.

Diet and Lifestyle

Diet

While it is known that many of the determinants of NCD risk factors, such as smoking, an unhealthy diet, lack of PA, and excessive alcohol consumption are largely avoidable, unhealthful modifiable lifestyle factors continue to increase worldwide.[30] The disease burden of poor diet quality has globally increased during the last 30 years.[31] Dietary risks (measured as the joint effect of 15 aggregate GBD dietary components; i.e. low intake of fruits, vegetables, fiber, and seafood omega-3, and high intakes of processed meats, sugar-sweetened beverages, and sodium, among others) were responsible for 188 million (95% UI: 156 – 225) DALYs and 7.94 million (6.47 – 9.76) deaths among adults aged 25 years and older in 2019.[6] Industrialized agriculture, ultra-processed foods, unethical conduct of some Food and Soda multi-national corporations, specific cultural forces (consumerism, hedonism, relativism), and the globalization of the *Westernized* diet are all implicated in the increased prevalence of NCDs and are addressable through several SDGs.[11]

During these past three decades, the development of nutritional epidemiology has been impressive.[32] A key contribution to this field has been the shift of focus from assessing isolated dietary factors to studying the effects of overall or complete dietary patterns.[33] This dietary pattern analysis considers the complex interactions among nutrients and foods, as well as the cumulative effect of all the individual components.[34] Food patterns can be defined as the quantities, proportions, variety, or combination of different foods and drinks in diets, and the frequency with which they are habitually consumed. *A priori* defined dietary indices, constructed to measure adherence to specific dietary patterns as indicators of overall diet quality, have allowed epidemiologists to establish inverse associations between a healthy food pattern and multiple health outcomes.[35–37] Research has also considered the effect of improving dietary quality by estimating long term measures of health. For instance, one study evidenced a dose-response pattern for healthier diet quality, according to the alternative healthy eating index (AHEI)-2010, with cardiometabolic disease-free life expectancy between the ages of 50 and 85 years.[38] Thus, health promotion strategies should be intervening on the factors that influence individuals to consume a low-quality diet, including a lack of knowledge, lack of availability, high cost, time scarcity, social and cultural norms, marketing of poor-quality foods, and palatability.[39]

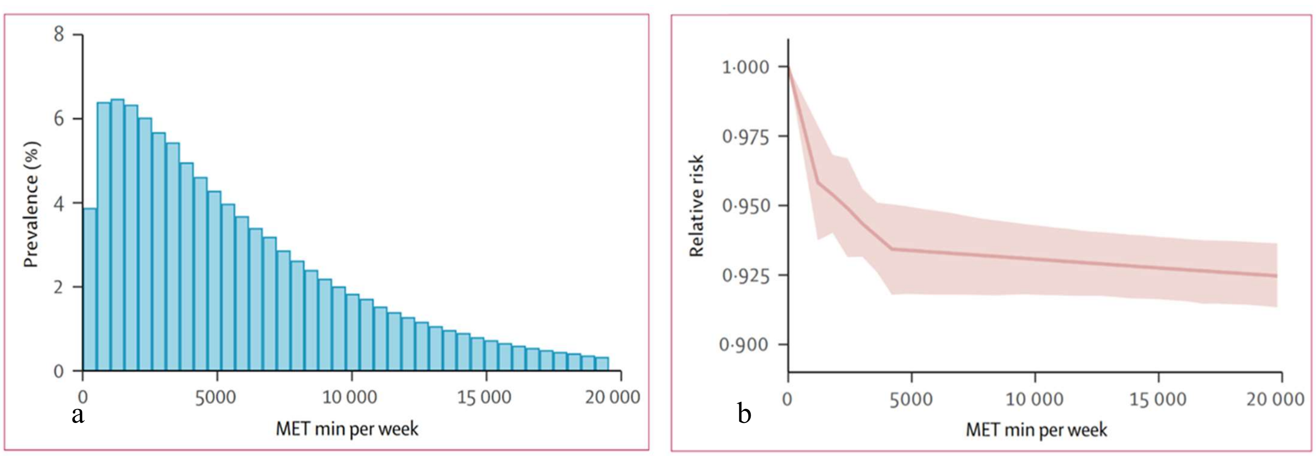
Evidence of the Mediterranean diet (MedDiet) in particular has attracted worldwide attention and has supported the global effect produced by the sum of its components, rich in antioxidants and anti-inflammatory properties, could partially explain the higher life expectancy in the Mediterranean region.[40,41] The frequency and quantity of food consumption, as well as dietary habits, that characterize the MedDiet have led this dietary pattern to be widely praised for its nutritional adequacy, high diet quality, and beneficial effects on several health outcomes, in particular for the prevention of chronic disease.[42–50] These health benefits include reduced incidence of CVD, total mortality, diabetes, metabolic syndrome, obesity, specific types of cancer, and cognitive function.[51] In this context, the MedDiet is internationally recognized as one of the best dietary strategies for the prevention of chronic diseases and premature death.[52–56]

Physical Activity

A lack of PA is also a major and globally relevant determinant of health.[57] There is abundant evidence of the effect of PA on health for the prevention of chronic diseases and premature mortality, whereas a lack of PA is a key risk factor for these health outcomes.[58–61] It has been demonstrated that the replacement of PA or exercise with inactivity or sedentary behavior will eventually adversely affect the aging process, whatever the age of the individual. Even a simple indicator of PA, such as time spent sitting, is an independent predictor of mortality. The increase in risk of lifestyle-related and age-associated chronic diseases are attributed to the decline in functional levels of many body systems and thus suboptimal maintenance of physiological functions in sedentary individuals.[61]

As the global lifestyle has become increasingly sedentary, greater interest has been placed on understanding the relationship between PA and health outcomes (Figure 5).[62] A recent systematic umbrella review evaluated the associations of moderate-to-vigorous PA with all-cause and CVD mortality, incident coronary artery disease, ischemic stroke and all-cause heart failure. The clear dose-response relationship showed there was no lower threshold for effect, but rather a steep, early slope with about 70% of the benefit obtained by PA alone reached at 8.25 MET-h/w (150 minutes of “brisk walking”) and there was no apparent upper threshold nor evidence for increased risk at the greatest amounts of PA.[63] Furthermore, increasing leisure time PA later in adulthood was associated with mortality benefits similar to those who maintained higher levels of PA across the adult life course, thus it is not too late for adults to improve their PA habits and begin an active lifestyle.[64]

Figure 5. a) Percentage of GBD population exposed to risk factor, b) All-cause mortality relative risk associated with average weekly PA, both sexes combined, 2019.



Source: http://www.healthdata.org/results/gbd_summaries/2019/low-physical-activity-level-2-risk

Diet & Physical Activity

Diet and PA are two of the most frequently addressed modifiable lifestyle risk factors, which increase morbidity and mortality from lifestyle diseases, including CVD, obesity, T2D, and some cancers. Hand-in-hand, diet and PA are frequently recommended in clinical practice for general health promotion, weight loss or weight maintenance, chronic disease prevention, and increased quality of life.[65,66] An ideal dietary pattern and PA are considered multidimensional variables that can influence each other.[67] According to data from NHANES 2003-2006, US adults were 32% more likely to eat a healthy diet if they met PA guidelines.[68] When considering the energy balance equation, diet (pertinent to energy intake) and PA (pertinent to energy expenditure) found themselves on either side of the equation, suggesting that both factors influence each other to maintain a healthy weight, possibly more so than the sources of energy themselves.[67,69,70] This concept has already been incorporated into the MedDiet Pyramid, which underlines the importance of PA and other lifestyle factors beyond diet.[71,72]

Defined as adherence to both the MedDiet and PA, the current scientific literature already supports the public health promotion of a Mediterranean lifestyle for better health biomarkers, decreased initiation of CVD medications, lower risk of disease, and lower mortality beyond those acquired from the MedDiet or PA alone.[48,53,54,73–76] A meta-review from 9 systematic reviews and 24 meta-analyses concluded that the MedDiet may reduce the risk of NCDs, improve health status, and reduce total lifetime healthcare costs, with a possibly even greater effect when combined with PA, as long as tobacco and excessive alcohol consumption are avoided.[48] Furthermore, a pooled analysis of 11 randomized controlled trials observed strong evidence suggesting the MedDiet and PA resulted in greater beneficial changes in ten out of eleven metabolic risk factors, (i.e., body weight, BMI, waist circumference, systolic and diastolic blood pressure, HOMA-IR index, and blood glucose, triglycerides (TG), and total- and HDL-cholesterol) compared to a control condition, indicating a metabolic risk reduction in an adult population.[76] A recent analysis within the PREDIMED study evidenced that the combination of high leisure time PA levels and a normo-caloric MedDiet adherence was synergistically associated with a delayed initiation of CVD related drug therapy.[75] Although the statistical interaction has been evaluated in some of these studies, questions remain on exactly how, to what extent, and to whom is this combined effect most beneficial to target interventions in public health.

The existing studies have almost never quantified the synergism between diet and PA. Little has been studied on the *a priori* analysis of the interaction between diet and PA to determine its impact on hard clinical events or mortality thus far.[77,78] More comprehensive, methodical, and robust evidence is needed to demonstrate that diet and PA are two sides of the same coin, as well as to identify to whom this combination may offer the greatest benefit in public health.

Interaction Analysis

An interaction (effect modification) is defined as the situation in which the effect of one exposure on an outcome differs across the strata of another exposure, implying that the risk differences vary across strata of the other exposure. Thus, the presence of interaction suggests that the effect of the two exposures is different than the mere sum or multiplication of their individual effects, depending on the nature of the association between exposures and the assumed scale (additive or multiplicative) for the interaction. This interrelation of effects suggests that the reduction of either factor would also reduce the risk of the other factor in producing a given outcome.[79] Different terminology is used throughout the scientific community to refer to the concept of interaction; joint effect or combined effect, synergy, interdependence, heterogeneity of effects, non-uniformity of effects, effect modification, or subgroup analysis.[80] For the purpose of this dissertation, the term interaction will refer to the 'mechanistic or biological interaction' created when two potential causal risk factors participate in the same causal mechanism, which implies either synergism or antagonism between factors on disease risk or death.[79,81]

The current criteria within the STROBE guidelines recommend describing any methods used to examine interactions or subgroup analyses within the statistical analysis section of the study methods.[82] However, many studies fall short of this recommendation.[80] In 2009, Knol et al. evaluated the presence of interaction in 225 epidemiological studies to examine how interaction was assessed and reported. This literature search found that not all studies that addressed effect modification or interaction provided satisfactory information on interactions between exposures (primarily treatments, medical conditions, and lifestyle factors). Moreover, only 1 out of 10 studies reported adequate information for a full assessment of additive or multiplicative interaction.[83] This is important because an adequate reporting of methods allows for higher transparency, direct interpretation, comparison, and independent recalculation of results.[84]

The concept of interaction is dependent on the chosen scale, namely, whether the scale is additive (then the interaction will mean a departure from additivity) or multiplicative as in relative risks (then the interaction will mean a departure from multiplicativity). Interaction may be present on one scale but absent on another. In fact, if both of the exposures have an effect on the outcome, then the absence of interaction on the additive scale implies the presence of multiplicative interaction for relative risks and likewise, the absence of multiplicative interaction for relative risks implies the presence of additive interaction.

The departure from additive effects is estimated by means of the Relative Excess Risk due to Interaction (RERI) that equals:

$$RERI = RR_{11} - RR_{10} - RR_{01} + 1$$

Where RR means relative risk and the sub-indexes 0 or 1 represent the exposure to both factors (RR_{11}) or to only one of them (RR_{10} or RR_{01}).

The RERI is also sometimes referred to as the “Interaction Contrast Ratio” (ICR) that provides an estimate similar to additive interaction (difference in absolute risk differences) but using risk ratios rather than absolute risks.

The Synergy Index (SI) can also be used to assess departure from additive effects.

$$SI = (RR_{11} - 1) / [(RR_{10} - 1) + (RR_{01} - 1)]$$

The Attributable Proportion (AP) due to additive interaction is a third estimate and it assesses the proportion of the risk in the doubly exposed group that is due to the interaction:

$$AP = (RR_{11} - RR_{10} - RR_{01} + 1) / RR_{11}$$

There are a variety of statistical approaches for considering interactions between potential causal factors. The most frequently reported method includes conducting a likelihood ratio test to compare regression models with and without the multiplicative interaction product-term (used for relative risks (RR), odds ratios (OR) or hazard ratios (HR)). However, this most common analysis of interaction on the multiplicative scale is limited to assessing statistical interaction. Reporting interactions on the *additive* scale is uncommon in standard epidemiologic reports. Current explanations as to why interactions may not be reported in greater detail include space constraints, word limits, or editorial intervention.[83] For instance, one study included interaction analysis on both scales, employing a cross-product term on the multiplicative scale, and AP, RERI, and SI on the additive scale, however the authors used brief descriptive statements to report no interactions were found and the data were not shown.[73] Furthermore, the current tendency among observational studies to simply report statistical significance of the likelihood ratio test on the multiplicative scale is due to the implicit nature of epidemiologic statistical modeling and software convenience.[81,85] When obtaining relative risks, the inclusion of a product term in multivariable regressions provides a quick analysis for investigators to report interactions with a corresponding p-value, usually implying that a p-value <0.05 for a product-term ($exposure_A$*$exposure_B$) implies a departure from pure multiplication of effects. This method, however, disregards the possibility of detecting additive interactions and quantifying the effect attributed to the interaction. Contrary to the common practices in standard articles of epidemiology, according to Rothman, the information provided on the *additive* scale, including interaction analysis, is most relevant for public health application.[85,86] Therefore, Knol et al. suggest using more extensive methods, including analyses for the single effects of each factor, joint effects for combinations of exposures, stratification, and measures of interaction on multiplicative and additive scales.[84]

Additive interaction analysis, on the absolute risk scale, estimates the number of attributable cases due to the combined effect. In the presence of interaction, these cases will either surpass or fall short of the sum of cases due to both exposures separately, suggesting that the excess of cases depends on the extent to which risk factors A (i.e., MedDiet) and B (i.e., PA) occur together in the same individuals. Moreover, relevant to public health, this analysis provides insights towards which subgroup of a population, not necessarily the high-risk subgroup, would observe a greater absolute risk reduction from disease prevention or intervention strategies.[79,80,87] When two independent risk factors are considered well suited to fit an additive model, the presence of biological interaction requires a departure from additivity in the scale of absolute incidence rate differences.[79,81] However, study results in epidemiology are most frequently presented on the relative risk multiplicative scale, which does not directly allow calculating an absolute risk difference. Nevertheless, alternative measures of interaction to the absolute additive model have been available for decades, including RERI, SI, and AP due to interaction.[84,85,88] The null value for RERI and AP is 0 and SI is 1.[89] Derived from the regressions on the multiplicative scale, these measures of interaction on the additive scale indicate the direction, because it can be positive (synergism, beyond the sum of effects) or negative (antagonism, below the sum of effects), as well as the magnitude of the interaction.[90]

Interactions on combined lifestyle factors are rarely a primary objective nor an initially intended analysis in most studies.[73,82,91] However, the inclusion of these analyses provides essential information on the potential public health impact and causal structure of combined effects of different relevant exposures.[83] Consequently, studying the conjoint effect of diet and PA and the subsequent impact associated with their interaction is especially relevant given the current research gap between the effects of individual factors and the complexity of an overall lifestyle.

Combined Lifestyle Factors

Research in epidemiology has assessed the association between lifestyle factors and health outcomes by creating *a priori* defined lifestyle scores. Just as the MedDiet and PA have been studied as the combined effect created by their individual components, such as *a priori* defined dietary patterns rather than single food groups or foods, lifestyle can be assessed by studying specific combinations of behaviors.[35,41,58,92] The definitions of these scores vary substantially across studies, however, PA, smoking status, alcohol intake and BMI are the most frequently included lifestyle factors.[93] While dietary indices may find important associations with health, overall lifestyle indices may better encompass the complexity of a population's multifactorial lifestyle in relation to health. For instance, the joint effect of lifestyle factors has suggested that the larger the number of healthy or low-risk lifestyle factors, the lower the risk of mortality.[94–99] Furthermore, evidence for individual and joint effects of lifestyle habits (i.e. diet, PA, smoking status, socializing, time spent working, etc.) has already indicated a greater decreased risk for depression and depressive symptoms when exhibiting a greater number of protective factors compared with individual factors.[100–103] Moreover, recent evidence has suggested adherence to healthy modifiable lifestyle factors may lower the risk of depression regardless of underlying genetic risk.[104]

This methodology was anticipated by Rothman, who stated "as more causal factors are associated with health outcomes, greater interest will be given to the joint effects created by combinations of exposures".[105] Given the clear existing evidence for the individual effects of lifestyle factors on chronic disease and mortality, studying numerous lifestyle factors simultaneously has been the next step forward.[93,106] Lifestyle scores in particular have been designed and employed to capture a multidimensional exposure with the use of algorithms, such as the *Healthy Heart Score*, or scoring systems/indices, such as *Life's Simple 7* (LS7) and the World Cancer Research Fund/American Institute for Cancer Research (WCRF/AICR) score.[29,107] The *Healthy Heart Score* is an algorithm or prediction model based on modifiable lifestyle factors that predicts the 20-year CVD risk (%) of men and women ≥ 40 years of age.[107] This score was composed of the nine factors that best estimated CVD risk, i.e. current smoking, higher BMI, low PA, lack of moderate alcohol consumption, low intakes of fruits, vegetables, cereal fiber, and nuts, and high intakes of sugar-sweetened beverages and red and processed meats.[108] On the other hand, the LS7 score is calculated by assigning 2 points for ideal, 1 point for intermediate, and 0 points for poor status of each of the seven individual factors previously mentioned, for a final score theoretically ranging from 0 to a maximum of 14 points, with a higher score indicating healthier status.[29] Both of these tools aim to facilitate the assessment of a limited number of critical lifestyle factors to identify individuals at high risk for CVD.

More and more, lifestyle scores, including simple scores, encompass a healthy dietary pattern complemented by PA, other lifestyle habits, and cardiometabolic parameters to define a larger concept of lifestyle.[109–113] Nevertheless, a recent meta-analysis observed the risk reductions for all-cause and CVD mortality similar or even greater for the simple score, compared to the LS7 score, indicating that more emphasis should be given to lifestyle factors, in addition to cardiometabolic markers, for the prevention of premature deaths.[112] Studying combinations of behavioral risk factors or lifestyle factors, such as smoking, poor diet, and physical inactivity, which tend to cluster within populations and may have synergistic effects on health, is relevant for understanding the global effect of a lifestyle score and the impact these multifaceted and interrelated habits have on individual and population health.[78] Such studies may provide key insight for implementing successful multicomponent lifestyle interventions.[114–118]

The Mediterranean Lifestyle

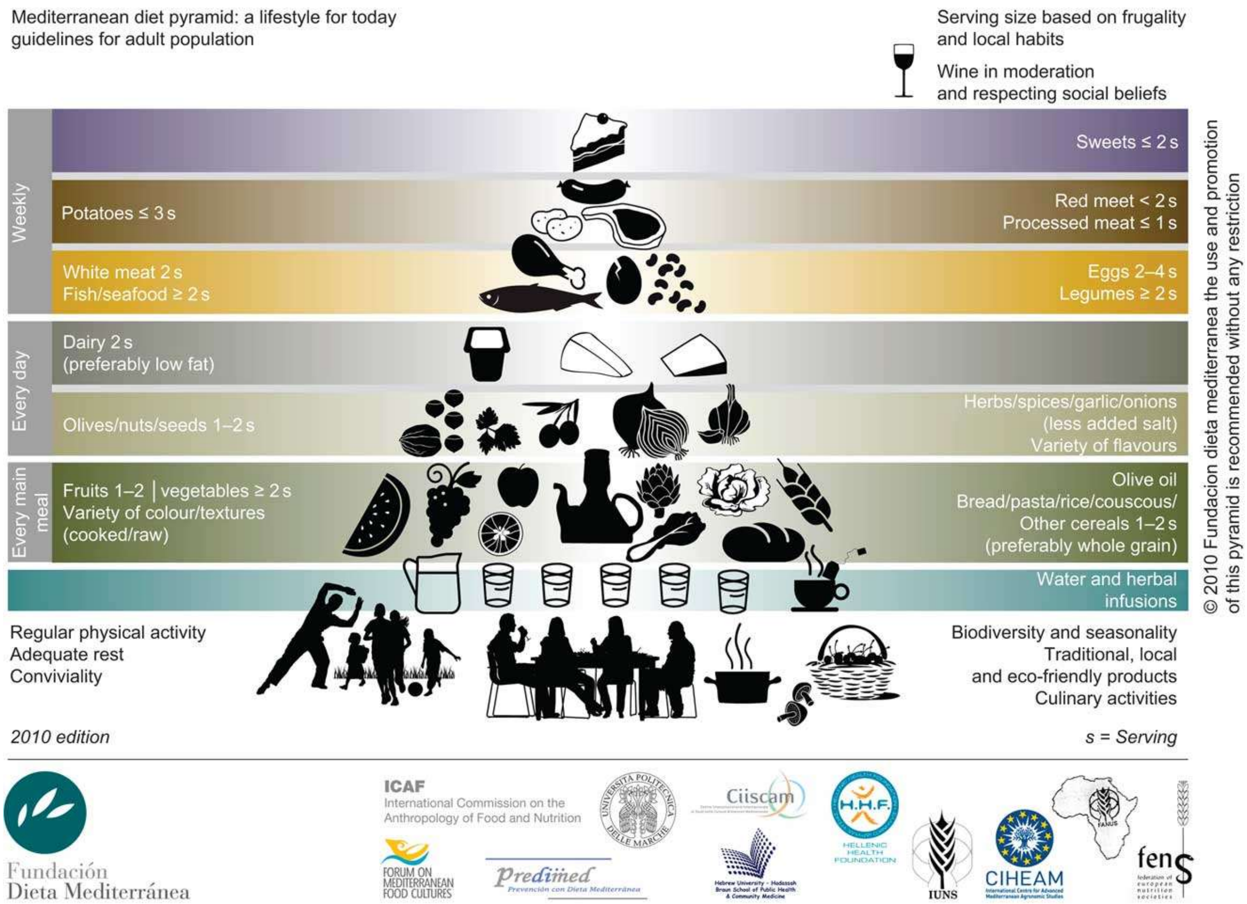
The Mediterranean lifestyle refers to the way of life unique to the Mediterranean basin. Although the MedDiet has become the most frequently assessed modifiable lifestyle factor associated with this population, the Mediterranean lifestyle encompasses several distinctive habits beyond the MedDiet, such as the way of selecting, cooking and eating food, PA, resting patterns, social structures and interactions.[72,119,120] Eating together is the foundation of the MedDiet, which reaffirms the family, group or community identity and emphasizes values of hospitality, neighborliness, intercultural dialogue, creativity and diversity.[120] Moreover, given its recognition as an intangible cultural heritage of humanity by the UNESCO Intergovernmental Committee and as a proposed universal model of a healthy diet by the EAT-

Lancet Commission, the broader concept of the MedDiet not only considers the combination of individual food groups, but also non-nutritional dietary habits including social, cultural, economic, and environmental aspects.[72,120–122] These key dietary, cultural, and social characteristics specific to this region, comprised of numerous cultures, have been described and represented by the Spanish MedDiet Foundation, in collaboration with several international experts in nutrition, anthropology, sociology, and agriculture, in a MedDiet Pyramid (Figure 6).[72]

This new model incorporates both quantitative and qualitative elements to better define the way of life that accompanies the habitual pattern of Mediterranean food consumption. Such elements that influence the frequency and consumption of foods include frugality, moderation, locally grown, biodiverse, seasonal, and traditional products, culinary practices, conviviality during meals, regular PA, adequate hydration and rest.[72] This pyramid specifies plant-based foods at the base, which should sustain the diet, while food from animal origin, rich in sugars or fats should be eaten in moderation and left for special occasions. More specifically, three main balanced meals should be comprised of one or two servings of cereals (preferably whole grain), more than two servings of vegetables (at least one raw), and one or two servings of fruit (most frequently served as dessert). About two servings of fish, white meat, and eggs are recommended per week, whereas red and processed meats should be consumed less than two and one serving per week, respectively. Other recommendations include preference for low-fat dairy products, olive oil as the primary source of fat, and wine and other fermented beverages at meals are limited to one glass per day for women and two glasses for men. Additionally, the abundant use of spices, herbs, garlic, and onion increase the palatability of traditional Mediterranean dishes and reduce the need for excessive salt in cooking. Meanwhile, olives, nuts, and seeds serve as healthy snacks. Based on these recommendations, in 2014 a Mediterranean lifestyle (MEDLIFE) index was developed and validated in a Spanish working population.[123,124]

Addressed to a healthy adult population, the three blocks of the MEDLIFE index consist of food consumption, dietary habits, PA, rest, social habits, and conviviality to holistically define the Mediterranean lifestyle as a way of living.[123] In Block 1, the MEDLIFE index identifies the shared components between the numerous varieties of MedDiets across cultures.[71,72,119,125] Block 2 encompasses drinking patterns, salt, ultraprocessed or refined foods, added sugar intake, and snacking. Lastly, PA, adequate rest, and social interactions in Block 3 also contribute to the overall effect of the Mediterranean lifestyle.[123]

Figure 6. The Mediterranean Diet Pyramid: a lifestyle for today.



In addition to being a healthy diet that does not increase adiposity or obesity, the MedDiet can also be considered part of a sustainable lifestyle and food system model adaptable to specific agricultural resources and cultures.[51] All culturally adapted varieties of the MedDiet are distinguished by the pleasure derived from its palatability and the social context that facilitates interaction, conversation, and relaxation through shared meals at fixed times with extended duration.[72,126] In addition to optimal nutrition, the Mediterranean lifestyle incorporates being physically active, including active transport, leisure time, and lifestyle tasks, an afternoon nap ≤30 minutes a day that complements a good night's rest (6-8 hours), conviviality, which refers to the warmth, friendliness, and sense of belonging associated with family life and interpersonal encounters throughout the day, such as eating meals with others, socializing with friends, and interacting with others in physical activities.[119]

The purpose behind the combination of several lifestyle factors in the MEDLIFE index is to better capture the heterogeneity and multifactorial etiology of chronic diseases and mortality.[123] Just as the concept of dietary pattern analysis has been a preferable methodology in nutritional epidemiology for better capturing the intercorrelation and interaction between nutrients and foods,[35] the MEDLIFE index provides a comprehensive assessment of the multifactorial habits that form this overall traditional way of life. As aforementioned, other lifestyle indices, such as LS7, combine behavioral and metabolic CVD risk factors for the primary prevention of cardiovascular events. However, the metabolic risk factors associated with NCDs and lower life expectancy are largely preventable by means of a healthy lifestyle.[29] Hence, the Mediterranean lifestyle employs diet and lifestyle factors for its applicability towards the primordial prevention of chronic diseases and mortality.[127,128]

Before the publication of the present articles in this dissertation, the MEDLIFE index had only been associated with lower CVD risk factors in Spanish and Croatian working populations.[124,129] Given the fact that the MedDiet has proven effective in both Mediterranean and non-Mediterranean populations[49,51,130,131] and it is currently promoted within the Dietary Guidelines for Americans,[132] other Mediterranean behaviors in combination with diet should be feasible and are likely to provide better health outcomes for non-Mediterranean populations as well.[49,133] Lastly, the MEDLIFE index implies a more comprehensive, multi-dimensional assessment of lifestyle, therefore it should continue to be used in epidemiology to provide robust evidence on the potential associations of the Mediterranean lifestyle with chronic disease and mortality.

HYPOTHESIS AND AIMS

Hypothesis – The main hypothesis of this dissertation was that a combination of traditional Mediterranean lifestyle factors would be associated with a decreased risk of chronic disease and overall mortality greater than the sum of individual lifestyle behaviors.

- The interrelationship between the MedDiet and PA and their potential interaction will drive a greater risk reduction on all-cause mortality than the sum of the individual effects of diet or PA, considered as isolated factors.
- The combined effect of dietary intakes of bioactive components, such as anthocyanins, with PA will be associated with favorable CVD risk parameters in a high-risk population.
- The Mediterranean lifestyle will provide a greater risk reduction on all-cause and cause-specific mortality than any individual lifestyle habit alone.
- Adherence to the traditional Mediterranean lifestyle will be associated with a decreased risk of depression.
- The joint effect of multicomponent lifestyle patterns, including interrelated factors such as PA and avoidance of sedentary behaviors within a general way of living, will be associated with a lower prevalence of metabolic syndrome.

General aim – The main overarching aim of this doctoral dissertation was to provide new insights into the role that combinations of lifestyle factors play on the risk of chronic disease and mortality in both Mediterranean and non-Mediterranean populations.

Specific aims –

1. To conduct a comprehensive review on reported interaction analyses between the MedDiet and PA on mortality and then comprehensively assess the potential interaction between these frequently combined lifestyle factors in the "Seguimiento Universidad de Navarra" (SUN) prospective cohort.
2. To cross-sectionally study the joint effect and possible synergism between anthocyanin intake and PA on the lipid profile of Midwestern US career firefighters in "Feeding America's Bravest".
3. To evaluate the association between the MEDLIFE index and the risk of all-cause and cause-specific mortality in a Spanish population of university graduates in the SUN cohort.
4. To longitudinally evaluate the association between the MEDLIFE index with the risk of depression among a primarily middle-aged educated population in the SUN cohort.
5. To cross-sectionally evaluate the association between the MEDLIFE index and metabolic syndrome in "Feeding America's Bravest", a non-Mediterranean working population at high CVD risk.

METHODS

The five published articles included in this dissertation were studies conducted within a Spanish cohort: *The SUN "Seguimiento Universidad de Navarra" Project* (The University of Navarra Follow-up Project) (publications 1, 3, and 4) and a US clinical trial: "*Feeding America's Bravest": Mediterranean Diet-Based Interventions to Change Firefighters' Eating Habits* (publications 2 and 5). The presented methods will describe the overall study design and aims of each study followed by the specific methods employed for each study.

1. The "Seguimiento Universidad de Navarra" (SUN) cohort

1. Study design and aims of this cohort

The SUN "Seguimiento Universidad de Navarra" Project is a prospective, dynamic, permanently open, multipurpose cohort first initiated by Dr. Martínez-González in 1999 at the Universidad de Navarra's Department of Preventive Medicine and Public Health. The SUN Project was designed in collaboration with investigators at the Harvard T. H. Chan School of Public Health, including Walter Willett, Alberto Ascherio, Frank B. Hu, and Meir J. Stampfer, using a methodology similar to that of the "Nurses' Health Studies" and the "Health Professionals Follow-up Study".

Conducted among university graduates throughout Spain, this study's main objective is to provide scientific evidence on the benefits of diet, particularly the MedDiet pattern, and lifestyle on health outcomes and disease prevention with a particular focus on NCDs such as CVD and cancer, among others.[134] Major findings from the SUN project on adherence to the MedDiet include reduced incidence of all-cause mortality, fatal and non-fatal major CVD events, T2D, weight gain, metabolic syndrome, depression, cognitive decline, and nephrolithiasis. The SUN Project has contributed to the acceptance and knowledge on the MedDiet as the ideal food pattern for the prevention of major chronic diseases highly prominent around the world today (www.publicaciones.proyectosun.es).[37]

2. Recruitment

The most recent SUN project dataset is currently comprised of 22,893 participants recruited up until December 2019. The recruitment of participants is promoted by sending invitations (Appendix 1A) to the University of Navarra alumni, other university alumni groups and professional associations.

Potential participants may also formally request to join the study via the weblink: https://participantes.proyectosun.es/registrarse or by sending an email to sun@unav.es. Participants are neither incentivized nor compensated for their participation in the SUN project. An important aspect of this study is the self-selected participation and commitment to the study by highly motived participants, which ensures a high retention rate (91%). This is attributed to the limited response rate to the initial invitation to participate without any incentive or monetary compensation. At the same time, this recruitment strategy of highly educated participants (lowest education level is a bachelor's degree) serves to control potential confounding due to heterogeneity of socioeconomic status and educational level. This restriction is the only inclusion criteria and ensures high-quality self-reported data from participants by providing greater reliability, validity, and retention rates associated with education status.[135]

3. Data collection

The study consists of a baseline questionnaire and follow-up questionnaires administered every two years that collect information on participants´ sociodemographic, diet, lifestyle, and health characteristics, including new-onset diagnoses of diseases. All invited individuals initially receive an invitation letter by mail that briefly describes the study objectives, expected involvement in the study, and information that will be collected throughout follow-up. This letter is accompanied by a baseline questionnaire, personal information form, and a pre-stamped envelope for the participant to complete and return of their own accord (Appendix 1B, 1C). As of 2004, this questionnaire may be answered via the SUN website (https://participantes.proyectosun.es/) after receiving a personal code through email. To avoid future attrition, the baseline questionnaire collects three addresses (home, work, and relative or friend), a telephone number, and email address from all participants. Newly received baseline questionnaires follow a standard procedure to add participants' personal information to the administrative database with restricted access. All questionnaires are manually reviewed for proper completion, consistency, and quality of responses, as well as the codification of some variables. Thereafter, the questionnaires are read by an optical reader through a computer scanner, which uploads the collected data digitally to the SUN's secured web-based database. To maintain participants' confidentiality at all times, this database only identifies participants with a numerical ID code that corresponds with the participant's name in the administrative database.

The baseline questionnaire contains a total of 554 items on sociodemographics (sex, birth date, marital status), anthropometrics (weight, weight change, birth weight, height, waist circumference), quality of life (health self-perception, body image, health quality), diet, eating behaviors, lifestyle (smoking, socializing, sleep, sedentary behaviors), clinical data and family/childhood medical history, disease diagnosis, preventive screenings, preventive strategies (sunscreen, sea belt, airbag or helmet), medications and supplements, personality traits, feelings and emotions, and PA (Appendix 1C).

Follow-up questionnaires are administered by mail or email every two years to all participants. These questionnaires collect information to track new disease incidence and changes in sociodemographic, medical and lifestyle variables. In order to achieve the highest retention rate possible, up to a total of five reminder emails (the last one via postal service) are sent to the participants. There are currently 12 questionnaires including the baseline, follow-up two through twenty years, and a short, abbreviated questionnaire. This latter questionnaire is sent to participants who have not responded to the 5 consecutive messages to complete and return a primary follow-up questionnaire. In between follow-up questionnaires, participants receive a newsletter with an update of the scientific progress made by the SUN project (www.proyectosun.es), as well as a Christmas card to thank participants for their dedication, commitment, and continued participation in the study. This card additionally serves to verify any changes in mailing addresses.

4. Ethical standards

The SUN project has been approved by the Research Ethics Committee of the University of Navarra and registered at clinicaltrials.gov (NCT02669602). This investigation is conducted according to the Declaration of Helsinki guidelines and participants' informed consent is given upon completion of the baseline questionnaire, which marks the date the participant entered the cohort. All potential participants are informed of their right to refuse to participate in the SUN study or to withdraw their consent to participate at any time without reprisal. All necessary study information is clearly stated, and any additional information facilitated to the potential participants.

5. Funding sources

The SUN Project has received funding from the Spanish Government-Instituto de Salud Carlos III, and the European Regional Development Fund (FEDER) (RD 06/0045, CIBER-OBN, Grants PI10/02658, PI10/02293, PI13/00615, PI14/01668, PI14/01798, PI14/01764, PI17/01795, PI20/00564 and G03/140), the Navarra Regional Government (27/2011, 45/2011, 122/2014), PNSD 2020/2021, and the University of Navarra.

6. Exposure assessments

Dietary assessment

Dietary intake was collected at baseline with a self-administered semiquantitative 136-item food frequency questionnaire (FFQ) that assessed food consumption in the previous year. This questionnaire is self-administered at baseline and after 10 years of follow-up. The FFQ has been repeatedly validated in Spanish participants[136–138] demonstrating that FFQ measurements have good reproducibility and a relative validity similar to those of FFQs used in other prospective studies. Each food item indicates a typical portion size. Total intake was calculated by multiplying the portion size with the frequency of consumption (never/seldom, 1–3 servings/month, 1 serving/week, 2–4 servings/week, 5–6 servings/week, 1 serving/day, 2–3 servings/day, 4–5 servings/day, and ≥ 6 serving/day). Nutrient intakes were estimated using the Spanish Food Composition Tables by a trained team of dietitians.[139,140] Additionally, some variables were calculated as the percentage of total daily energy intake (%E). Dietary intakes reported in the present studies included total daily energy intake (kcal/d), carbohydrate intake (%E), protein intake (%E), fat intake (%E), saturated fat (g/d), polyunsaturated fat (g/d), monounsaturated fat (g/d), monounsaturated:saturated fat (%E), fiber intake (g/d), vegetables (g/d), fruits (g/d), legumes (g/d), cereals (g/d), meat (g/d), fish (g/d), dairy products (g/d), nuts (g/d), olive oil (g/d), and alcohol consumption (g/d) among others.

Adherence to the Mediterranean diet score (MDS) was assessed with a 9-item *a priori* defined index, originally designed and developed by Trichopoulou et al., and reproduced in the SUN cohort (Table 1).[141] This operational definition of the MedDiet calculated adherence scores for all participants by assigning one point for each criteria based on the median intake of the study population. Final MDS scores could theoretically range from 0 to 9 points.

Table 1: Trichopoulou's Mediterranean diet score employed in the SUN cohort

MDS items 1-point criteria
1. Vegetables ≥ median
2. Legumes ≥ median
3. Fruits and nuts ≥ median
4. Dairy products < median
5. Cereals ≥ median
6. Meat and meat products < median
7. Fish ≥ median
8. Monounsaturated:saturated fat ≥ median
9. Alcohol 10-50 g/d men 5-25 g/d women

Physical Activity assessment

The frequency and time dedicated to habitual leisure time physical activities, sports, and sedentary behavior was collected with a validated 17-item PA questionnaire.[142] Frequency of leisure time PA was calculated as the sum of all activity durations. Metabolic equivalents (METs)-h/week for each participant were estimated as the sum of the hours per week dedicated to each activity multiplied by the activity's corresponding metabolic equivalents. A MET is defined as the ratio of the rate of energy expended during a given activity to the resting metabolic rate.

Previously designed and developed in the SUN cohort by Alvarez-Alvarez et al.,[74] an 8-item *a priori* defined index measured the degree to which participants exhibited a physically active lifestyle (Table 2). Final PA scores were calculated as the sum of points assigned to each item according to the criteria for one point, creating a theoretical scoring range from 0 to 8 points. The index was reproduced using the following variables: doing exercise (%), intensity (METs/h), energy expenditure (METs-h/wk), walking pace: brisk or very fast (%), walking (min/d), climbing stairs (floors/d), watching television (h/d), and sitting down (h/d).

Table 2. Physical activity score employed in the SUN cohort

PA score items criteria in the SUN cohort	points
1. Do you exercise?	
No	0
Yes	1
2. Exercise intensity (0-10 scale)	
Light (<6)	0
Vigorous (≥ 6)	1
3. Leisure-time energy expenditure	
≤ 16 METs-h/wk	0
≥ 16.1 METs-h/wk	1
4. Walking pace	
Slow or normal/average	0
Brisk or very fast	1
5. Walking time	
< 0.5 h/d	0
≥ 0.5 h/d	1
6. Climbing upstairs	
< 3 floors/d	0
≥ 3 floors/d	1
7. Television viewing time	
≥ 1.5 h/d	0
< 1.5 h/d	1
8. Sitting time (between working and leisure time)	
≥ 5 h/d	0
< 5 h/d	1

The MEDLIFE index

Based on the MedDiet pyramid developed by the Spanish MedDiet Foundation and international nutrition experts, in 2014, a MEDLIFE index was developed and validated in a Spanish working population.[123,124] Previous to the present publications, the MEDLIFE index had only been associated with lower levels of cardiovascular risk factors in working populations.[124,129] More recently, this index has been used in epidemiological studies, including two pertaining to this dissertation, to study the traditional Mediterranean lifestyle.[110,143–145] The MEDLIFE index was reproduced for the SUN cohort employing the baseline questionnaire.

The MEDLIFE index consists of 28 items divided into three blocks describing 1) food consumption; 2) traditional dietary habits (frugality; moderation; locally grown, biodiverse, seasonal, and traditional products; culinary practices; conviviality during meals); and 3) PA, rest and social interactions (Table 3).[123] Adapting the original MEDLIFE index for the SUN cohort required the modification of 13 items to best fit the baseline questionnaire and existing evidence within this cohort (Appendix 1D). Each item is weighted equally with 0 or 1 point, creating a categorical scoring range from 0 (worst) to 28 (best). Final scores were then categorized into quartiles of adherence to the Mediterranean lifestyle.

The validation study for the MEDLIFE index was conducted in a working population at an academic institution in Madrid, Spain. Its validity was assessed by comparing this index with a full 147-item lifestyle questionnaire, including a validated FFQ, PA, and other traditional Mediterranean habit questions. The results showed nearly 60% (16 items out of 28 items) had an absolute agreement from very good to moderate (kappa = 0.41-1). Only three items had a poor agreement (kappa < 0.2), namely dairy products, cereals, and processed meats. These values indicated a correct classification of adherence for more than half of the participants evaluated. Furthermore, the correlation between the final composite score with the lifestyle questionnaire showed a moderate-to-good correlation ($r = 0.626$, $p < 0.05$).[124]

Table 3. Description of the Mediterranean Lifestyle (MEDLIFE) index modified for the SUN cohort.

Score items	Components (serving size)	Criteria for 1 point
Block 1: Mediterranean food consumption		
1. Sweets	Cookies, chocolate cookies, pastries, donuts, homemade baked goods, store-bought baked goods (50g), muffins (25-50g), tea biscuits (90g), chocolates (30g), churros (100g), turrón (35g)	≤ 2 servings/wk
2. Red meat	Beef, pork, lamb (100-150g)	< 2 servings/wk
3. Processed meat	Sausage, soft spicy sausage, bacon (50g), cured ham (60g), cooked ham (30g), hamburger (150g), liver (100-150g), organ meats (100-150g), pâté (25g)	≤ 1 serving/wk
4. Eggs	Eggs (1 unit)	2-4 units/wk
5. Legumes	Lentils, beans, chickpeas, peas (60g uncooked)	≥ 2 servings/wk
6. White meat	Chicken/turkey with skin, chicken/turkey without skin, rabbit (100-150g)	2 servings/wk
7. Fish/seafood	White fish, fatty fish, codfish, salted or smoked fish, shrimp, octopus, calamari (100-150g), oysters and shellfish (6 units)	≥ 2 servings/wk
8. Potatoes	Baked or boiled potatoes (150g)	≤ 3 servings/wk
9. Low-fat dairy products	Skim milk (200cc), low-fat milk (200cc), low fat yogurt (125g), fresh soft cheese (50g)	2 servings/d
10. Nuts and olives	Almonds, peanuts, hazelnuts, walnuts (50g), olives (10 units)	1-2 servings/d
11. Sofrito	Olive oil, pepper, other vegetables (250g), tomato (150g)	>2/4 ingredients above the median
12. Fruit	Orange, banana, apple, pear, kiwi, mango, avocado, peach, apricot, nectarine (1 unit), clementine (2 units), strawberry (6 units), cherries, plums, figs, grapes (1 dessert plate), watermelon, melon (200-250g), dates and dried fruits (150g)	3-6 servings/d
13. Vegetables	Spinach, cauliflower, broccoli, lettuce, carrot, squash, green beans, eggplant, zucchini, cucumber, pepper, asparagus, gazpacho, garden salad, other vegetables (250g), tomato (150g) *(excludes potatoes)*	≥ 2 servings/d
14. Olive oil	Olive oil (1Tbsp)	≥ 3 tablespoons/d
15. Cereals	White bread, whole-grain bread (3 slices), white rice, pasta (60g uncooked), pizza (200g), breakfast cereal (30g)	3-6 servings/d
Block 2: Mediterranean dietary habits		
16. Water	Tap water, bottled water (200 cc), coffee, decaffeinated coffee (50cc)	≥ 6 servings/d
17. Wine	Red/white wine (1 glass)	women:>0 to ≤ 0.5 serving/d men:>0 to ≤ 1 serving/d
18. Limit salt at meals	Do you limit salt at meals?	Yes
19. Preference for whole grains	Do you try to consume a lot of fiber? + fiber from grains	Yes, ≥ 6g/d fiber from grains
20. Snacks	Potato chips (150g)	< 1 serving/wk
21. Limit snacking in between meals	Do you tend to eat in between meals?	No
22. Limit sugar in beverages (including sugar-sweetened beverages)	Do you add sugar to some beverages? + soda + bottled juice (200cc)	No, ≤ 1/wk, ≤ 1/wk
Block 3: Physical activity, rest, social habits, and conviviality		
23. Physical activity	Walking, jogging, running, climbing stairs, bicycling, stationary cycling, swimming, dance, aerobic exercise, martial arts, gymnastics, gardening, tennis, soccer, skiing, ice skating, team sports like basketball, other physical activities or sports	> 300 min/wk
24. Nap	Napping throughout the week	≤ 30 min/d
25. Hours of sleep	Sleeping throughout the week	6-8 h/d
26. Watching television	Watching TV/videos throughout the week	≤ 2 h/d
27. Socializing with friends	Socializing throughout the week	> 1 h/d
28. Collective sports	Playing soccer, tennis, squash or other racket sports, basketball, and other team sports	≥ 1 h/wk

Abbreviations: min: minutes, h: hours, d: day, wk: week, cc: cubic centimeter, g: grams, Tbsp: tablespoons

7. Covariate assessments

Anthropometric measurements have been previously validated in a cohort subgroup.[146] BMI was calculated by dividing weight by height squared (kg/m^2). Other sociodemographic characteristics included sex (male/female), age (years), marital status (single, married, widowed/divorced/other), living alone (yes/no), and maximum attained educational level at enrollment (bachelors/postgraduate studies). Lifestyle variables included smoking status (never, current, former), cigarettes smoked (pack-years), special dieting (yes/no), alcohol intake (g/d), and hours working (none, <40 h/wk, ≥40 h/wk). Analyses with the MEDLIFE index were adjusted for alcohol intake defined as servings of beer and distilled beverages per week because wine consumption was already included in the MEDLIFE index. Personality traits characterized each participants' level of competitiveness, psychological tension, and dependence on a scale of 0 (more conformist, relaxed, or autonomous) to 10 (more competitive, tense, or dependent). Participants' family/personal medical history included family history of CVD, baseline prevalence of hypertension, hypercholesterolemia, diabetes, CVD, depression, cancer, and regular aspirin use (yes/no). Prevalent cases of disease were identified if they reported a previous diagnosis and/or treatment with antihypertensive, antidiabetic, or lipid-lowering medications, respectively.

8. Outcome assessments

All-cause and cause specific mortality

Primary outcomes in papers 1 and 3 included all-cause mortality and cause-specific mortality from CVD, cancer, and other causes. Other causes of death were comprised primarily of external injuries, respiratory, neurological, digestive, endocrine, infectious diseases, and suicides. As of 2018, only about 4% of reported deaths had unconfirmed causes. In the SUN project, both families and postal authorities frequently report deaths, which are confirmed by death certificates and medical records sent by next of kin or computerized record linkage to the Spanish National Statistics Institute (NSI, www.ine.es).[147] The date and cause of death are recorded and encoded using the International Classification of Diseases (ICD-10). The initial reports of death until 2018 had been communicated by a family member via a questionnaire, email or WhatsApp (n=250), telephone (n=23), mail (n=21), newspapers or newsletters (n=8), or friends (n=5). The remaining cases were identified with the Spanish National Death Index (S-NDI) or the S-NDI system search requested annually (n=113).

The S-NDI was initiated relatively recently in the year 2000 and serves as the standard source of data for assessing deaths in cohort studies throughout Spain. Access to the NSI is considered more accurate and allows utilizing microdata with personal identifiers to link the NSI databases with research files. This is only made possible after a rather arduous process that requires signing an agreement between the NSI and the University of Navarra. Prerequisites involve submitting the research databases in a specified format, paying a stipulated price, and lastly, collaborative work between one member of the research team and officials of the NSI is required to best decide whether a death can be assigned to cases with partial agreement of personal identifiers. The positive predictive value for these sources of information regarding fatalities nationally is very high, expected to be around 100%.

Depression

Depression was the outcome of interest for publication 4. Incident cases of depression in the SUN cohort are defined as self-reported new cases of depression throughout follow-up by participants free from any previous history of depression at baseline, not using any antidepressants at baseline, and indicated yes to the question "Have you ever been diagnosed with depression by a medical doctor" or self-reported a new habitual use of antidepressants. Either one or both a medical diagnosis and onset of antidepressant treatment was classified as incident depression. Given that antidepressant prescriptions for medical purposes other than depression are extremely uncommon in Spain, we considered these criteria valid for defining incident depression. Furthermore, self-reported cases of depression have been previously validated in a subsample of the SUN cohort using the Structured Clinical Interview for DSM-IV (conducted by a senior psychiatrist or clinical psychologist) as the gold standard. The specificity was 96% (percentage of confirmed depression was 74.2% (95% CI: 63.3–85.1) and confirmed non-depression was 81.1% (95% CI: 69.1–92.9).[148]

***Specific methods** (this section is a reiteration of the Statistical Analysis of each article)*

1.1. The Mediterranean Diet and Physical Activity: better together than apart for the prevention of premature mortality

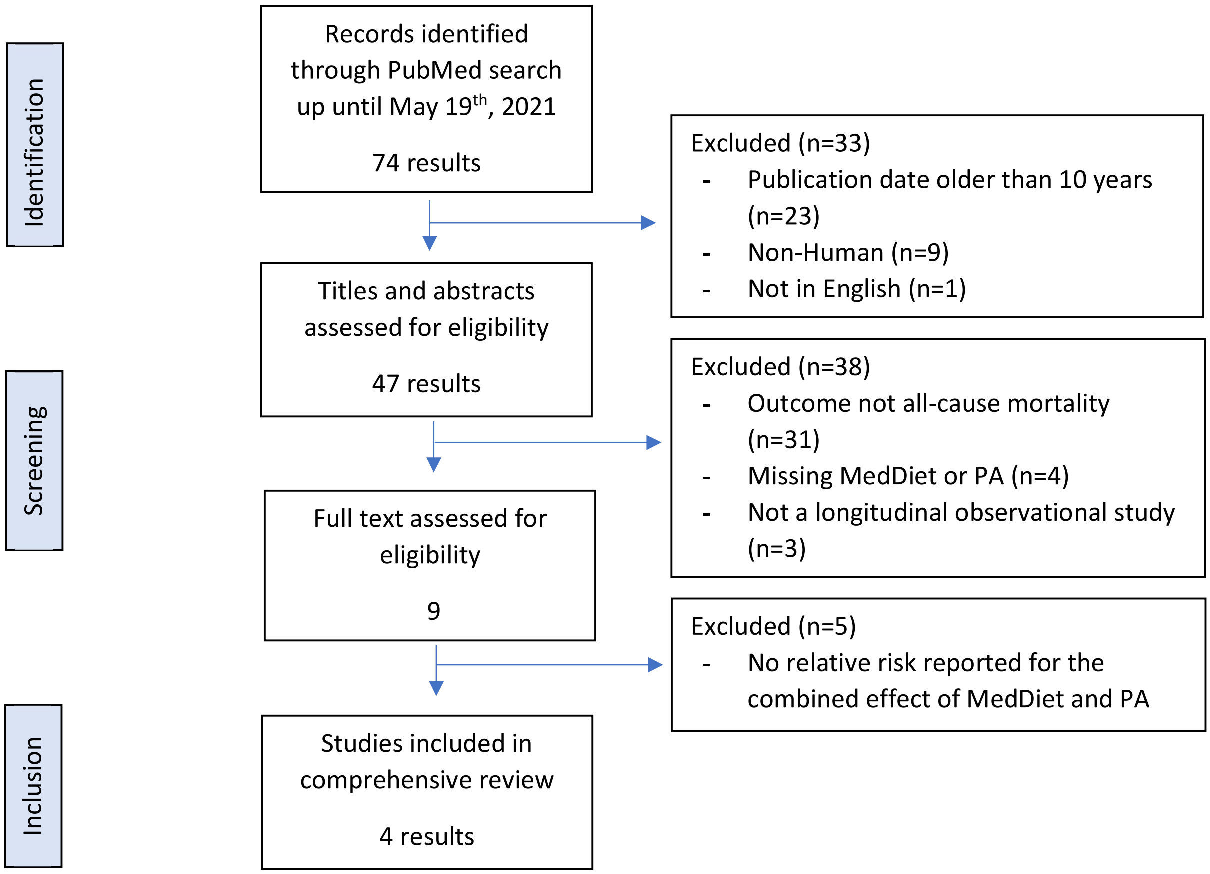
1. Inclusion and exclusion criteria of eligible studies for review

We searched PubMed database for original observational research articles (in the last 10 years, English, and humans) that studied the combined effect of the MedDiet with PA on all-cause mortality, from the time when Knol and Vanderweele first published recommendations for presenting analyses of effect modification and interaction in 2012 (Table 4).[84] We first identified the methods employed to assess the combination of diet and PA. Variations among studies included reporting relative risks for: a lifestyle score that included diet and PA items; a lifestyle score and its individual components; diet and a lifestyle score combined; diet and PA combined; or a lifestyle score, its individual components, and combinations of components. Since we were interested in accessing the presence of interaction analysis between the MedDiet and PA, we excluded all studies that did not specifically assess the relative risk for the combination of the MedDiet and PA on mortality.[96,113,149–151] After these exclusions, only 4 articles met the inclusion criteria for further assessment (Figure 1).

Table 4. Search strategy, database searched until May 19th, 2021.

Database	Search strategy
PubMed	TOPIC: ((“Mediterranean diet” OR “Mediterranean dietary pattern” OR “Mediterranean diets” OR “Diets, Mediterranean”)) AND ((“Physical Activity” OR “Physical Activities” OR “Activity, Physical”)) AND ((“survival" OR “survive" OR “mortality" OR “fatal" OR “death")) AND (("follow up" OR "longitudinal studies" OR "cohort studies" OR "prospective studies"))

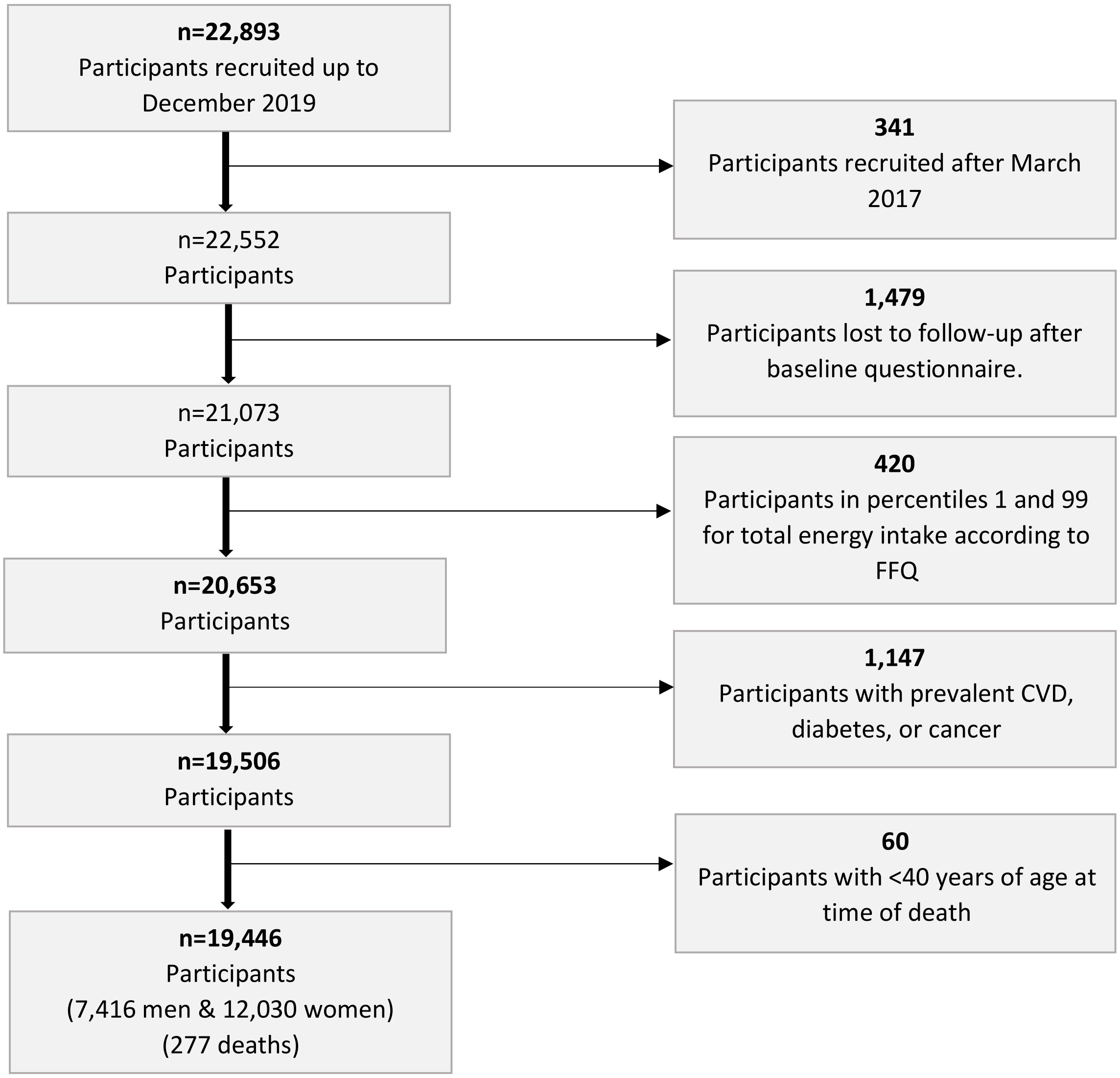
Figure 1. Flow chart of eligible studies for comprehensive review.



2. Inclusion and exclusion criteria for analysis

From December 1999 until December 2019, a total of 22,893 participants had been recruited for the SUN cohort. We excluded participants recruited within the past 2 years and 9 months (March 2017) to ensure equal opportunity for the completion of the first follow-up questionnaire by all participants without creating a misclassification bias by those highly motivated who quickly return the questionnaire [n=341], participants lost to follow-up after the baseline questionnaire [n=1,479], participants in percentiles 1 and 99 for total energy intake according to the FFQ [n=420], participants with prevalent CVD, diabetes, or cancer [n=1,147], and participants with less than 40 years of age at the time of death [n=60]. After exclusions, a total of 19,446 participants, consisting of 7,416 men and 12,030 women (61%), were included in the analysis.

Figure 2. Flowchart for selection of participants in the SUN cohort



3. Statistical analyses

Traditionally presented as protective factors, Trichopoulou's operational definition of the MedDiet, a 9-point MDS, and a 8-point PA score were transformed and presented as risk factors by selecting the lowest risk category as the reference group by recommendation of Knol et al. when conducting interaction analyses on the additive scale.[152] Thus, MDS score was presented as approximate quartiles (Q4: high adherence to Q1: low adherence) and PA score was dichotomized into categories of high (4-8 points) and low (0-3 points) activity levels. This categorization of each exposure identified the most appropriate distribution of individuals with differentiated MedDiet adherence and PA level. Table 5 shows how the combined exposures were created with a contingency table for quartiles of MDS and dichotomous PA variables.

Table 5. Combined exposures of adherence to the MedDiet and PA.

Exposures†		*MedDiet adherence*			
		Q4 MDS *(7-9 pts.)*	Q3 MDS *(5-6 pts.)*	Q2 MDS *(4 pts.)*	Q1 MDS *(0-3 pts.)*
Physical Activity	high PA *(4-8 pts.)*	Q4 MDS - high PA (reference)	Q3 MDS - high PA	Q2 MDS - high PA	Q1 MDS - high PA
	low PA *(0-3 pts.)*	Q4 MDS - low PA	Q3 MDS - low PA	Q2 MDS - low PA	Q1 MDS - low PA

†Variables are presented as risk factors.
MDS: Mediterranean diet score, PA: physical activity pts: points, Q: quartile

Multivariable statistical analyses were conducted using Cox regression models for the assessment of individual and combined effects between adherence to the MedDiet and PA on all-cause mortality. Follow-up for each participant was calculated from the date the baseline questionnaire was returned to the date the last questionnaire was received or the reported date of death. Age was the underlying time variable, and all Cox regression models were stratified by age in decades (7 categories) and the year in which participants entered the study (6 categories). Multivariable adjusted HRs with 95% confidence intervals (CIs) were adjusted for sex, BMI (5 categories), educational level (bachelor's degree/masters or doctorate), smoking status (never, active, former smoker), cigarettes smoked (packs/d-yr), alcohol consumption (continuous), total energy intake (continuous), family history of CVD, prevalent hypertension, hypercholesterolemia, and history of depression at baseline (ever/never). Individual exposures were additionally adjusted for the remaining lifestyle factors. Linear trend tests were performed by assigning medians to each category and treating it as a continuous variable.

Interactions were analyzed according to the methodology proposed by Knol and Vanderweele by studying the single and joint effects of the exposures followed by an interaction analysis on both the multiplicative and additive scales (Table 6).[84,153] Knol et al. made particular emphasis that protective factors be recoded as risk factors, selecting the reference group as those not exposed to either risk factor, representing the lowest risk on the given outcome, for the correct calculation and interpretation of the RERI.[152] On the multiplicative scale, a likelihood ratio test compared Cox regression models with and without a product term for the lowest MDS and low PA level. On the additive scale, the lowest MDS quartile and low PA category were employed for calculating the RERI, as well as the AP due to interaction.

All *p*-values were 2-sided and considered statistically significant at $p<0.05$. All statistical analyses were conducted using STATA 14 (StataCorp, College Station, TX, USA).

Table 6. Methods for interaction analyses on multiplicative and additive scales.

Multiplicative Interaction:

Likelihood ratio test – Comparison of Cox regression models using the likelihood ratio test.

STATA code:

```
generate I_A=g*e
stcox e g   c1 c2 c3
est store A
stcox I_A e g   c1 c2 c3
lrtest A .
```

g = 1; low Mediterranean diet score (Q1: 0-3 points)   e = 1; low physical activity (0-3 points)
g = 0; reference (Q4: 7-9 points)   e = 0; reference (4-8 points)
c = covariables

Additive Interaction:

Relative excess risk due to interaction (RERI) – Proportion of the *effect* of both exposures on the additive scale that is due to their interaction.

$$RERI_{HR} = HR_{11} – HR_{10} – HR_{01} + 1$$

HR_{11} is the adjusted hazard rate ratio comparing the doubly exposed higher risk combination to the reference combination with the lowest risk HR_{00}.[154] RERI = 0 means no interaction or exactly additivity; RERI > 0 means positive interaction or more than additivity; RERI < 0 means negative interaction or less than additivity; RERI can go from – infinity to + infinity. [155]

STATA code:

```
stcox I_A e g   c1 c2 c3
nlcom (exp(_b[I_A] + _b[g] + _b[e]) - exp(_b[g]) - exp(_b[e]) + 1)
```

Where:
exp (b[I_A] + _b[g] + _b[e]) = HR_{11}
exp(b[g]) = HR_{10}
exp(b[e]) = HR_{01}

Attributable proportions due to the joint effect – proportion of the joint effect (total hazard) that is due to each component among those who present both exposures.

Portion of the effect attributable to the MedDiet alone: $(HR_{10} – 1) / (HR_{11} – 1)$
Portion of the effect attributable to PA alone: $(HR_{01} – 1) / (HR_{11} – 1)$
Portion of the effect attributable to their interaction: $(RERI_{HR} – 1) / (HR_{11} – 1)$

STATA CODE:

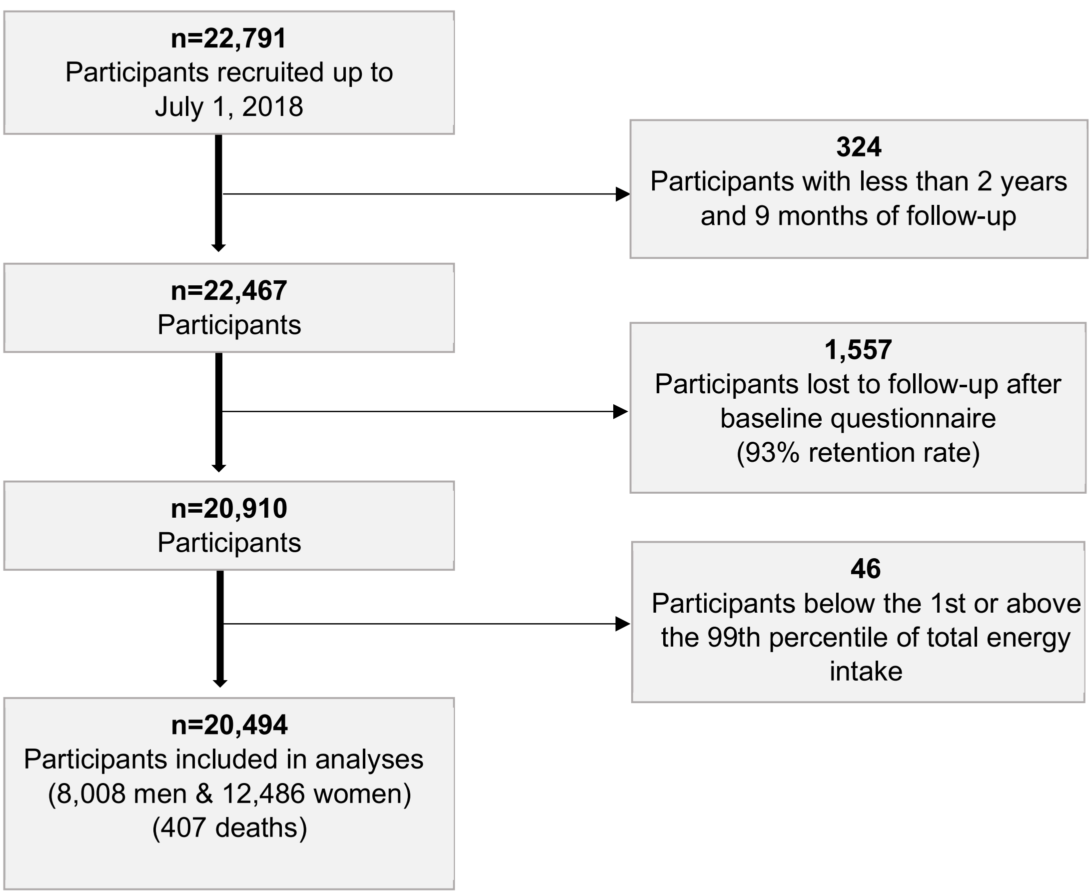
```
nlcom (exp(_b[g])-1)/(exp(_b[I_A]+_b[g]+_b[e])-1)
nlcom (exp(_b[e])-1)/(exp(_b[I_A]+_b[g]+_b[e])-1)
nlcom (exp(_b[I_A]+_b[g]+_b[e])- exp(_b[g]) - exp(_b[e]) + 1)/(exp(_b[I_A]+_b[g]+_b[e])-1)
```

1.2. The Association Between the Mediterranean Lifestyle Index and All-Cause Mortality in the "Seguimiento Universidad de Navarra" (SUN) Cohort

1. Inclusion and exclusion criteria for analysis

From December 1999 until July 2018, a total of 22,791 participants were recruited in the SUN cohort. The exclusion criteria applied for this study were: participants with <2 years and 9 months of follow-up [n=324], participants lost to follow-up after the baseline questionnaire [n=1,557] (93% retention rate), and participants with sex-specific extreme energy intakes <1st or >99th percentiles [n=416]. A total of 20,494 participants, including 8,008 men and 12,486 women, were included in this study's analyses (Figure 3).

Figure 3. Flowchart of eligible participants in the SUN cohort 1999-2018.



2. Statistical analyses

Baseline characteristics of participants adjusted for age and sex with the inverse probability weighting method were described according to MEDLIFE quartiles, expressed as means with standard deviations (SD) for numerical variables or percentages for categorical variables.

To determine the contribution of each item to the between-person variance of MEDLIFE scores, stepwise-selection regression analyses and nested least-squares linear regression models were conducted. The change in the cumulative coefficient of determination (R^2) identified each item's contribution to the total variability in the score.

Cox proportional regression models were fitted with age as the underlying time variable to assess the risk of all-cause, CVD, cancer, and other causes of death across MEDLIFE quartiles. Follow-up for each participant was calculated from the date the baseline questionnaire was returned to the date of death reported or the last questionnaire was received, whichever came first. The proportional-hazards assumption was tested using the Schoenfeld residuals method. We calculated HRs and 95% CIs across MEDLIFE quartiles for all-cause and cause-specific mortality, using the lowest adherence quartile (Q1; range 3-10 points) as the reference for all models. In addition, HRs and 95% CIs were calculated for each 2-point increment in the MEDLIFE score as a continuous variable and linear trend tests performed by assigning medians to each quartile and treating it as a continuous variable.

To control for potential confounding, the multivariable-adjusted models were stratified by age group (deciles) and year of recruitment (6 categories) and adjusted for sex (male/female), BMI (tertiles), total energy intake (kcal/day), special diets (yes/no), alcohol intake excluding wine (g/d) (tertiles), smoking status (never, former, current), cigarette pack-years (tertiles), postgraduate education (yes/no), family history of CVD (yes/no), prevalent hypercholesterolemia, hypertension, diabetes, cancer, and CVD (yes/no).

Adjusted restricted cubic splines for all participants and after stratification by age at baseline and at last contact were presented for each 1-point increment of MEDLIFE on all-cause mortality. HRs and 95% CIs for the 28 items and three blocks of MEDLIFE were assessed individually for all participants and stratified by age at last contact, adjusting for all confounding variables and the remaining items or blocks, respectively. The reference category for each item was the absence of the given MEDLIFE item (0 points). Nelson Aalen survival plots were adjusted for all potential confounders mentioned above with inverse probability weighting to show mortality rates according to MEDLIFE quartiles (Q2 and Q3 were merged to form a medium adherence category).

Within our subgroup analysis, we presented HRs and 95% CIs across MEDLIFE quartiles and assessed statistically significant multiplicative interaction terms using likelihood ratio tests for sex (male, female), smoking status (non-smokers, smokers), BMI (<25 kg/m^2, ≥25kg/ m^2), age at baseline and age at final contact (<50 years, ≥50 years). Lastly, we conducted several sensitivity analyses to test the robustness of our findings, presenting HRs with 95% CIs and linear trend tests across quartiles for each modification.

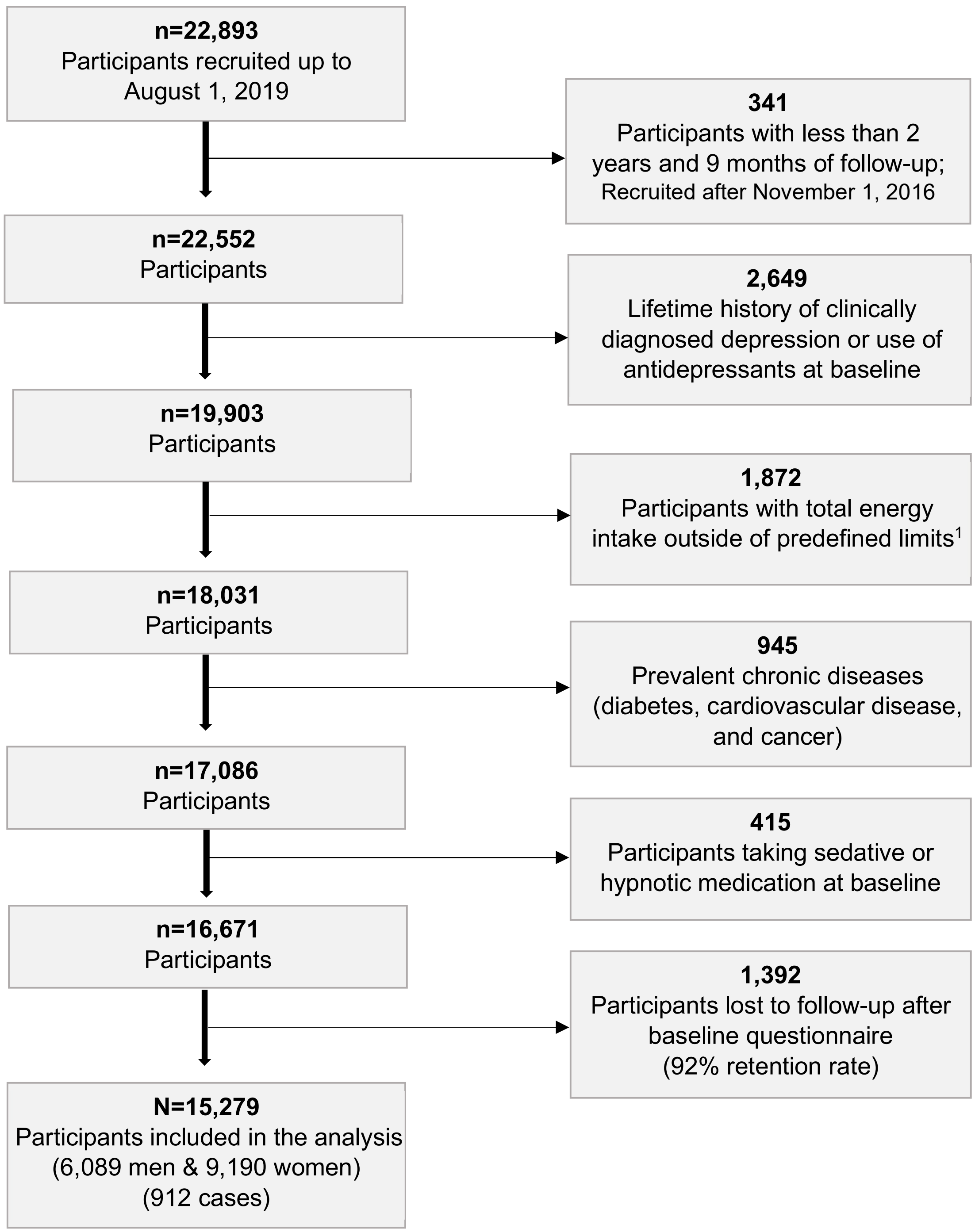
All analyses were conducted in 2019 with Stata version 12.0 (StataCorp, College Station, TX). All *p*-values are two-sided and were considered statistically significant at $p<0.05$.

1.3. The Mediterranean Lifestyle and the Risk of Depression in Middle-Aged Adults

1. Inclusion and exclusion criteria for analysis

From December 1999 until August 2019, a total of 22,893 participants were recruited. The exclusion criteria included: participants with less than 2 years and 9 months of follow-up (recruited after November 2016) (n=341), participants with a lifetime-history of clinically diagnosed depression or use of antidepressants at baseline (n=2,649), total energy intake outside of predefined limits (men: <800 kcal/day or >4000 kcal/day and women: <500 kcal/day or >3500 kcal/day) (n=1,872), prevalent chronic diseases; diabetes, CVD, and cancer (n=945), use of sedative or hypnotic medication at baseline (n=415), and participants lost to follow-up after the baseline questionnaire (n=1,392; 92% retention rate). A total of 15,279 participants, including 6,089 men and 9,190 women, were included in the primary analysis of this study (Figure 4).

Figure 4. Flowchart for selection of participants in the SUN cohort



*<800 kcal/day or >4000 kcal/day in men and <500 kcal/day or >3500 kcal/day in women[156]

2. Statistical analyses

Baseline and MEDLIFE characteristics of participants adjusted for age and sex, using the inverse probability weighting method,[157] were described according to MEDLIFE quartiles using means with SDs for numerical variables and relative frequencies for categorical variables.

To determine the contribution of each item to the between-person variance of MEDLIFE scores, stepwise selection regression analyses and nested least squares linear regression models were conducted. The change in cumulative R^2 identified each item's contribution to the total variability in the score.

Cox proportional regression models were fitted with age as the underlying time variable to assess the risk of incident depression across MEDLIFE quartiles. Person-years of follow-up for each participant were calculated from the date the baseline questionnaire was returned to the date the last questionnaire was received, the date of diagnosis of depression, or the date of death, whichever occurred first. HRs and 95% CIs were calculated across MEDLIFE quartiles using the lowest adherence quartile (Q1; range, 3–10 points) as the reference group for all models. In addition, HRs and 95% CIs were calculated for each additional point of MEDLIFE as a continuous variable and linear trend tests were performed by assigning medians to each quartile and treating it as a continuous variable.

Multivariable models were stratified by age group and year of recruitment and adjusted for sex (dichotomous), BMI (continuous), total energy intake (kcal/d; continuous), special dieting (yes, no), alcohol intake (not including wine; ≥1 serving/d, <1 serving/d), smoking status (never, former, current), cigarette pack-years (continuous), marital status (single, married, widowed-divorced-other), level of competitiveness, psychological tension, dependence (continuous), and hours working (none, <40 hr/wk, ≥40 hr/wk).

In addition, HRs and 95% CIs for the 28 items and 3 blocks were assessed individually for all participants, adjusting for all confounding variables and the remaining MEDLIFE items or blocks, respectively. The reference category for each item was the absence of the given MEDLIFE item (0 points). Adjusted restricted cubic splines for all participants were plotted to graphically assess the dose-response association of the MEDLIFE index with incident depression.

Subgroup analyses showed HRs and 95% CIs across MEDLIFE quartiles and multiplicative interaction terms were assessed using likelihood ratio tests for sex (male/female), BMI (<25 kg/m^2, ≥25 kg/m^2), age at baseline (<50 years, ≥50 years), and personality traits; psychological tension, competitiveness, and level of dependence (<, ≥ median) for the association between the MEDLIFE index and incident depression.

Sensitivity analyses showed multivariable HRs with 95% CIs and linear trend tests across quartiles (Q2 and Q3 were merged to form a medium adherence category); awarding 1 extra point to non-smokers, 1 extra point to non-drinkers, including participants taking sedative or hypnotic medication at baseline, and defining the outcome as having both a reported diagnosis and treatment of depression. Additional exclusions from the study population included participants missing ≥30 FFQ items and those diagnosed with depression within the first 2 years of follow-up. Substitutions included: Trichopoulou's MDS[141] instead of Block 1, high adherence (7 to 9 points) to the Mediterranean alcohol drinking pattern (MADP)[158] instead of wine, PA >150 minutes/week, napping on weekends, 6-8 hours of sleep during weekdays, watching <1 hour of television on weekdays, socializing with friends ≥2 hour/day on weekends, collective sports ≥2 hours/week, and the original criteria used by Sotos-Prieto[123] for Block 3.

All analyses were conducted with Stata, version 14.0. All *p*-values are 2-sided and were considered statistically significant at $p<0.05$.

2. “Feeding America’s Bravest”: Mediterranean Diet-Based Interventions to Change Firefighters' Eating Habits

1. Study design and aims of this trial

“Feeding America’s Bravest” is a cluster-randomized diet intervention trial (RCT) that included 44 fire stations from the Indianapolis Fire Department (IFD) and 6 fire stations from Fishers, Indiana Fire Department with almost 500 enrolled firefighters between 2016 and 2019. Led by Dr. Stefanos Kales and Dr. Steven Moffatt of Public Safety Medical (PSM), this research study was funded by the US Department of Homeland Security. Based on the study’s ability to foster firefighters’ well-being, it has been endorsed by the International Association of Fire Fighters and the National Fallen Firefighter Foundation. The aim of this trial was to motivate firefighters and their families to incorporate MedDiet principles at work and home through education, participation, and incentives. The ultimate purpose of the study was to lower firefighters’ CVD and cancer risk by successfully getting more firefighters and their families to adopt and implement healthy eating principles into their daily lifestyle through the promotion of greater understanding, acceptance, and adherence to this traditional dietary pattern. The rationale for this nutritional intervention was evidenced by previously conducted national surveys that demonstrated US firefighters most often chose the MedDiet as the most popular way to eat healthier. More information can be found on the study’s website, which currently serves as a resource for ongoing nutritional education among firefighters across the country (www.hsph.harvard.edu/firefighters-study/feeding-americas-bravest/).

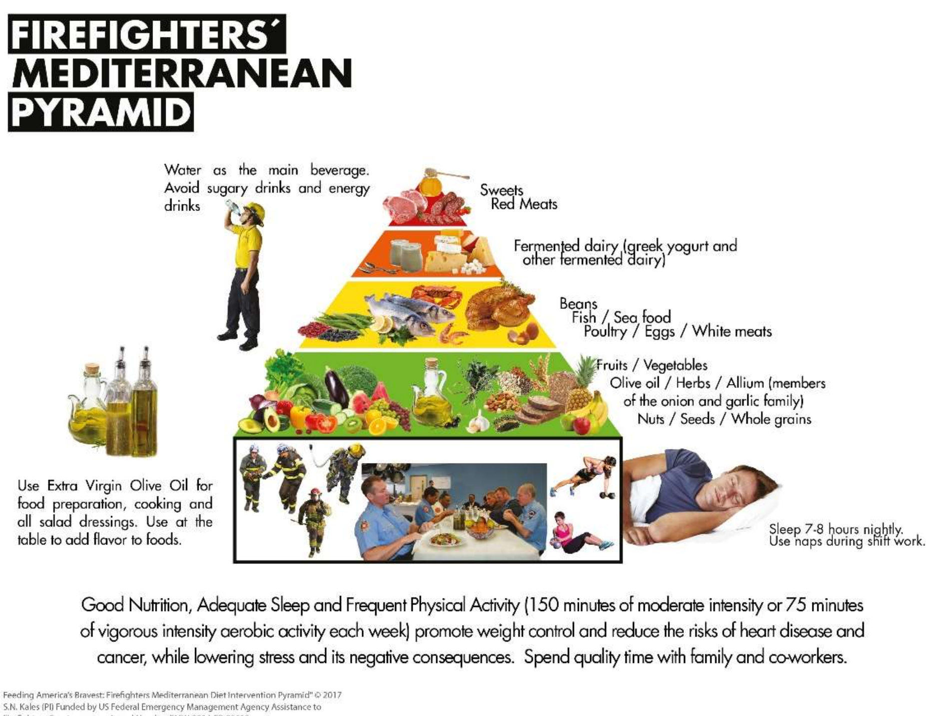
The Mediterranean Diet Nutritional Intervention (MDNI) addressed behavioral and environmental components with educational materials; opportunities for discounted access to healthy foods consistent with the MedDiet for both participating firefighters and their families; group education/support, online learning, and email/text message encouragement and reminders. Educational materials included a brochure, shopping list recommendations, sample recipes and cooking demonstrations, tips to practice both at home and at work, and a firefighter specific MedDiet Pyramid (Figure 5). While an intervention group received the MDNI for 12 months followed by a 12-month self-sustained phase, the control group received the MDNI for

6 months to test the efficacy of a shorter intervention followed by 6 months of a self-sustained phase after receiving 12 months of usual care.

The primary objective of this RCT was to compare a multiple behavior change strategies MDNI (group 1) against a Midwestern-style diet or "usual care" group (control, group 2) using a cross-over study design over a 2-year period: 12-month change in mMDS comparing group 1 vs. group 2; the 12- and 24-month change in group 1, and 6- and 12-month change in group 2, from baseline. Secondary outcomes included changes in body weight, body composition and other cardiometabolic risk markers: blood pressure, glucose, total cholesterol, HDL cholesterol, LDL cholesterol, TG, high sensitivity C-reactive protein (hs-CRP) and metabolic syndrome, as well as correlations between self-reported dietary habits and adherence biomarkers (urine tyrosol and hydroxityrosol, plasma fatty acids).

This innovative trial tested a novel worksite approach to introduce the MedDiet among US firefighters through a multicomponent MDNI combining evidence-based behavior change strategies with economic incentives, family and peer support, and environmental changes. Evidence obtained from this trial may help inform recommendations for improving the health of the US fire service and potentially other similar workforces, such as the police, military and veterans.[159–161]

Figure 5. The Mediterranean diet nutritional intervention pyramid for US career firefighters.



2. Recruitment and data collection

Eligible IFD members for the trial included those: a) permanently assigned to one of the 44 IFD or 6 Fishers fire stations; b) with a fire department medical examination in the last two years conducted at the PSM clinic; c) at least 18 years of age; and d) full duty status, modified or restricted duty. The fire department notified PSM regarding criteria for medical exams. A sample invitation letter used by the Fishers Fire Department is provided in Appendix 2A.

Baseline data collection included a nutrition/lifestyle (history of CVD, tobacco, sleep patterns, PA and diet behavior) Qualtrics questionnaire, 13-item modified Mediterranean diet score (mMDS)[162], validated 131-item semi-quantitative FFQ[163], behavioral readiness for change (e.g. receptivity, resistance, psychosocial factors), anthropometric measurements taken by the study team; blood pressure, weigh-in, waist circumference, and body composition, medical exams (blood glucose, lipid profile), and physical fitness test results. Coded de-identified results from the last fire department medical examinations were imported from the existing electronic medical record database at PSM.

Figure 6. Timeline of data collection for the elaboration of "Feeding America's Bravest" dataset.

365d<x<30d	±45d	±45d	±45d	±45d	±45d	±45d	±45d	±45d	
Visit 0 baseline	Visit 1 3 months	Visit 2 6 months	Visit 3 9 months	Visit 4 12 months	Visit 5 15 months	Visit 6 18 months	Visit 7 21 months	Visit 8 24 months	Visit 9 >24 months

*flags mark the first date of each follow-up visit. d:days.

Figure 6 shows the methodology developed to assign data to study visits for the elaboration of the "Feeding America's Bravest" database during a 3-month research stay with principle investigator Dr. Stefanos Kales at the Harvard TH Chan School of Public Health, Boston, Massachusetts, USA. Study visits collected sociodemographics, anthropometrics, lifestyle and diet questionnaires, fitness, and medical lab results, unless otherwise noted (Figure 7). The FFQ was collected at baseline and biomarkers for a study subgroup at the 12-month visit. Table 7 shows the sample size for each type of data collected throughout the intervention. The final database was comprised of 486 participants with a total of 4,042 variables.

Table 7. Data collection (n) across "Feeding America's Bravest" study visits.

enrolled IFD (n=428) *enrolled Fishers (n=58)* *Total enrolled (n=486)*		Baseline 0m	3m	6m	9m	12m	15m	18m	21m	24m
Anthropometrics	**Total**	**486**		**347**		**265**		**152**		**226**
	IFD	428		294		228		152		226
	Fishers	58		53		37				
FFQ	**IFD**	**426**								
Qualtrics	**Total**	**323**		**338**		**262**		**152**		**162**
	IFD	265		293		228		152		162
	Fishers	58		45		34				
Fitness	**Total**	**478**	**151**	**143**	**90**	**65**	**89**	**79**	**51**	**45**
	IFD	422	150	110	76	62	87	71	51	45
	Fishers	56	1	33	14	3	2	8		
Medical labs	**Total**	**481**	**204**	**97**	**41**	**63**	**120**	**64**	**46**	**69**
	IFD	428	174	73	41	60	98	49	46	69
	Fishers	58	30	24	0	3	22	15		
Biomarkers						**48**				

IFD: Indianapolis fire department, FFQ: food frequency questionnaire, m: month, n: sample size

3. Ethical standards

The overarching "Feeding America's Bravest" protocol was approved by the Harvard IRB (IRB16-0170) and is registered at Clinical Trials (NCT02941757). All participants provided full consent for their participation in the study. Each member had a chance to individually ask the PSM staff questions and decide whether to sign or decline the form. IFD members were assured that their participation status in the study was completely voluntary and had no bearing on their employment with the department or on the occupational health care they received from PSM. All procedures performed in this study involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and the 1964 declaration of Helsinki and its later amendments or comparable ethical standards.

Furthermore, the cross-over design of the study was most appropriate for ethical considerations to support buy in and enrollment within this particular population by offering a beneficial intervention to all participants. Receiving the assignment for the control group for two years would not have been accepted by those concerned with their health. Given the nature of an intervention study design, a data advisory monitory board (DAMB) was formed based on the member's nutrition, clinical, methodologic, statistical, or fire service expertise. This board was responsible for monitoring interim data analyses in collaboration with the team statistician.

In addition, the sponsors of this study had no involvement in the overall study design; data collection, analysis, and interpretation of data; writing of the report; or the decision to submit subsequent publications.

4. Funding sources

The "Feeding America's Bravest" trial was funded by the US Department of Homeland Security's US Federal Emergency Management Agency Assistance to Firefighters Grant program: Award Number EMW-2014-FP-00612. Commercial sponsors of this study included: Kroger Company (coupons and customer loyalty discounts); Barilla America (Barilla Plus Products), Arianna Trading Company, Innoliva and Molino de Zafra, Spain (extra virgin olive oil samples and discounts) and the Almond Board of California (free samples of roasted unsalted almonds).

5. Exposure assessments

Dietary Assessment

Dietary intake was collected at baseline at the study team visit using a validated 131-item semi-quantitative 2007 grid Harvard FFQ, also known as the Willett FFQ.[163] This FFQ reflects the previous year's habitual intake and has been validated by the "Health Professionals Follow-Up Study". Participants were asked how often, on average, they consumed each food of a standard portion size in the past year. The nine possible frequency responses ranged from "never or less than once per month" to "six or more times per day". Intraclass correlation coefficients for nutrient intakes ranged from 0.47 to 0.80 between two FFQs one year apart and showed a mean correlation coefficient of 0.59 with energy-adjusted nutrient intakes measured by diet records. This correlation was even stronger (mean r=0.65) after adjusting for week-to-week variation in diet record intakes.[164] A sample FFQ can be found at the following link: regepi.bwh.harvard.edu/health/nutrition.html (Appendix 2B).

For our first analysis, flavonoid subclasses were calculated as the habitual daily intake (mg/day), estimated using the US Department of Agriculture (USDA) flavonoid content of foods database, according to previously described methods.[165,166] Flavonoids are polyphenols, secondary plant metabolites and bioactive compounds naturally occurring in plants and plant-derived products, which can be differentiated into six main classes: flavones, flavonols, flavanols, flavanones, anthocyanins, and flavan-3-ols. Anthocyanins are further classified into six subclasses: pelargonidin, cyanidin, delphinidin, peonidin, petunidin, and malvidin. Common US dietary sources of anthocyanins include berries, blackcurrants, red grapes, plums, and cherries, as well as red wine, fruit juice, and some vegetables such as radishes.[167,168]

Physical activity assessment

PA level was collected at baseline using a PA questionnaire, measured as a single question with categorical responses, administered within the lifestyle questionnaire (Table 8).[169] On a scale from 0–7, participants were asked to identify the statement which best described their habitual level of PA over the past month: (0) Avoid walking or exertion (e.g., always use elevator, drive whenever possible instead of walking, biking, or rollerblading); (1) walk for pleasure, routinely use stairs, occasionally exercise sufficiently to cause heavy breathing or perspiration; (2) 10 to 60 minutes per week; (3) over one hour per week; (4) run less than 1 mile per week or spend less than 30 min per week in comparable PA; (5) run 1 to 5 miles per week or spend 30 to 60 min per week in comparable PA; (6) run 5 to 10 miles per week or spend 1 to 3 hours per week in comparable PA; and (7) run over 10 miles per week or spend over 3 hours per week in comparable PA.

PA information was self-reported and has not been validated.[170] Nonetheless, previous validation studies have confirmed the accuracy of prediction models for functional aerobic capacity with self-reported PA data, without exercise testing, compared to objective maximal oxygen consumption; VO2max.[169,171] Our PA question specifically captured PA defined as all modes of movement caused by muscle activity resulting in increased energy expenditure. Nevertheless, we were able to compare the self-reported PA question with physical fitness. The correlation coefficient between PA and cardiorespiratory fitness as measured by VO2max was r=0.41, indicating a moderate correlation. Although these concepts are not identical, as fitness reflects muscle strength, endurance and motor ability, these measures are well known to have moderate to very good correlations.

Table 8. Self-reported physical activity questionnaire.

0-1: I did not participate regularly in programmed recreation, sport, or heavy physical activity.	
0 -	Avoid walking or exertion (as an example, always use elevator, drive whenever possible instead of walking, biking or rollerblading).
1 -	Walk for pleasure, routinely use stairs, occasionally exercise sufficiently to cause heavy breathing or perspiration.
2-3: I participated regularly in recreation or work requiring modest physical activity, such as golf, horseback riding, calisthenics, gymnastics, table tennis, bowling, weightlifting, yard work.	
2 -	10 to 60 minutes per week.
3 -	Over one hour per week.
4-7: I participated regularly in heavy physical exercise such as running or jogging, swimming, cycling, rowing, skipping rope, running in place, or engaging in vigorous aerobic activity type exercise such as tennis, basketball, or handball.	
4 -	Run less than 1 mile per week or spend less than 30 minutes per week in comparable physical activity.
5 -	Run 1 to 5 miles per week or spend 30 to 60 minutes per week in comparable physical activity.
6 -	Run 5 to 10 miles per week or spend 1 to 3 hours per week in comparable physical activity.
7 -	Run over 10 miles per week or spend over 3 hours per week in comparable physical activity.

Physical Activity in the Past Month. Below circle ONE of the values (0 to 7) which best represents your general ACTIVITY LEVEL for the PREVIOUS MONTH.

The MEDLIFE Index

A description of the adapted MEDLIFE index for the "Feeding America's Bravest" trial is provided in Table 9. Due to differences in data collection, Appendix 2C indicates the eleven modifications and 2 exclusions made to the original MEDLIFE index to best fit the available baseline data and holistically define the overall concept of the Mediterranean lifestyle. Each item was weighted equally with 0 or 1 point, creating a theoretical scoring range from 0 (worst) to 26 (best). Final scores were then categorized into tertiles of MEDLIFE adherence. Items regarding food consumption were derived from the FFQ, while information on eating habits and dietary behaviors, sleep behaviors, and PA were obtained from the self-reported lifestyle questionnaire administered at baseline for the participants to complete and return online of their own accord.[162,169,172,173]

Table 9. Description of the Mediterranean Lifestyle (MEDLIFE) index modified for "Feeding America's Bravest".

Item	Components (serving size)	Criteria for 1 point
Block 1: Mediterranean food consumption		
1. Sweets	milk chocolate, dark chocolate, candy bars, candy (1 oz.), cookies, brownies, doughnuts, cake, pie, muffins, biscuits (1 unit) pancakes, waffles (2 small units)	≤ 2 servings/wk
2. Red meat	Beef, pork, lamb as main dish, mixed dish, or sandwich (4-6 oz.)	< 2 servings/wk
3. Processed meats	Hamburger, hotdog (1 unit), salami, bologna, other processed meat (2 oz.), chicken or turkey hotdogs or sandwich, sausage, frozen dinner (1 unit), bacon (2 slices), beef liver (4 oz.), chicken liver (1 oz.)	≤ 1 serving/wk
4. Eggs	Regular eggs including yolk and omega-3 fortified including yolk	2-4 units /wk
5. Legumes	Beans or lentils, baked, dried, or soup, peas, lima beans (1/2 cup)	≥ 2 servings/wk
6. White meat	Chicken or turkey with or without skin (3 oz.)	2 servings/wk
7. Fish	Dark meat fish (tuna steak, mackerel, salmon, sardines, bluefish, swordfish), other fish (3-5 oz.), canned tuna (3-4 oz.), shrimp, lobster, scallops as a main dish	≥ 2 servings/wk
8. Potatoes	potatoes, baked, boil (1 unit) or mashed (1 cup) and French fries (6 oz. or 1 serving)	≤ 3 servings/wk
9. Dairy products	Skim, 1 or 2%, whole, soy milk (8 oz.), cream (1 Tbs), frozen yogurt, sherbet, ice cream, plain or sweetened yogurt (1 cup), cottage or ricotta cheese (1/2 cup), margarine, butter, cream cheese, other cheese (1 oz./1 slice)	2 servings/d
10. Nuts	Nuts (e.g., walnuts, almonds, hazelnuts, pistachio, peanuts)	1-2 serving/d
11. Fruit	Raisins (1 oz), grapes (1/2 cup), prunes (6 units), apple, orange, grapefruit, prune, and other fruit juices (small glass), bananas (1 unit), cantaloupe (1/4 melon), grapefruit, avocado (1/2 fruit or cup), apples, pears, oranges, peaches, plums, apricots (1 unit), strawberries, blueberries fresh, frozen, or canned (1/2 cup)	3-6 servings/d
12. Vegetables	Tomatoes (2 slices), tomato or carrot juice (small glass), broccoli, string beans, cauliflower, cabbage, Brussel sprouts, raw or cooked carrots, corn, mixed vegetables, yams, sweet potatoes, squash, eggplants, zucchini, kale, cooked spinach, cooked onions (1/2 cup), spinach (1 cup), head or leaf lettuce (1 serving), celery (2-3 sticks), peppers (3 slices), raw onion (1 slice)	≥ 2 servings/d
13. Olive oil	Olive oil added to food or bread (1 Tbs.) Main oil usually used for frying and sautéing at home?	≥ 1 servings/d + Olive oil
14. Cereals	Cold breakfast cereal (1 serving), oatmeal (1 cup), other cooked cereals (1 cup) white bread (1 slice), rye bread (1 slice), whole grain bread (1 slice), English muffin, bagel, rolls (1 unit), brown rice (1 cup), white rice (1 cup), pasta, noodles, couscous (1 cup), tortillas (2 units), pizza (2 slices)	3-6 servings/d
Block 2: Mediterranean dietary habits		
15. Water, coffee, and tea	Which of the following non-alcoholic beverages do you most frequently drink at home? Which of the following non-alcoholic beverages do you most frequently drink at the firehouse? Water (bottled, sparkling, or tap), herbal tea, tea with caffeine, decaffeinated tea, coffee with caffeine, decaffeinated coffee (8 oz.)	Water, coffee, and tea are most frequent non-alcoholic drinks at home and firehouse + ≥ 6 cups/d of water, coffee, or tea
16. Wine	When you drink alcoholic beverages, what type do you drink?	Red or white wine are the usual alcoholic beverage
17. Limit salt	Sodium (mg)	< 2.3 g/d
18. Preference for whole grain products	Is whole grain the main type of bread or starch that you eat?	Yes
19. Snacks	Regular popcorn (3 cups), fat free popcorn (3 cups), potato chips (small bag or 1 oz), crackers (6), pretzels (1 small bag or serving), breakfast, energy, and low carb bars (1 unit)	≤ 2 serving/wk
20. Limit sugar in beverages (sugary beverages)	Carbonated beverages caffeinated or caffeine-free with sugar and other sugared beverages: punch, iced tea, lemonade, sports drinks (1 glass, bottle, or can)	≤ 1 serving/wk
21. Local, seasonal, or organic products	Do you usually consume local/seasonal or organic products?	Yes
Block 3: Physical activity, rest, social habits, and conviviality		
22. Physical activity	Statement that best describes your physical activity in the past month	Run >10 miles/wk or spend >3 hrs/wk in comparable physical activity
23. Siesta/nap	How many times do you take a nap per week?	≥ 3 naps/wk
24. Hours of sleep	Total hours of actual sleep in a typical 24-hour period	6-8 hrs/d
25. Watching TV	During the past weeks, what was your average total time per week at each of the following activities?	≤ 4 hrs/wk
26. Time spent eating	Time you usually spend eating each meal at home (in minutes). Time you usually spend eating each meal at the firehouse (in minutes).	≥ 20 min

Abbreviations: min: minutes, h: hours, d: day, wk: week, oz.: ounces, g: grams, Tbsp: tablespoons

6. Covariate assessments

Information on sociodemographic characteristics, dietary intake, a 13-item mMDS, lifestyle habits, anthropometric measurements, and medical history were collected at baseline through in-person data collection, an online lifestyle questionnaire, or medical record after informed consent was given. Anthropometric measurements were collected by the study team at the time of enrollment, which marked the participants' baseline visit. BMI was calculated by dividing weight by height squared (kg/m^2). Total daily energy intake and micronutrient intakes were calculated using the baseline FFQ. Adjustment for alcohol intake considered servings of beer and distilled beverages per week because wine consumption was already included in the MEDLIFE index. The mMDS score, described in Table 11, was reproduced based on previously employed definitions by Yang et al. and Sotos-Prieto et al..[162,172] Participants with dyslipidemia, hypertension, or T2D were identified if they self-reported a previous diagnosis or were being treated with lipid-lowering, antihypertensive, or antidiabetic medications, respectively within the previous year to enrollment.

The mMDS index for firefighters has been validated with a panel of plasma and urine biomarkers and the Harvard FFQ. The validation study showed a high correlation between the lifestyle questionnaire's 13-item mMDS and the mMDS derived from the 131-item FFQ ($r = 0.74$). Furthermore, a good correlation was found between the FFQ nutrient intake and plasma biomarkers (omega-3, EPA and DHA).[172] This firefighter-specific mMDS was developed and assessed in Midwestern career firefighters. The index was based on previously validated scores, mainly the MDS, mMDS, and MEDAS, as well as dietary data both at home and at the firehouse.[130,141,174] This mMDS has been previously associated with a decreased total cholesterol:HDL cholesterol ratio and increased HDL cholesterol among our study population.[173] Previous to the "Feeding America's Bravest" trial, greater mMDS adherence was associated with a lower prevalence of metabolic syndrome, better lipid profiles, and lower risk of weight gain within a longitudinal study of firefighters.[162]

Table 11. Description of the mMDS score developed using the "Feeding America's Bravest" diet and lifestyle questionnaire.

mMDS items	Components	Score range	Points for each answer								
1. Fast-food or Take-out food	How many times per week do you eat the following?	0-4	never	≤ 1	2-3	3-4	5-6	every day	8-10	≥ 11	missing
			4 pts	3 pts	2 pts	2 pts	1 pts	1 pts	0 pts	0 pts	0 pts
2. Fruits	How many servings of each of the following do you consume per day?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7	missing		
			0 pts	1 pts	2 pts	3 pts	4 pts	4 pts	0 pts		
3. Vegetables (not including potatoes)	How many servings of each of the following do you consume per day?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7	missing		
			0 pts	1 pts	2 pts	3 pts	4 pts	4 pts	0 pts		
4. Legumes (e.g. beans, chickpeas, lentils)	How many servings of each of the following foods do you eat per week?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7	missing		
			0 pts	0 pts	1 pts	2 pts	3 pts	4 pts	0 pts		
5. Nuts (e.g. walnuts, almonds, hazelnuts, pistachio, peanuts)	How many servings of each of the following foods do you eat per week?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7	missing		
			0 pts	0 pts	1 pts	2 pts	3 pts	4 pts	0 pts		
6. Sweet Desserts (cake, cookies, pie, ice cream, etc.)	How many times per week do you eat the following?	0-4	never	≤ 1	2-3	3-4	5-6	≥ 7	missing		
			4 pts	4 pts	3 pts	2 pts	1 pts	0 pts	0 pts		
7. Primary cooking oil/fat use at home[1]	Which oil or fat do you use most often for cooking and serving food at home?	0-5	Butter	Lard or other animal fat	Margarine	Corn or vegetable oil	Benechol or Smart Balance	Olive oil	EVOO	other	missing
			0	0 pts	1 pts	2 pts	3 pts	4 pts	5 pts	0 pts	0 pts
8. Primary cooking oil/fat use at work[2]	Which oil or fat do you use most often for cooking and serving food at the firehouse?	0-5	Butter	Lard or other animal fat	Margarine	Corn or vegetable oil	Benechol or Smart Balance	Olive oil	EVOO	other	missing
			0 pts	0 pts	1 pts	2 pts	3 pts	4 pts	5 pts	0 pts	0 pts
9. Fried foods (French fries, fried chicken, chicken nuggets, etc.)	How many times per week do you eat the following?	0-4	never	≤ 1	2-3	3-4	5-6	every day	8-10	≥ 11	missing
			4 pts	3 pts	2 pts	1 pts	0 pts	0 pts	0 pts	0 pts	0
10. Breads/starches consumed at home[1]	Which bread or starch do you most frequently eat at home?	0-4	I do not eat bread or starch	White bread, filled pasta, white rice, or potatoes	Durum wheat bread or dry pasta	French bread or Italian bread or multigrain or other crusty bread	Whole wheat bread or brown rice or whole wheat pasta	missing			
			3 pts	0 pts	3 pts	2 pts	4 pts	0 pts			

11. Breads/starches consumed at work[2]	Which bread or starch do you most frequently eat at the firehouse?	0-4	I do not eat bread or starch	White bread, filled pasta, white rice, or potatoes	Durum wheat bread or dry pasta	French bread or Italian bread or multigrain or other crusty bread	Whole wheat bread or brown rice or whole wheat pasta	missing								
			3 pts	0 pts	3 pts	2 pts	4 pts	0 pts								
12. Baked, broiled, grilled, or blackened (NOT fried) ocean fish (salmon, tuna, cod, haddock, etc.)	How many times per week do you eat the following?	0-4	never	≤ 1	2-3	3-4	5-6	every day	8-10	≥ 11	missing					
			0 pts	1 pts	2 pts	3 pts	4 pts	4 pts	4 pts	4 pts	0 pts					
13. Non-alcoholic beverages at home[1]	Which of the following non-alcoholic beverages do you most frequently drink at home?	0-4	Cola/ soda	Diet cola/soda	Fruit drink or punch	Milk	Tea/ coffee	Juice	Water	Other	missing					
			0 pts	1 pts	1 pts	1 pts	2 pts	2 pts	4 pts	0 pts	0 pts					
14. Non-alcoholic beverages at work[2]	Which of the following non-alcoholic beverages do you most frequently drink at the firehouse?	0-4	Cola/ soda	Diet cola/soda	Fruit drink or punch	Milk	Tea/ coffee	Juice	Water	Other	missing					
			0 pts	1 pts	1 pts	1 pts	2 pts	2 pts	4 pts	0 pts	0 pts					
15. Quantity of alcoholic beverages	How many alcoholic beverages (beer, wine, hard liquor, etc.) do you drink over a typical week?	0-4	I do not drink	0	1-2	3-4	5-6	7-8	9-10	11-12	13-14	15-16	17-18	19-20	≥ 21	missing
			0 pts	0 pts	2 pts	2 pts	4 pts	4 pts	4 pts	4 pts	4 pts	4 pts	4 pts	4 pts	1 pts	0 pts
16. Wine consumption	When you drink alcoholic beverages, what type do you drink?	0-2	White wine	Red wine	Beer	Hard liquors	Don't drink									
			2 pts	2 pts	0 pts	0 pts	0 pts									

[1] Weighted by the proportion of meals at home relative to the total number of meals per week (breakfast + lunch + dinner)

[2] Weighted by the proportion of meals at the firehouse (or on work time) relative to the total number of meals per week (breakfast + lunch + dinner)

The possible responses indicated in the top row are matched with their corresponding points in the row directly below.

Calculation of the modified Mediterranean diet score (mMDS):mMDS1+(mMDS2+mMDS3)+mMDS4+mMDS5+mMDS6+mMDS9+mMDS12+mMDS15+mMDS16+(mMDS7+mMDS10+mMDS13)*(1-fh) +(mMDS8+mMDS11+mMDS14)*fh

7. Outcome assessments

Primary outcomes assessed in this study population included lipid profile measures; TG, total cholesterol, HDL cholesterol, LDL cholesterol, and ratios for LDL cholesterol:HDL cholesterol, TG:HDL cholesterol, and total cholesterol:HDL cholesterol, as well as metabolic syndrome. Baseline measures within the last year from enrollment in the study were gathered from the PSM electronic medical record database. Baseline characteristics, anthropometric measurements, and cardiometabolic parameters are described by group assignment in Appendix 2D.

Lipid profile

Baseline lipid panels were collected separately during participants' fire department medical examinations, which were conducted at the contracted PSM clinics independent of the research study. This collaboration with the PSM clinics facilitated a much wider data collection on health outcomes and biochemical assessments that would not have been possible via questionnaires and study team visits. Blood samples were collected after an overnight fast at baseline and at follow-up. Plasma and serum samples were collected in the 15-mL tubes as appropriate for each assay, aliquoted, frozen at −80 °C, and then stored. Blood lipid profiles were determined using standardized automated high-throughput enzymatic analyses, which achieved coefficients of variation of ≤3% for cholesterol and ≤5% for TG, using cholesterol assay kit and reagents Ref:7D62–21 and TG assay kit and reagents Ref:7D74–21 by ARCHITECT c System, Abbott Laboratories, IL, USA.

Metabolic Syndrome

We used the harmonized definition of metabolic syndrome established in 2009, which requires meeting at least three of the following five criteria; abdominal obesity (waist circumference ≥102 cm in men, or ≥88 cm in women for Caucasians); elevated blood glucose (≥100 mg/dl or treatment with antidiabetic drugs); high blood pressure (systolic ≥130 mm Hg or diastolic ≥85 mm Hg, or receiving antihypertensive drugs); TG ≥150 mg/dl; serum HDL cholesterol <40 mg/dl in men or <50 mg/dl in women.[26] Secondary outcomes included each of the five potential metabolic syndrome components, in addition to BMI, waist circumference, percent body fat, TG, total cholesterol, HDL cholesterol, LDL cholesterol, total cholesterol:HDL cholesterol, and plasma glucose levels as continuous outcomes.

Specific methods *(this section is a reiteration of the Statistical Analysis of each article)*

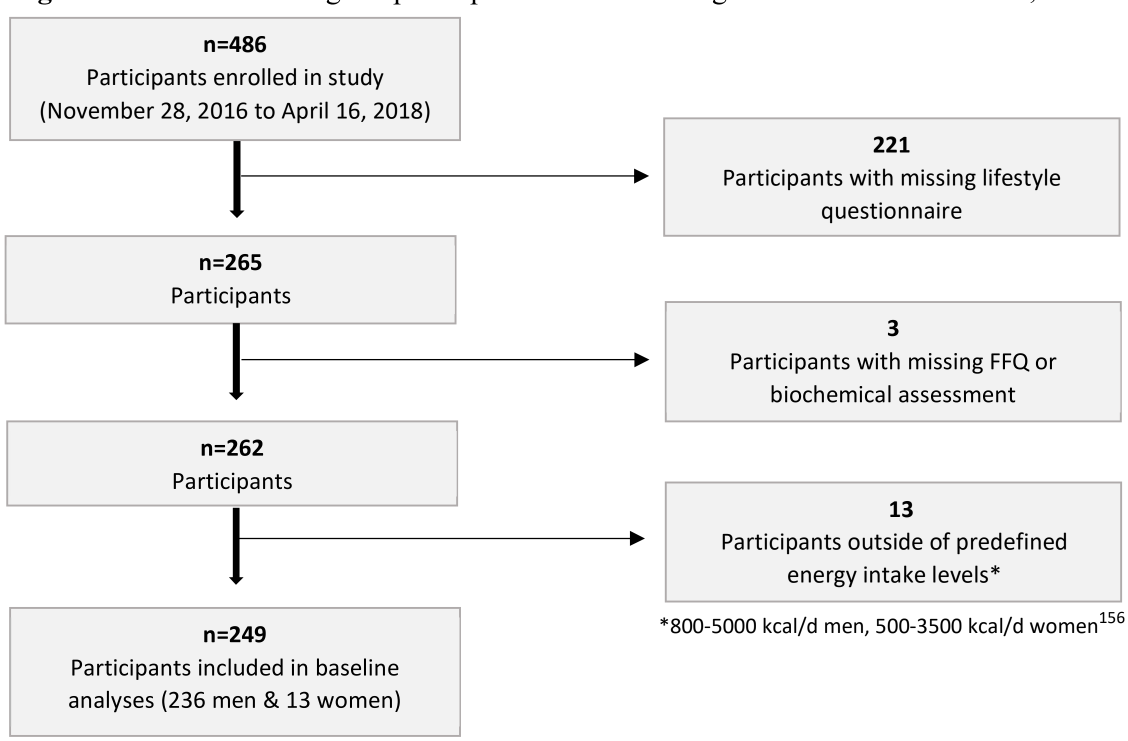
2.1. Anthocyanin Intake and Physical Activity: Associations with the Lipid Profile in a US Working Population

Specific study design and aim – In this study, we conducted a cross-sectional analysis with the "Feeding America's Bravest" baseline data collection. Our aim was to cross-sectionally study the joint effect and possible synergism between anthocyanin intake and PA on the lipid profile of Midwestern US career firefighters.

1. Inclusion and exclusion criteria

Although 486 persons were enrolled in the "Feeding America's Bravest" trial, only 265 participants completed the baseline lifestyle questionnaire between November 28, 2016, and April 16, 2018. Additionally, participants with missing FFQ or biochemical assessment (n=3) and participants whose energy intake exceeded predefined levels (men: 800–5000 kcal/d, women: 500–3500 kcal/d) (n=13) were excluded, leaving a total of 249 participants available for analyses (Figure 7).

Figure 7. Flowchart of eligible participants in the "Feeding America's Bravest" trial, 2016-2019.



2. Statistical analysis

A continuous variable for total anthocyanin intake was transformed into units of SDs using the standardization method. Thereafter, a dichotomous variable of total anthocyanins was created to define high and low anthocyanin intake using the median as the cut-off point; the median intake was equivalent to 19.14 mg/day. PA was used as a continuous variable considering each unit (level) increase. As a dichotomous variable, high PA was defined as regularly participating in heavy physical exercise, such as running or jogging, swimming, cycling, etc., or engaging in vigorous aerobic activity, such as tennis, basketball, or handball (levels 4–7), whereas low PA represented none to regular recreation or work requiring modest PA, such as golf, horseback riding, calisthenics, gymnastics, table tennis, bowling, weightlifting, and gardening (levels 0–3).

Baseline characteristics of participants were presented according to low and high categories of anthocyanin intake and PA. Quantitative values were expressed as mean ± SD and qualitative variables as a percentage. Statistical significance of between-group variation between low and high categories for each exposure were tested using Student's t-test for quantitative variables and chi-squared test for qualitative variables.

To determine the contribution of each food source to the between-person variance of total anthocyanin intake, stepwise-selection regression analyses and nested least-squares linear regression models were conducted. The cumulative R^2 indicates the proportion of variability with the addition of each source, whereas the change in cumulative R^2 identifies each source's contribution to the total variability of anthocyanin intake. Moreover, the contribution of anthocyanins from each food source was presented as a percentage of the total anthocyanin intake. Additionally, subclasses of anthocyanins were presented as percentages of the total anthocyanin intake.

Multivariable linear regression models were used to determine the extent to which each continuous exposure of anthocyanin intake and PA level predicted lipid profile measures. Beta coefficients were reported with 95% CIs and *p*-values presented for each adjusted model. To control for potential confounding, multivariable adjusted models included age (years), sex (male/female), BMI (kg/m^2), total energy intake (kcal/d), mMDS (points), smoking status (never, current, or former), maximum attained educational level (technical school, some college, associate degree/Bachelor's degree or higher), marital status (married/single), multivitamin use (yes/no), supplement use (yes/no), sleep (hours/day), prevalent hypertension, dyslipidemia, and T2D (yes/no). Additionally, the multivariable linear regression models for anthocyanin intake were adjusted for PA level, total time spent sitting down (hours/week), and time spent in front of the television, computer and in the car (hours/week), whereas the fully adjusted models for PA were adjusted for anthocyanin intake. Total time sitting and sedentary behavior showed a correlation coefficient of 0.28, indicating these covariates measured different forms of inactivity. A sensitivity analysis considered additional exclusions for chronic diseases, women, and supplement use.

To assess the potential effect modification between anthocyanin intake and PA on HDL cholesterol, we followed the recommendations provided by Knol and Vanderweele.[84] First, the prevalence within each subgroup was presented as a percentage and the joint effect of the four possible combinations of low and high exposures to both anthocyanin intake and PA on HDL cholesterol < 40 mg/dL as relative risks, adjusted for age, sex, and total energy intake. Relative risks were calculated using generalized linear models with Poisson distribution and robust standard errors.[175] A stratification analysis tested effect modification by assessing each dichotomous exposure stratified by the other. This was followed by a comprehensive interaction analysis by applying both multiplicative and additive interaction analyses. Multiplicative interaction was tested by comparing age-, sex-, and energy-adjusted models with and without the interaction term, whereas the RERI was assessed on the additive scale.[88]

All analyses were conducted with Stata version 14.0 (StataCorp, College Station, TX, USA). All *p*-values are two-sided and were considered statistically significant at $p < 0.05$.

2.2. The Mediterranean Lifestyle (MEDLIFE) Index and Metabolic Syndrome in a non-Mediterranean Working Population

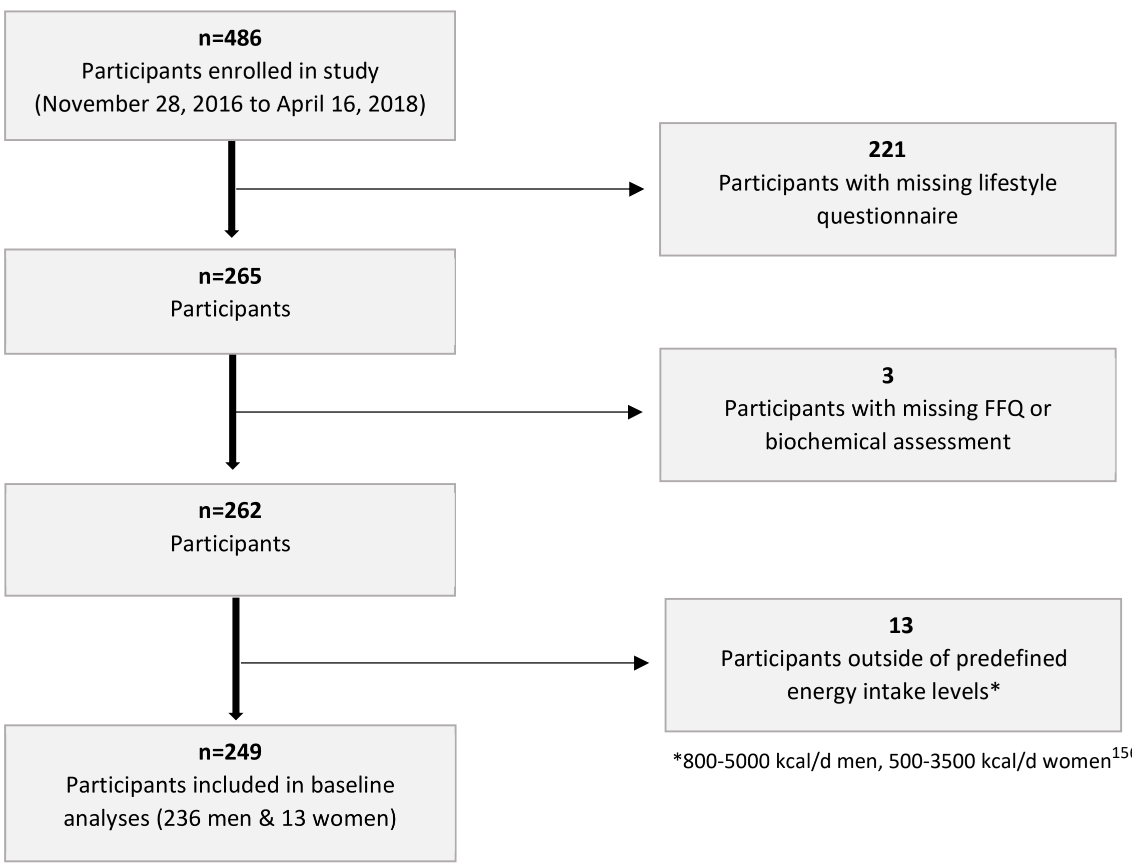
Specific study design and aim – In this study, we conducted a cross-sectional analysis with the “Feeding America’s Bravest” baseline data collection. Our aim was to cross-sectionally evaluate the association between the MEDLIFE index and metabolic syndrome in a non-Mediterranean working population at high CVD risk.

1. Inclusion and exclusion criteria

Among the US career firefighter study population, we included participants who completed a baseline lifestyle questionnaire between November 28, 2016 and April 16, 2018 (n=265) and excluded participants with a missing FFQ or biochemical assessment (n=3) or whose total daily energy intake exceeded predefined levels (men: 800–5000 kcal/d, women: 500–3500 kcal/d)[156] (n=13). A total of 249 participants were left for evaluation (Figure 8).

MEDLIFE scores required data from the online lifestyle questionnaire, which was completed and returned on the participant's own accord. Of the total study population, 45% of the missing data for the calculation of MEDLIFE scores primarily derives from the lack of completion of this questionnaire. Due to incompatibilities with the firefighter's work schedule, completion of the FFQ was required in person at the baseline study team visit, whereas the lifestyle questionnaire (online Qualtrics) was to be completed out of work hours and thus was not completed by all active participants.

Figure 8. Flowchart of eligible participants in "Feeding America's Bravest", 2016-2019.



2. Statistical analysis

Variables with quantitative values were expressed as means ± SD and those characterized qualitatively as a percentage. The inverse probability weighting method was used to present age-, sex-, and energy intake-adjusted baseline characteristics of participants, as well as age-and-sex adjusted MEDLIFE characteristics, according to tertiles of MEDLIFE adherence. Statistical significance of between-group comparisons for each characteristic was tested with a post-estimation contrast of adjusted means across MEDLIFE tertiles.

To determine the contribution of each block to the between-person variance of total MEDLIFE scores, a linear regression with Shapley and Owen decomposition of R^2 analysis was conducted.[176] The R^2 as a percentage identifies each block's contribution to the total variability of MEDLIFE scores.

Multivariable logistic regression models were used to assess the association between metabolic syndrome and adherence to the MEDLIFE index. We also assessed the association between the MEDLIFE index and each component of the metabolic syndrome: abdominal obesity, hyperglycemia, hypertension, hypertriglyceridemia, and low HDL cholesterol. ORs were reported with 95% CIs and linear *p*-for-trends calculated across tertile medians for each model. To control for potential confounding, an initial multivariable adjusted model included age (years), sex (male/female), BMI (kg/m^2), total energy intake (kcal/d), smoking status (never, current, or former), and education level (technical school, some college, associate degree/Bachelor's degree or higher). A final multivariable model additionally adjusted for potential confounders, including alcohol intake other than wine (g/d), marital status (married/single), multivitamin use, supplement use, sleep medication use (yes/no), prevalent T2D, hypertension, and dyslipidemia (yes/no). Possible confounders, including BMI, prevalent T2D, hypertension, and dyslipidemia, were excluded from models with corresponding outcomes, respectively.

Multivariable linear regression models were used to determine the extent to which each tertile of MEDLIFE adherence predicted continuous outcomes, including BMI, waist circumference, body fat percentage, TG, total, HDL cholesterol, LDL cholesterol, total cholesterol:HDL cholesterol, and plasma glucose levels. Beta coefficients with 95% CI and *p*-for-trends were reported across MEDLIFE tertiles for each model. An initial multivariable model adjusted for age, sex, BMI, total energy intake, smoking status, and education level. A fully adjusted model additionally adjusted for other sources of alcohol intake different from wine, civil status, multivitamin use, supplement use, sleep medication, prevalent hypertension, dyslipidemia, and T2D.

Lastly, multivariable logistic regression models were conducted to assess the effect of each item (1 pt. vs 0 points), block, and additional point of the MEDLIFE score (as continuous variable) on metabolic syndrome, adjusting for age, sex, total daily energy intake, other sources of alcohol intake different from wine, smoking status, education level, civil status, multivitamin use, supplement use, sleep medication, and the remaining items or blocks, respectively.

Sensitivity analyses were conducted for metabolic syndrome across MEDLIFE tertiles with additional exclusions of women (n=13), participants reporting caloric intake beyond Willett's total daily energy intake limits; 800-4000 kcal/d for men and 500-3500 kcal/d for women[156] (n=20), participants with baseline clinical measurements earlier than 6 months prior to enrollment (n=126) and those with prevalent hypertension, T2D, or dyslipidemia (n=50). Additional subgroup analyses on metabolic syndrome were conducted for age (median cut-off point = <47 years, ≥47 years), BMI (<30 kg/m^2, ≥30 kg/m^2), total daily energy intake (median cut-off point = <2204, ≥2204 kcal/d) and smoking status (never/former or current), with corresponding *p*-for-trend across tertiles and *p*-for-interaction using tertile 1 as the reference category and tertile 2 + tertile 3 as a single category for higher MEDLIFE adherence. Lastly, a substitution of Block 1: Mediterranean food consumption for the mMDS was conducted to further test our primary findings.

All analyses were conducted with Stata version 14.0 (StataCorp, College Station, TX). All *p*-values are two-sided and were considered statistically significant at $p<0.05$.

RESULTS

1. Br. J. Nutr.: Q2 NUTRITION & DIETETICS 48/88; IF: 3.718

Hershey MS, Martínez-González MA, Álvarez-Álvarez I, Martínez-Hernández JA, & Ruiz-Canela M. The Mediterranean diet and physical activity: Better together than apart for the prevention of premature mortality. *British Journal of Nutrition*. 2021;1-12. doi:10.1017/S0007114521002877

2. Molecules: Q2 BIOCHEMISTRY & MOLECULAR BIOLOGY 115/295; IF: 4.412

Hershey MS, Sotos-Prieto M, Ruiz-Canela M, Martínez-González MA, Cassidy A, Moffatt S, & Kales SN. Anthocyanin Intake and Physical Activity: Associations with the Lipid Profile of a US Working Population. *Molecules*. 2020;25(19):4398. doi:10.3390/molecules25194398

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PUBLICATION 1

Hershey MS, Martínez-González MA, Álvarez-Álvarez I, Martínez-Hernández JA, & Ruiz-Canela M. The Mediterranean diet and physical activity: Better together than apart for the prevention of premature mortality. *British Journal of Nutrition*. 2021;1-12. doi:10.1017/S0007114521002877

British Journal of Nutrition, page 1 of 12 doi:10.1017/S0007114521002877

The Mediterranean diet and physical activity: better together than apart for the prevention of premature mortality

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(Submitted 17 March 2021 – Final revision received 15 July 2021 – Accepted 27 July 2021)

Abstract

Diet and physical activity (PA) have been studied extensively in epidemiology as single or combined lifestyle factors; however, their interaction has not been studied thoroughly. Studying potential synergisms between lifestyle components with a comprehensive interaction analysis, including additive measures of interaction, provides key insights into the nature of their joint effect and helps target interventions more effectively. First, a comprehensive review was conducted to assess the potential research gap regarding reported interaction analyses conducted in studies assessing the Mediterranean diet (MedDiet) in combination with PA on all-cause mortality. Thereafter, we prospectively assessed the joint association of the MedDiet with PA on all-cause mortality in the *Seguimiento Universidad de Navarra* (SUN) cohort, followed by both multiplicative and additive interaction analyses. The conjoint effect of low adherence to the MedDiet and low PA observed an increased risk greater than the individual risk factors, suggesting a potential additive interaction or synergism between both exposures, with relative risk due to interaction (RERI) and (95 % confidence interval (95 % CI)) = 0·46 (–0·83 to 1·75) and attributable proportion (95 % CI) due to interaction of 36 % (–0·62, 1·34). No multiplicative interaction was detected. Studying interactions between lifestyle factors, such as the MedDiet and PA, is particularly relevant given the current research gaps in studying the complexities of combined aspects of lifestyle in comparison with isolated behaviours. Our findings underline the important public health message of adhering to both the MedDiet and PA for the prevention of premature mortality.

Key words: Mediterranean diet: Physical activity: Interactions: Additive interaction: All-cause mortality: Lifestyle factors

Individual and combined effects of diet and physical activity on health

The disease burden of poor diet quality has globally increased during the last 30 years with more than 11 million deaths attributable to dietary risk factors in 2017[1]. During this time, the development of nutritional epidemiology has been impressive[2]. A key contribution to this field has been the shift of focus from assessing isolated dietary factors to studying the effects of overall or complete dietary patterns. Dietary indices, constructed to measure adherence to specific dietary patterns as indicators of overall diet quality, have allowed epidemiologists to establish inverse associations between a healthy food pattern and multiple health outcomes[3]. In this context, the Mediterranean dietary pattern (MedDiet) is internationally recognised as one of the best dietary strategies for the prevention of chronic diseases and premature death[4–7].

Physical inactivity is also a major and globally relevant determinant of health[8]. There is abundant evidence of the effect of

Abbreviations: AP, attributable proportion; HR, hazard ratio; MDS, Mediterranean diet score; MedDiet, Mediterranean dietary pattern; PA, physical activity; RERI, relative excess risk due to interaction; SUN, Seguimiento Universidad de Navarra.

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physical activity (PA) on health for the prevention of chronic diseases and premature mortality, whereas a lack of PA is a key risk factor for these health outcomes[9–12]. It has been demonstrated that the replacement of PA or exercise with inactivity or sedentary behaviour will eventually adversely affect the ageing process, whatever the age of the individual. Even a simple indicator of PA, such as time spent sitting, is an independent predictor of mortality. The increase in risk of lifestyle and age-associated diseases are attributed to the decline in functional levels of many body systems and thus suboptimal maintenance of physiological functions in sedentary individuals[12].

Moreover, diet and PA are two of the most frequently addressed modifiable lifestyle risk factors, which increase morbidity and mortality from lifestyle diseases, including CVD, obesity, type 2 diabetes and some cancers. Hand in hand, diet and PA are frequently recommended in clinical practice for general health promotion, weight loss or weight maintenance, chronic disease prevention, and increased quality of life[13]. Diet and PA are considered multidimensional variables that can influence each other[14]. According to data from NHANES 2003–2006, US adults were 32 % more likely to eat a healthy diet if they met PA guidelines[15]. When considering the energy balance equation, diet (pertinent to energy intake) and PA (pertinent to energy expenditure) find themselves on either side of the equation, suggesting that both factors influence each other to maintain a healthy weight, possibly more so than the sources of energy themselves[14,16,17].

The Mediterranean diet pyramid underlines the importance of PA and other lifestyle factors beyond diet[18,19]. Furthermore, existing evidence already suggests that greater adherence to both the MedDiet and PA is associated with better health biomarkers, lower risk of disease and lower mortality when compared with the MedDiet or PA alone[6,7,20–22]. A meta-review from nine systematic reviews and twenty-four meta-analyses concluded that the MedDiet may reduce the risk of non-communicable diseases, improve health status and reduce total lifetime healthcare costs, with a possibly even greater effect when combined with PA, as long as tobacco and excessive alcohol consumption are avoided[21]. Existing literature supports that a healthy MedDiet and PA may be more effective when acting in combination rather than separately, but questions remain on exactly how, to what extent, and to whom is this combined effect most beneficial to target interventions in public health. Little has been studied on the *a priori* analysis of the interaction between diet and PA to determine its impact on hard clinical events or mortality thus far[23,24]. The existing studies have almost never quantified the synergism between diet and PA. More comprehensive, methodical, and robust evidence is needed to demonstrate that diet and PA are two sides of the same coin, as well as to identify to whom this combination may offer the greatest benefit in public health.

Interaction analysis for the potential synergism between the Mediterranean diet and physical activity

An interaction is defined as the situation in which the effect of one exposure on an outcome differs across the strata of another exposure, implying that the risk differences vary across strata of the other exposure. Thus, the presence of interaction suggests that the effect of the two exposures is different from the mere sum or multiplication of their individual effects, depending on the nature of the association between exposures and the assumed scale (additive or multiplicative) for the interaction. This interrelation of effects suggests that the reduction of either factor would also reduce the risk of the other factor in producing a given outcome[25]. Different terminology is used throughout the scientific community to refer to the concept of interaction: joint effect or combined effect, synergy, interdependence, heterogeneity of effects, non-uniformity of effects, effect modification, or subgroup analysis[26]. For the purpose of this article, the term interaction will refer to the 'mechanistic or biological interaction' created when two potential causal risk factors participate in the same causal mechanism, which implies either synergism or antagonism between factors on disease risk or death[25,27].

The current criteria within the Strengthening the Reporting of Observational Studies in Epidemiology guidelines recommend describing any methods used to examine interactions or subgroups within the statistical analysis section of the study methods[28]. However, many studies fall short of this recommendation[26]. In 2009, Knol et al. evaluated the presence of interaction in 225 epidemiological studies to examine how interaction was assessed and reported. This literature search found that not all studies that addressed effect modification or interaction provided satisfactory information on interactions between exposures (primarily treatments, medical conditions and lifestyle factors). Moreover, only one out of ten studies reported adequate information for a full assessment of additive or multiplicative interaction[29]. This is important because an adequate reporting of methods allows for higher transparency, direct interpretation, comparison and independent recalculation of results[30].

There are a variety of statistical approaches for considering interactions between potential causal factors. The most frequently reported method includes conducting a likelihood ratio test to compare regression models with and without the multiplicative interaction product term. However, this most common analysis of interaction on the multiplicative scale is limited to assessing statistical interaction. The current tendency among observational studies to simply report statistical significance of the likelihood ratio test on the multiplicative scale is due to the implicit nature of epidemiological statistical modelling and software convenience[27,31]. When obtaining relative risks, the inclusion of a product term in multivariable regressions provides a quick analysis for investigators to report interactions with a corresponding *P*-value, usually implying that a P-value $< 0{\cdot}05$ for a product term ($\text{exposure}_A \times \text{exposure}_B$) implies a departure from pure multiplication of effects. This method, however, disregards the possibility of detecting additive interactions and quantifying the effect attributed to the interaction. Contrary to the common practices in standard articles of epidemiology, according to Rothman, the information provided on the additive scale, including interaction analysis, is most relevant for public health application[31,32]. Therefore, Knol et al. suggest using more extensive methods, including analyses for the single effects of each factor, joint effects for combinations of exposures, stratification, and measures of interaction on multiplicative and additive scales[30].

Additive interaction analysis, on the absolute risk scale, estimates the number of attributable cases due to the combined effect. In the presence of interaction, these cases will either surpass or fall short of the sum of cases due to both exposures separately, suggesting that the excess of cases depends on the extent to which risk factors A (i.e. MedDiet) and B (i.e. PA) occur together in the same individuals. Moreover, relevant to public health, this analysis provides insights towards which subgroup of a population, not necessarily the high-risk subgroup, would observe a greater absolute risk reduction from disease prevention or intervention strategies[25,26,33]. When two independent risk factors are considered well suited to fit an additive model, the presence of biological interaction requires a departure from additivity in the scale of absolute incidence rate differences[25,27]. However, study results in epidemiology are most frequently presented on the relative risk multiplicative scale, which does not directly allow calculating an absolute risk difference. Nevertheless, alternative measures of interaction to the absolute additive model have been available for decades, including the relative excess risk due to interaction (RERI), synergy index of additivity (SI) and attributable proportion (AP) due to interaction[30,31,34]. The null value for RERI and AP is 0 and SI is 1[35]. Derived from the regressions on the multiplicative scale, these measures of interaction on the additive scale indicate the direction, because it can be positive (synergism, beyond the sum of effects) or negative (antagonism, below the sum of effects), as well as the magnitude of the interaction[36].

Reporting interactions on the additive scale is uncommon in standard epidemiological reports. Current explanations as to why interactions may not be reported in greater detail include space constraints, word limits or editorial intervention[29]. For instance, one study included interaction analysis on both scales, employing a cross-product term on the multiplicative scale, and AP, RERI, and SI on the additive scale; however, the authors used brief descriptive statements to report that no interactions were found and the data were not shown[20]. Moreover, interactions on combined lifestyle factors are rarely a primary objective nor an initially intended analysis in most studies[20,28,37]. However, the inclusion of these analyses provides essential information on the potential public health impact and causal structure of combined effects of different relevant exposures[29]. Thus, more research is needed that report data on interactions as part of the primary hypothesis evaluated.

To demonstrate this research gap, we present the findings of a comprehensive review on reported interaction analyses between the MedDiet and PA on all-cause mortality, followed by an original analysis with the proposed methodology for a complete interaction analysis. The comprehensive review included original research that studied the MedDiet in combination with PA on mortality to identify the use of interaction analysis. Although we are not the first to study additive interactions between lifestyle factors, including diet and PA, to our knowledge there is no previous review that has focused on the additive interaction between the MedDiet and PA on all-cause mortality. Following this review, we provide a novel original analysis within a Spanish cohort, the *Seguimiento Universidad de Navarra* (SUN), to prospectively assess the joint association of the MedDiet and PA on all-cause mortality, applying both the multiplicative and additive interaction analyses for its relevance to public health.

Comprehensive review of reported interaction analyses for the Mediterranean diet and physical activity in association with mortality

We searched PubMed database for original observational research articles (in the last 10 years, English, and humans) that studied the combined effect of the MedDiet with PA on all-cause mortality, from the time when Knol and Vanderweele first published recommendations for presenting analyses of effect modification and interaction in 2012[30]. The search strategy and diagram can be found in Supplementary Table S1 and Fig. S1. We first identified the methods employed to assess the combination of diet and PA. Variations among studies included reporting relative risks for: a lifestyle score that included diet and PA items; a lifestyle score and its individual components; diet and a lifestyle score combined; diet and PA combined; or a lifestyle score, its individual components, and combinations of components. Since we were interested in accessing the presence of interaction analysis between the MedDiet and PA, we excluded all studies that did not specifically assess the relative risk for the combination of the MedDiet and PA on mortality[38–42]. After these exclusions, only four articles met the inclusion criteria for assessment (Table 1)[22,43–45].

Three articles studied diet and PA as individual factors on mortality[22,43,44]; meanwhile, one article included PA and diet as components of a lifestyle score and analysed the combined effect of diet and PA as a secondary analysis[45]. Three studies included the sample size and hazard ratios (HR) with 95 % CI for each combination[22,44,45]. The most recent article employed the parametric G-formula to estimate the relative risk associated with hypothetical interventions on the individual and combined effects of the Mediterranean-style diet and PA on all-cause mortality[43]. Three articles assessed the MedDiet and PA in tertiles[22,43,44], whereas the other used dichotomous variables[45]. The diverse cut-off points indicated great heterogeneity for the categorisation of exposures, reinforcing categories are subject to the available data[46]. All four articles presented the combination of the MedDiet and PA as protective factors and observed relative risk reductions on all-cause mortality (Table 1). Graphical representations of the joint effects and measures of interaction varied across studies, including a contingency table, multidimensional histogram and relative risk tables for various combinations of lifestyle factors.

Only one article by Alvarez-Alvarez et al. reported *P*-values for the possible interaction between the MedDiet and PA on all-cause mortality[22]. The two reported measures of interaction in this article were obtained for two different scores of adherence to the MedDiet and were conducted on the multiplicative scale by incorporating an interaction term in the Cox proportional hazards model. Alvarez-Alvarez et al. observed a synergistic, but not significant, multiplicative interaction between a modified Mediterranean diet score and PA ($P_{interaction} = 0{\cdot}580$), as well as between a Mediterranean Diet Adherence Screener (MEDAS) and PA ($P_{interaction} = 0{\cdot}293$). Thus, the interpretation

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Table 1. Summary table of the literature review conducted on the presence of interaction analysis for the combined effect between the MedDiet and PA on all-cause mortality

	Population characteristics (mean follow-up)			Mediterranean diet			Physical activity			Combined			
Author, et al., year		Mean age	SD		HR	95 % CI†		HR	95 % CI†		Risk	95 % CI†	$P_{interaction}$*
As independent factors:													
Williamson et. al., 2019[43]	22 213 healthy middle-aged adults (median 58 years), 62·9 % female, from the Melbourne Collaborative Cohort Study 1995–2011, (median 13·6 years)			Low MDS (T0; 0–3 pts) *v.* high MDS (T2; 6–9 pts)	0·81	0·70, 0·93	Low PA (low activity ≤ 2/week) *v.* high PA (intensive activity ≥ 3/week)	0·71	0·62, 0·81	High + high; (RR)	0·82	0·64, 1·00	
Cárdenas-Fuentes et al., 2018[44]	7356 older adults at high vascular risk, 57·5 % female, from the PREDIMED study 2003–2008, (6·8 years)	67	6·2 years	MEDAS low (T1; < 130 pts) *v.* high (T3; > 119 pts)	0·47	0·37, 0·59	Low leisure-time PA (T1; < 9 pts) *v.* high (T3; > 10·4 pts)	0·64	0·51, 0·81	T3 + T3	0·27	0·19, 0·38	
Alvarez-Alvarez et. al., 2018[22]	19 467 adult, 60·2 % female, from the SUN cohort 1999–2013, (median 10·3 years)	38·2	12·2 years	mMDS low (T1; ≤ 19 pts) *v.* high (T3; 23–30 pts)	0·66	0·46, 0·96	PA score low (T1; ≤ 2 pts) *v.* high (T3; 6–8 pts)	0·48	0·33, 0·71	T3 + T3	0·36	0·19, 0·67	0·580
				MEDAS low (T1; ≤ 5 pts) *v.* high (T3; 7–12 pts)	0·53	0·31, 0·91	PA score low (T1; ≤ 2 pts) *v.* high (T3; 6–8 pts)	0·48	0·33, 0·71	T3 + T3	0·38	0·19, 0·73	0·293
As components of lifestyle scores:													
Behrens et. al., 2013[45]	170 672 men and women, 41 % female, from the NIH-AARP Diet and Health Study 1996–2009, (12·5 years)	62·5	5·3 years	aMDS healthy diet (no; ≤ 4 pts) *v.* (yes; 5–8 pts)	0·86	0·83, 0·88	Recommended PA (no; vigorous activity < 3/week) *v.* (yes; vigorous activity ≥ 3/week)	0·86	0·84, 0·89	yes + yes	0·82	0·79, 0·85	

MedDiet, Mediterranean diet; PA, physical activity; HR, hazard ratio; RR, relative risk; MDS, Mediterranean diet score; pts, points; MEDAS, Mediterranean Diet Adherence Screener; mMDS, modified Mediterranean diet score; aMDS, alternate Mediterranean diet score; PREDIMED, Prevención con Dieta Mediterránea.
† HR provided are the multivariable-adjusted values.
*All interaction analyses reported were presented on the multiplicative scale.

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of the combined effect of both lifestyle factors was the existence of a synergistic interaction, beyond additivity, but not beyond multiplicativity, equivalent to the mere multiplication of relative effects[22].

Despite the absence of an interaction analysis, Williamson et. al presented measures of association on the risk difference (additive) scale and measures of impact, in addition to the relative risk (multiplicative) scale. The combined hypothetical repeated intervention estimated an absolute reduction in all-cause mortality of 1·82 deaths per 100 people (95 % CI 0·03, 3·6). Moreover, when the authors considered an intervention only on participants with obesity, the overall risk differences and risk ratios were closer to the null, suggesting that a greater absolute effect would be obtained by intervening on the general population[43]. A comprehensive interaction analysis would have further addressed the mechanism behind the observed joint effect, which suggested a potential synergism in the Melbourne Collaborative Cohort Study, with 22 213 middle-aged participants. This comprehensive review sheds light on the absence of reported interaction analyses and the research gap that exists between the frequently reported measures of association (i.e. relative risks) and less common absolute measures of association and impact for public health.

Mediterranean diet and physical activity on all-cause mortality in the *Seguimiento Universidad de Navarra* cohort

One of the above-mentioned studies, which assessed the combined effect of the MedDiet and PA on all-cause mortality, was nested in the SUN cohort. The SUN project is a prospective, multipurpose, cohort of Spanish university graduates, with continually open recruitment (i.e. a dynamic design), consisting of a baseline questionnaire and biennial follow-up questionnaires. Participants' informed consent was given upon completion of the baseline questionnaire. All participants are university graduates, ensuring greater reliability, validity and retention rates associated with education status. The SUN project has been approved by the Institutional Review Board of the University of Navarra and registered at clinicaltrials.gov (NCT02669602). Further explanation of this study's objective, design and methods has been published previously[47,48].

In the previous study by Alvarez-Alvarez et. al., a descriptive evaluation of the relation between the MedDiet and PA showed that physically active participants had lower BMI and adhered better to the MedDiet by consuming more vegetables, fruits, legumes, cereals, fish and nuts, but less red and white meat. The authors calculated an eight-item active lifestyle score as a proxy of PA that considered the volume, intensity and frequency of leisure-time physical activities and sedentary behaviour (i.e. exercise, walking, climbing stairs, watching television and sitting). They found that participants who engaged in a more physically active lifestyle (6–8 points) and also presented higher adherence to the modified Mediterranean diet score (23–30 points) showed a 64 % relative reduction (HR = 0·36; 95 % CI 0·19, 0·67) on all-cause mortality compared with participants in the lowest category of PA and MedDiet. In addition, high adherence (7–9 points) to MEDAS in combination with high PA (6–8 points) was associated with a 61 % decreased relative risk of all-cause mortality (HR = 0·39; 95 % CI 0·21, 0·72) compared with the lowest adherence to PA and MedDiet category[22,49]. These observations and joint effects suggested the interrelationship between these two lifestyle factors and the potential interaction that may drive a greater risk reduction on all-cause mortality than the individual effects of diet or PA alone.

Alvarez-Alvarez et al. tested the potential synergism between the MedDiet and PA using a likelihood ratio test, comparing Cox proportional hazards models with and without the interaction product term created by the MedDiet and PA. *P*–values for multiplicative interaction were not statistically significant for neither the modified Mediterranean diet score ($P = 0·580$) nor MEDAS ($P = 0·293$). Therefore, their combined effect was inferred to only have synergistic effects on mortality risk reduction, but not beyond multiplicativity[22]. This interpretation, nonetheless, should be further assessed with appropriate statistical methods. Hence, this initial joint effect analysis provided the foundation for the following comprehensive assessment of interactions in search of a clearer understanding of the nature between these two lifestyle factors which are so frequently combined.

Comprehensive interaction analysis in the *Seguimiento Universidad de Navarra* cohort

From December 1999 to August 2020, a total of 22 893 participants had been recruited for the SUN cohort. After exclusions, a total of 19 446 participants, consisting of 7416 men and 12 030 women (61 %), were included in the present analysis (online Supplementary Fig. S2). Dietary data in the SUN cohort were collected using a validated 136-item semiquantitative FFQ at baseline[50,51]. For our analysis, Trichopoulou's operational definition of the MedDiet, a nine-item Mediterranean diet score (MDS) in which each item scored 0 or 1 point, assessed adherence to the MedDiet[49]. Additionally, a seventeen-item PA questionnaire collected at baseline inquired about the frequency and time dedicated to leisure-time physical activities, sports and sedentary behaviour[52]. PA was measured with an eight-item *a priori* defined index with final scores ranging between 0 and 8 points[22]. PA items included exercise (yes and no), intensity (moderate and vigorous), Metabolic equivalent of task-h/week (< 16·1 and ≥ 16·1), walking speed (low/normal and brisk/fast), walking time (< 0·5 h/d and ≥ 0·5 h/d), climbing upstairs (< 3 floors/d and ≥ 3 floors/d), television viewing time (≥ 1·5 h/d and < 1·5 h/d) and sitting time (≥ 5 h/d and < 5 h/d). These exposures, traditionally presented as protective factors, were transformed and presented as risk factors by recommendation of Knol et al. when conducting interaction analyses on the additive scale[53]. Thus, MDS scores were presented as quartiles (Q4: high adherence to Q1: low adherence) and PA scores were dichotomised into categories of high (4–8 points) and low (0–3 points) activity levels. This categorisation of each exposure identified the most appropriate distribution of individuals with differentiated MedDiet adherence and PA level. Combinations of both exposures were created with a contingency table for quartiles of MDS and dichotomous PA.

After a median follow-up of 12 years (±4·5 SD), a total of 277 deaths (including 9 (3·25 %) deaths with unconfirmed cause) were observed. Deaths were confirmed by death certificates and medical records sent by next of kin or computerised record linkage to the Spanish National Statistics Institute (INE, www.ine.es). The date and cause of death were recorded and encoded using the International Classification of Diseases (ICD-10). Follow-up for each participant was calculated from the date the baseline questionnaire was returned to the date the last questionnaire was received or the reported date of death.

A multivariable statistical analysis was conducted using a Cox regression model for the assessment of individual and combined effects between adherence to the MedDiet and PA on all-cause mortality. Age was the underlying time variable, and all Cox regression models were stratified by age in decades (seven categories) and the year in which participants entered the study (six categories). Multivariable-adjusted HR were adjusted for sex, BMI (five categories), education level (bachelor's degree/masters or doctorate), smoking status (never, active and former smoker), cigarettes smoked (packs/d-year), alcohol consumption (continuous), total energy intake (continuous), family history of CVD, prevalent hypertension, hypercholesterolemia and history of depression at baseline (ever/never). Individual exposures were additionally adjusted for the remaining lifestyle factor. Linear trend tests were performed by assigning medians to each category and treating it as a continuous variable.

Interactions were analysed according to the methodology proposed by Knol and Vanderweele by studying the single and joint effects of the exposures followed by an interaction analysis on both the multiplicative and additive scales[30,54]. Knol et al. made particular emphasis that protective factors should be recoded as risk factors, selecting the reference group as those not exposed to either risk factor, representing the lowest risk on the given outcome, for the correct calculation of RERI[53]. On the multiplicative scale, a likelihood ratio test compared Cox regression models with and without a product term for the lowest MDS and low PA level. On the additive scale, the lowest MDS quartile and low PA category were employed for calculating the RERI, as well as the AP due to interaction (online Supplementary Table S4).

All P-values < 0·05 were considered statistically significant. All statistical analyses were conducted using STATA version 14 (StataCorp).

Understanding how diet and physical activity interact on mortality in the *Seguimiento Universidad de Navarra* cohort

Descriptive baseline characteristics for our final study population are described by means and standard deviation or percentage in Supplementary Table S2. As expected, our final study population from the SUN cohort further demonstrated the interrelatedness between dietary and PA habits. Those with higher levels of PA exhibited slightly higher MDS, greater total daily energy intake with a greater percentage from carbohydrates, higher intakes of fibre, vegetables, fruits, cereals, fish, dairy products and nuts, as well as lower percentage of total energy intake from fat and lower meat consumption compared with participants with a low PA level. On the other hand, those with higher adherence to the MDS showed slightly higher PA scores, more frequent exercise, higher weekly energy expenditure, faster walking pace, more minutes walking per d, climbed more stairs and spent fewer hours sitting per d as compared with participants with lower MDS adherence. Supplementary Table S3 shows the frequency of points awarded to each item of the MDS and PA scores. Statistically significant differences were observed across categories of the opposite lifestyle factor, with the exception of dairy product consumption and monounsaturated to saturated fat ratio between PA levels (P > 0·05). These differences suggest that a greater adherence to the MDS is associated with a greater PA level and vice versa.

The main causes of death included cancer (53·8 %) and CVD (18·4 %) with a mean age at death of 61 years. As shown in Table 2, each protective factor as a continuous variable was associated with a statistically significant decreased risk on all-cause mortality. Additionally, poorer adherence across quartiles of the MDS and a low level of PA showed statistically significant increased risks of mortality compared with the highest MDS adherence quartile and the high PA category, (Q1 HR = 1·70; 95 % CI 1·10, 2·62) (HR = 1·32; 95 % CI 1·02, 1·70), respectively. The joint effect of the lowest MDS adherence with low PA showed an even greater increased risk (HR = 2·31; 95 % CI 1·33, 4·01) compared with the highest MDS and high PA combination (Table 2). As represented in Fig. 1, this joint association showed a linear increasing trend as MDS and PA combinations worsened ($P_{\text{for trend}}$ < 0·001).

The joint effect analysis suggested a potential synergism between the two independent variables. This finding was supported by the comprehensive interaction analysis for the doubly exposed category, which observed a RERI coefficient greater than 0 for the point estimate, although it had wide CI and it was not statistically significant (RERI = 0·46; 95 % CI −0·83, 1·75). An additional analysis to increase statistical power was conducted with continuous risk factors, yet no statistical significance was observed (RERI = 0·21; 95 % CI −0·03, 0·07). Table 2 shows that 36 % of the joint effect was attributed to the interaction, whereas low MDS and low PA accounted for 47 % and 16 %, respectively. Lastly, no multiplicative interaction was detected in this analysis since the comparison of regression models with and without a multiplicative interaction term did not observe statistical significance (P = 0·73). Our results indicated the joint association between the lowest adherence to the MDS (Q1) and low level of PA (0–3 pts) on all–cause mortality most likely involves an interaction beyond additivity, but below multiplicativity.

The potential synergism between the MedDiet and PA, as risk factors for premature mortality, may be explained in part by the complex dynamic balance between energetic intake and energy expenditure, in addition to a wide array of other biological mechanisms[55]. Energy intake exceeding energy needs has been associated with an increased mortality risk[56]. Both a healthy diet and adequate PA maintain body weight and composition through interconnected pathways regulated by the neural and endocrine systems[16]. Moreover, a high-quality diet, represented by higher adherence to the MedDiet, has been associated with benefits regarding lipid oxidation[57], HDL function[58], insulin sensitivity[59], endothelial function[60], inflammation[61,62] and telomere

Table 2. Prevalence, individual and joint effects (HR), and measures of interaction on multiplicative and additive scales between adherence to the MedDiet and PA on all-cause mortality
(Numbers and percentages; hazard ratios and 95 % confidence intervals)

		Deaths (%)		Time at risk		95 % CI	
	n	*n*	%	(person-years)	Multivariable-adjusted HR*	Lower limit	Upper limit
Continuous exposures							
Nine-item MDS	19 446	277	1·42	225 057	0·90	0·84	0·97
Eight-item PA score	19 446	277	1·42	225 057	0·88	0·82	0·94
Individual effects†							
Q4 MDS (7–9 pts)	2179	38	1·74	23 929	1 Ref.		
Q3 MDS (5–6 pts)	6527	110	1·69	74 063	1·66	1·11	2·47
Q2 MDS (4 pts)	3968	58	1·46	46 437	1·59	1·02	2·48
Q1 MDS (0–3 pts)	6772	71	1·05	80 629	1·70	1·10	2·62
High PA (4–8 pts)	12 606	156	1·24	145 869	1 Ref.		
Low PA (0–3 pts)	6840	121	1·77	79 189	1·32	1·02	1·70
4 × 2 Joint effects							
Q4 MDS-high PA	1607	26	1·62	17 617	1 Ref.		
Q3 MDS-high PA	4406	65	1·48	50 122	1·77	1·09	2·89
Q2 MDS-high PA	2526	29	1·15	29 508	1·51	0·86	2·65
Q1 MDS-high PA	4067	36	0·89	48 622	1·82	1·05	3·14
Q4 MDS-low PA	572	12	2·10	6312	1·31	0·63	2·71
Q3 MDS-low PA	2121	45	2·12	23 941	2·16	1·29	3·62
Q2 MDS-low PA	1442	29	2·01	16 929	2·48	1·40	4·39
Q1 MDS-low PA	2705	35	1·29	32 007	2·31	1·33	4·01
Measures of interaction					Estimate		
Multiplicative scale							
Likelihood ratio test					*P* = 0·73		
Additive scale							
Relative excess risk due to interaction					0·46	–0·83	1·75
Attributable proportions of the joint effect							
Due to interaction					0·36	–0·62	1·34
Due to low MDS					0·47	–0·11	1·06
Due to low PA					0·16	–0·53	0·85

HR, hazard ratio; MedDiet, Mediterranean diet; PA, physical activity; *n*, sample population size; MDS, Mediterranean diet score; pts: points.
*Adjusted for sex, BMI, education level, smoking status, cigarettes smoked, alcohol, total energy intake, family history of CVD, prevalent hypertension, hypercholesterolemia, depression, and stratified by year entering the cohort and age in decades. Individual exposures were additionally adjusted for the remaining lifestyle factor.
† Variables are presented as risk factors.

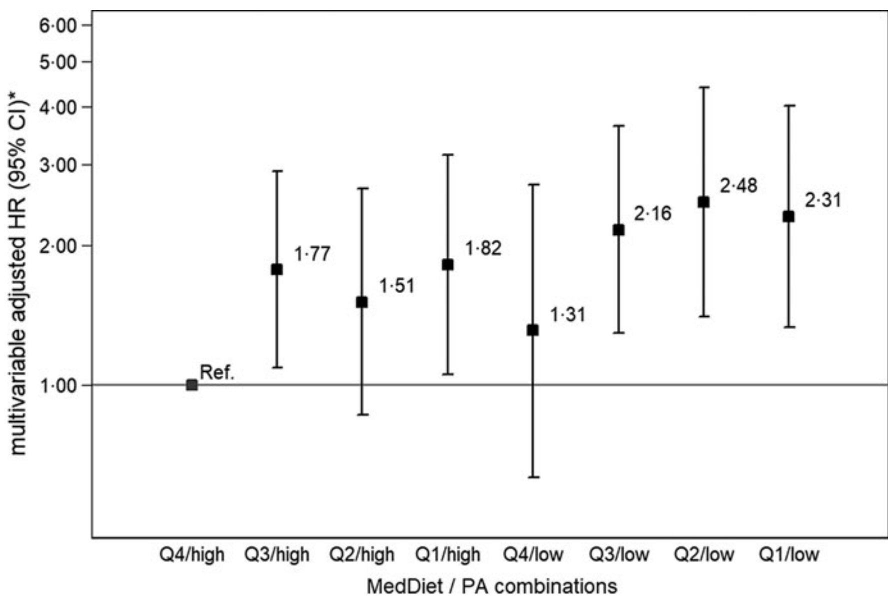

Fig. 1. HR (95 %CI) for the combinations of adherence to the MedDiet and PA levels on all-cause mortality. MedDiet, Mediterranean diet; PA, physical activity; HR, hazard ratios.

length[63], suggesting potential biological mechanisms for a lower risk of mortality[5]. Similarly, regular exercise alleviates the negative effects caused by free radicals, reducing the risk of sarcopenia, insulin resistance, chronic disease, and consequently, premature death[64]. Thus, the detrimental effects of inadequate nutrition and lack of PA, which increase morbidity and mortality from lifestyle diseases, are most likely due to an energy imbalance, the modification or disruption of regulatory processes, and harmful effects caused by inflammation and oxidative stress on health.

Furthermore, the absolute measures presented, RERI and the AP due to interaction, provide informative estimates regarding the impact of the joint effect. The effect of the interaction varies according to the prevalence of the two exposures and the outcome within a given subgroup. Hence, the public health implications of the MedDiet and PA depend on the proportion of the population in which these factors occur jointly[25]. The greater the number of subgroups, the fewer cases of mortality correspond to each combination, the smaller the effect observed from the interaction. According to our data, which observed increased relative risks as lifestyle factor combinations worsened, surpassing the risks of the individual factors, we quantified the effect due to the interaction was 36 % of the total joint effect. Hence, the subgroup with low MedDiet adherence and low PA would benefit from an intervention targeting both habits simultaneously to reduce the risk posed by this synergism. This subgroup received three or less points for both the MDS and PA scores, indicative of individuals with ample room for improvement in many possible aspects of diet and PA compared with the rest of the study population. Similarly, from a more applicable perspective to public health, increasing risk reductions were observed across combinations of MDS and PA on protective scales (online Supplementary Table S5 and Fig. S3). Although quantifying the interaction requires transforming healthy lifestyle factors into their corresponding risk factors, the observed joint effect of adhering to the MedDiet and PA, greater than the sum of the effect of each individual lifestyle habit, offers a more translatable message to the public.

Strengths of this analysis include the large population size, long follow-up, adjustment for numerous potential confounders and greater validity of self-reported data from an educationally homogenous population of university graduates. Nevertheless, considering a multivariable analysis requires a large sample size, an interaction analysis requires an even greater sample size and, therefore, the AP due to interaction may add strength to the RERI estimate. Although the remaining cases of mortality were few after exclusions were applied, a recent meta-analysis showed that the association between a healthy lifestyle and all-cause mortality was stronger in studies with longer follow-up or among younger participants, indicating larger benefits could be obtained if people adopt healthy lifestyles at an early age and follow for a long time[65]. Although we may not have had the sufficient statistical power for a more robust interaction analysis, the power to detect interactions tends to be greater on the additive scale than the multiplicative scale when the main effects are positive[66,67]. In addition, the primary limitations posed by measuring long-term habitual patterns of dietary intake and PA from self-reported measurements include residual confounding due to variations in habits over time and recall bias[68]. Furthermore, our analysis employed baseline data, whereas an analysis with repeated measures may detect associations and interactions between decreased adherence to the MedDiet and PA over time. Lastly, our results should be interpreted with caution due to the use of recoded variables as risk factors (i.e. non-adherence to the MedDiet rather than high adherence and physical inactivity rather than PA), which may not infer the same results for exposures in their preventive form[53]. The categorisation of exposures may be debatable given the irregular distribution of participants; however, the biological relevance and case distribution were considered to present the most appropriate analysis[46].

As previously mentioned, measures of interaction are commonly non-significant and considered unnecessary to authors and therefore are often not presented[29]. One of the main methodological reasons for the absence of statistically significant interaction terms previously acknowledged and the reason for underreporting interaction analyses is the lack of statistical power to detect RERI and reduce type II error[66,67,69]. Both follow-up duration and sample size should be considered when conducting additive interaction analysis, provided that the detection of causal interactions may depend on the progression of time and more precise estimates may require very large study populations[70]. This frequent concern may be solved in part by calculating the AP due to interaction, which does not solely rely on statistical significance, making it a valuable measure of interaction[34]. Furthermore, statistical significance for interactions is frequently established at $P < 0{\cdot}10$, rather than $P < 0{\cdot}05$, due to the limitations of statistical power[71].

Significance of studying interactions between lifestyle factors

Just as the MedDiet and PA have been studied as the combined effect created by their individual components, such as a *priori* defined dietary patterns rather than single food groups or foods, lifestyle can be assessed by studying specific combinations of behaviours[3,9,72,73]. This methodology was anticipated by Rothman, who stated 'as more causal factors are associated with health outcomes, greater interest will be given to the joint effects created by combinations of exposures'[74]. More and more lifestyle scores, including simple scores, Life's Simple 7 (LS7) and the World Cancer Research Fund/American Institute for Cancer Research (WCRF/AICR) score, encompass a healthy dietary pattern complemented by PA, other lifestyle habits and cardiometabolic parameters to define a larger concept of lifestyle[41,65,75–77]. A recent meta-analysis observed the risk reductions for all-cause and CVD mortality related to LS7 were similar or even weaker compared with the simple score, indicating that more emphasis should be given to lifestyle factors, in addition to cardiometabolic markers, for the prevention of premature deaths[65]. In addition to studying the global effect of a lifestyle score, studying combinations of lifestyle factors is relevant for understanding the impact these multifaceted and interrelated habits have on individual and population health. These studies provide key insight for implementing successful multicomponent lifestyle interventions[78–82]. Consequently, studying

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the conjoint effect of diet and PA is especially relevant given the current research gap between the effects of individual factors and the complexity of an overall lifestyle.

Not only the MedDiet and PA but other lifestyle factors as well should be studied in combination with each other to understand the interaction between multifactorial causes of disease and mortality and create effective guidelines for general, at risk, and diseased populations. Translating the findings of an interaction analysis into a public health message, however, is difficult. Future strategies will require educating health professionals on the synergism between lifestyle factors to communicate the synergistic health benefits to patients. There are considerable limitations when asking a dietitian to speak on PA or a PA expert to speak on diet, let alone other lifestyle factors. Thus, clinicians should be specifically trained to discuss lifestyle factors as proposed by Frates et al.[83] This issue is similar at the public health level, we need to better combine the dietary guidelines with PA guidelines in a more integrative manner, such as the Dietary Guidelines for Americans[84] and the Physical Activity Guidelines for Americans[85]. These recommendations must be supported by long-term policies, communication and implementation strategies across sectors[86].

Provided that chronic disease affects all aspects of health and the combination of poor diet and PA may play a greater role in the burden from chronic disease, more so than overall mortality, future research should study cause-specific mortality, premature mortality and death free of chronic disease, including CVD, diabetes, cancer and the metabolic syndrome[87,88]. Research in this line has already been devised and conducted with network analysis (analysis of regularities or patterns of interaction within the network) to understand the multiple connections between associations of healthy ageing. In a similar manner to studying biological interactions, this methodology has been used to focus on the meaning of the interactions between aspects of health and vitality along the path that leads to frailty and its adverse consequences, and how they change over time[89]. Complementary to the presently suggested interaction analysis, network analysis may also contribute to the research gap regarding the pathways involved in interactions with the MedDiet in the field of public health[90].

In conclusion, this article addresses the current research gap regarding interaction analyses reported for the combination of the MedDiet and PA, beyond individual and joint measures of association, and presents an original analysis within the SUN cohort. Our analysis focused on quantifying the interaction between the MedDiet and PA; however, more studies are needed to study other dietary patterns for greater generalisability and a meta-analysis of the effect attributed to the interaction would provide further evidence. Similar to studying an overall dietary pattern as a cumulative effect of several individual components, lifestyle indices are used to study the cumulative effect of individual behaviours. Nevertheless, the mechanism by which these individual components interact is complex, suggesting the use of interaction analysis as an essential statistical method to complement frequently reported joint effects. Our analysis in the SUN cohort suggested a synergism between low adherence to the MDS and low level of PA on all-cause mortality. While quantifying the synergism between the MedDiet and PA focuses on one interaction among many possible lifestyle interactions, this methodology and network analysis may be advantageous towards understanding the potential synergism between multiple lifestyle factors. More studies on interactions are needed to fill this gap in nutritional epidemiology and provide high-quality evidence as interest grows in studying overall lifestyle patterns on health.

Acknowledgements

The authors other members of the SUN Group: Aguilera-Buenosvinos I, Alonso A, Álvarez-Álvarez I, Balaguer A, Barbería-Latasa M, Barrio-López MT, Basterra-Gortari FJ, Battezzati A, Bazal P, Benito S, Bertoli S, Bes-Rastrollo M, Beunza JJ, Buil-Cosiales P, Carlos S, de Irala J, de la Fuente-Arrillaga C, de la O V, de la Rosa PA, Delgado-Rodríguez M, Díaz-Gutiérrez J, Díez Espino J, Domínguez L, Donat-Vargas C, Donazar M, Eguaras S, Fernández-Lázaro CI, Fernández-Montero A, Fresán U, Galbete C, García-Arellano A, Gardeazábal I, Gea A, Gutiérrez-Bedmar M, Gomes-Domingos AL, Gómez-Donoso C, Gómez-Gracia E, Goñi E, Goñi L, Guillén F, Hernández-Hernández A, Hershey MS, Hidalgo-Santamaría M, Hu E, Lahortiga F, Leone A, Llavero M, Llorca J, López del Burgo C, Marí A, Martí A, Martín-Calvo N, Martín-Moreno JM, Martínez JA, Martínez-González MA, Mendonça R, Menéndez C, Molendijk M, Molero P, Muñoz M, Navarro AM, Pano O, Pérez de Ciriza P, Pérez-Cornago A, Pérez de Rojas J, Pimenta AM, Ramallal R, Razquin C, Rico-Campà A, Romanos-Nanclares A, Ruiz L, Ruiz-Canela M, San Julián B, Sánchez D, Sánchez-Bayona R, Sánchez-Tainta A, Sánchez-Villegas A, Santiago S, Sayón-Orea C, Toledo E, Vázquez Z and Zazpe I. We especially thank all participants in the SUN cohort for their long-standing and enthusiastic collaboration and our advisors from Harvard TH Chan School of Public Health Walter Willett, Alberto Ascherio, Frank B. Hu and Meir J. Stampfer who helped us design the SUN project, the Prevención con Dieta Mediterránea (PREDIMED) study and the PREDIMED-PLUS ongoing trial.

The SUN project has received funding from the Spanish Government-Instituto de Salud Carlos III, and the European Regional Development Fund (FEDER) (RD 06/0045, CIBER-OBN, Grants PI10/02658, PI10/02293, PI13/00615, PI14/01668, PI14/01798, PI14/01764, PI17/01795, PI20/00564 and G03/140), the Navarra Regional Government (27/2011, 45/2011, 122/2014), and the University of Navarra. MSH receives financial support from the Programa de Ayudas de la Asociación de Amigos de la Universidad de Navarra.

M. R. C. and J. A. M. formulated the research question, M. A. M. and M. R. C. designed the study, M. S. H., I. A. A., and M. R. C. carried out the study, M. S. H., I. A. A., and M. R. C. analysed the data, M. S. H., M. R. C., and M. A. M. interpreted the findings, M. S. H., M. A. M., and M. R. C. wrote the article, and all authors reviewed and contributed to the final version of the manuscript.

There are no conflicts of interest

Supplementary material

For supplementary material/s referred to in this article, please visit https://doi.org/10.1017/S0007114521002877

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PUBLICATION 2

Hershey MS, Sotos-Prieto M, Ruiz-Canela M, Martínez-González MA, Cassidy A, Moffatt S, & Kales SN. Anthocyanin Intake and Physical Activity: Associations with the Lipid Profile of a US Working Population. *Molecules*. 2020;25(19):4398. doi:10.3390/molecules25194398

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Anthocyanin Intake and Physical Activity: Associations with the Lipid Profile of a US Working Population

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Academic Editors: Luciana Mosca and Paula Silva
Received: 30 July 2020; Accepted: 22 September 2020; Published: 24 September 2020

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Abstract: While growing evidence exists on the independent associations between anthocyanins and physical activity on cardiovascular disease (CVD) risk determinants, the possible interaction between these exposures has not yet been studied. We aimed to study the potential synergism between anthocyanin intake and physical activity on lipid profile measures. This cross-sectional study was conducted among 249 US career firefighters participating in the *Feeding America's Bravest* trial. Anthocyanin intake was calculated using a validated food frequency questionnaire (FFQ) and physical activity level by a validated questionnaire. Multivariable linear regression models determined the extent to which anthocyanin intake and physical activity predicted lipid parameters. Generalized linear models were used for joint effect and interaction analyses on the multiplicative and additive scales. Both anthocyanins and physical activity were independently inversely associated with total cholesterol:high density lipoprotein (HDL) cholesterol. Only physical activity was inversely associated with triglycerides, low density lipoprotein (LDL) cholesterol:HDL, and triglycerides (TG):HDL. Although the combined exposure of low anthocyanin intake and low physical activity was associated with lower (RR = 2.83; 95% CI: 1.42 to 5.67) HDL cholesterol <40 mg/dL, neither multiplicative ($p = 0.72$) nor additive interactions were detected (relative excess risk due to interaction (RERI): 0.02; 95% CI: −1.63 to 1.66; $p = 0.98$). Our findings provide insight on the potential synergism between anthocyanin intake and physical activity on the lipid profile.

Keywords: anthocyanins; physical activity; lipid profile; cardiovascular disease; working population

1. Introduction

The prevalence of low fitness, obesity, and cardiovascular disease (CVD) risk factors, such as hypercholesterolemia, hypertension, and high blood glucose, among United States (US) career firefighters is high [1]. When these risk factors interact with the strenuous physical activity, emotional stress, and environmental pollutants characteristic of the firefighter profession, the risk of CVD events is increased [2]. Sudden cardiac death is the leading cause of on-duty deaths, 82% of which are attributed to underlying coronary heart disease and cardiomegaly/left ventricular hypertrophy; moreover, CVD contributes to important morbidity and disability among this working population [2–5]. Therefore, the primary prevention of CVD through the promotion of healthy dietary patterns and physical activity should be a current priority. Polyphenols in particular play an important role as one of the dietary components associated with the cardioprotective effects of certain foods common across different healthy diets.

Polyphenols are secondary plant metabolites and bioactive compounds naturally occurring in plants and plant-derived products, which can be differentiated into six main classes: flavones, flavonols, flavanols, flavanones, anthocyanins, and flavan-3-ols. Anthocyanins are most abundant in red-, purple-, or blue-pigmented plants, flowers, seeds, fruits, and other plant-based foods. A combination of animal and human studies have attributed cardioprotective effects to anthocyanins, including the inhibition of platelet aggregation, increased HDL, arterial vasorelaxation, and improvement of lipid profile and platelet function [6]. These cardioprotective effects suggest biological pathways in which an anthocyanin-rich diet contributes towards the prevention of CVD. A systematic review of randomized controlled trials (RCTs) suggested anthocyanins may have the potential to influence CVD development and progression among individuals with hyperlipidemia [7]. Among men, a prospective cohort study observed a higher intake of anthocyanins was associated with a 14% lower risk of nonfatal myocardial infarction [8]. Evidence suggests positive potential for the use of anthocyanins in the prevention and treatment of CVD risk factors among firefighters.

Anthocyanins have also been associated with nitric oxide production, exercise performance, and physiological responses before, during, and post-exercise, suggesting their antioxidant, anti-inflammatory, and vasoactive properties improve fitness performance [9,10]. Understanding the intercorrelation and combined effect of dietary bioactive components, such as anthocyanins, with physical activity on CVD risk parameters may contribute towards better prevention and treatment of CVD in high-risk populations. Therefore, we aimed to study the joint effect and the possible synergism between anthocyanin intake and physical activity on the lipid profile of Midwestern US career firefighters.

2. Results

2.1. Study Participants

Among all 249 participants, 95% were male and the mean (±standard deviation (SD)) age was 47 ± 7.6. The average total energy intake among males was 2395 kcal/day, whereas women consumed an average of 1886 kcal/day. The average body mass index (BMI) among participants was 29.8 kg/m^2. The mean (±SD) values for lipid profile measures at baseline were as follows: TG = 126.37 ± 68.57 mg/dL, total cholesterol = 195.40 ± 36.24 mg/dL, HDL cholesterol = 48.50 ± 10.83 mg/dL, LDL cholesterol = 121.85 ± 31.66 mg/dL, LDL:HDL ratio = 2.61 ± 0.81, TG:HDL ratio = 2.90 ± 2.15, and total cholesterol:HDL ratio = 4.18 ± 1.02.

Baseline characteristics of the overall participants (n = 249) from *Feeding America's Bravest* included in this study are presented in Table 1 according to low and high anthocyanin intake and physical activity level. Across both subgroups, high anthocyanin intake and physical activity level observed significantly greater mMDS scores and supplement use than those with low intake or activity. Total flavonoid content, protein intake, whole grains, total fiber, polyunsaturated fat, and alcohol consumption were significantly higher among high anthocyanin intakes. Meanwhile, added sugar intake and prevalent dyslipidemia were significantly lower in the high anthocyanin subgroup. Age, BMI, and hours sitting per week were significantly lower among participants with high physical activity levels.

Table 1. Baseline characteristics according to high and low anthocyanin intake and physical activity level.

	Anthocyanin Intake (SD) [a]			Physical Activity Level [b]		
	Low	High	*p*-Value	Low	High	*p*-Value
N	124	125		95	154	
Women (%)	4.8	5.6	0.79	2.11	7.14	0.08
Age (yrs)	47.2 (7.4)	46.3 (7.5)	0.30	48.7 (6.9)	45.5 (7.6)	0.002
BMI (kg/m^2)	30 (4.5)	29.7 (4.2)	0.59	31.7 (4.4)	28.7 (3.9)	<0.001
Total energy intake (kcal/d)	2244 (941)	2491 (852)	0.03	2423 (993)	2334 (846)	0.45
mMDS † (pts)	22.3 (6.6)	25.3 (6.9)	<0.001	21 (7.6)	25.6 (5.8)	<0.001
Flavonoids (mg/d)	286 (212.0)	460 (282.0)	<0.001	383 (306.0)	367 (238.0)	0.65
Anthocyanins (mg/d)	10.9 (6.0)	53 (40.2)	<0.001	29.7 (35.5)	33.4 (35.7)	0.43
Protein intake (g/d)	97 (43.0)	110 (38.0)	0.01	104 (46.0)	103 (37.0)	0.95
Carbohydrate intake (g/d)	243 (106.0)	267 (99.0)	0.07	262 (106.0)	251 (101.0)	0.40
Whole grains (g/d)	33.2 (18.7)	39.9 (23.2)	0.01	35.7 (17.9)	37 (23.2)	0.64
Total fiber intake (g/d)	21 (8.7)	27.7 (9.2)	<0.001	23.7 (8.8)	24.8 (10.0)	0.39
Added sugar (g/d)	65.2 (48.7)	54.2 (35.2)	0.04	66.1 (47.7)	55.7 (39.0)	0.06
Fat intake (g/d)	93.5 (45.8)	101.4 (39.3)	0.09	101.2 (47.8)	95.1 (39.3)	0.27
Saturated fat (g/d)	31.4 (17.3)	31.8 (12.9)	0.85	33.3 (17.1)	30.6 (13.9)	0.18
Polyunsaturated fat (g/d)	19.4 (9.3)	22 (9.1)	0.02	21.6 (9.9)	20.2 (8.9)	0.27
Monounsaturated fat (g/d)	35.2 (17.6)	39.4 (16.8)	0.05	38.5 (18.8)	36.6 (16.3)	0.39
Alcohol (g/d)	9.1 (12.4)	15.6 (25.5)	0.01	11.2 (18.8)	13.1 (21.2)	0.48
Nondrinkers (%)	18.5	16.8	0.72	17.9	17.5	0.94
Smoking status (%)			0.29			0.54
never	57.3	52.8		54.7	55.2	
current	18.5	14.4		13.7	18.2	
former	24.2	32.8		31.6	26.6	
Education (%)			0.38			0.28
Technical school/some college/associates degree	66.9	61.6		68.4	61.7	
Bachelor's degree or higher	33.1	38.4		31.6	38.3	
Marital status (%)			0.41			0.92
married	79.2	83.4		80	80.5	
single	21.8	17.6		20	19.5	
Multivitamin use (%)	38.7	38.4	0.96	31.6	42.9	0.08
Supplement use (proteins, glutamine, amino acids, etc.) (%)	24.2	39.2	0.01	20.0	39.0	0.002
Sitting (hrs/wk)	19.5 (13.0)	18.7 (16.8)	0.64	22.4 (19.4)	17.1 (11.1)	0.01
TV, computer, and driving (hrs/wk)	8.02 (3.8)	7.7 (4.0)	0.52	8.33 (4.4)	7.56 (3.5)	0.14
Sleep (hrs/d)	6.57 (1.1)	6.38 (0.9)	0.13	6.41 (1.1)	6.51 (1.0)	0.42
Prevalent hypertension (%)	5.65	6.4	0.80	7.37	5.19	0.48
Prevalent dyslipidemia (%)	20.2	7.2	0.003	10.5	15.6	0.26
Prevalent type 2 diabetes (%)	1.61	1.6	0.99	1.05	1.95	0.51

Values are means ± (SD), unless specified as a percentage (%). Percentages may not equal 100 due to rounding. [a] Low intake was defined as standard deviations of anthocyanin intake (mg/d) below the median and high intake above the median. [b] Low activity was defined as levels 0–3 representing those who avoid walking or exertion to >1 hr/wk of modest PA, whereas high activity levels 4–7 indicated running <1 mile/wk or spending <30 min/wk in heavy PA to running over 10 miles/wk or >3 hrs/wk of comparable PA. † An explanation of how this score was developed is shown in Table S3 (Supplementary Materials). BMI: body mass index is weight in kilograms divided by meters squared, d: day, g: grams, hrs: hours, kcal: kilocalories, mg: milligrams, mMDS: modified Mediterranean Diet Score, N: population size, pts: points, TV: television, wk: week.

2.2. Characteristics of Anthocyanin Intake

Blueberries were the richest source of anthocyanins contributing 43% of total anthocyanin intake (Table 2). Raisins and grapes represented the second richest source (15%); however, they showed the least between-person variability. Strawberries, red wine, apples, and pears followed in contribution and descending between-person variability, respectively. The composition of anthocyanin subclasses was as follows: 27% malvidin, 24% cyanidin, 18% delphinidin, 17% pelargonidin, 10% petunidin, and 4% peonidin, according to the above-mentioned anthocyanin-rich sources, which accounted for 88% of total anthocyanins.

Table 2. Top contributors of total anthocyanin intake; source content, between-person variability, and contribution to total anthocyanin intake according to the FFQ items in Feeding America's Bravest trial (2016–2019).

Sources (Serving Size)	mg/Serving	Cumulative R^2	Change in R^2	Contribution (%)
Blueberries (1/2 cup)	120.8	0.961	—	43
Strawberries (1/2 cup)	20.5	0.981	0.020	11
Red wine (5 oz. glass)	28.3	0.989	0.008	11
Apple or pears (1 fresh)	6.05 *	0.994	0.005	8
Raisins or grapes (1 oz or small pack) or (1/2 cup)	36.5	0.997	0.003	15

* anthocyanin content is an average of the anthocyanins in apples (8.4 mg/serv) and pears (3.7 mg/serv). Cumulative R^2 indicates the proportion of variability with the addition of each source. The change in R^2 indicates the between-person variability corresponding to each source.

2.3. *Individual Associations between Anthocyanin Intake and Physical Activity on Lipid Profile*

Figure 1 shows regression coefficients for each independent association of anthocyanin intake and physical activity on lipid concentrations and lipid ratios. Multivariable-adjusted models for anthocyanin intake were inversely associated with total cholesterol:HDL (β = −0.14; 95% CI: −0.27 to −0.01, p = 0.04) (Table S1). Although not statistically significant, inverse associations were observed between anthocyanin intake and TG, total cholesterol, LDL cholesterol, and LDL:HDL and TG:HDL ratios and a positive association was observed with HDL cholesterol. After stratifying by low and high physical activity level in Table 3, the association with HDL cholesterol was stronger among the high physical activity subgroup for the age-, sex-, and energy intake-adjusted model (β = 1.97; 95% CI: 0.25 to 3.69) compared to participants with low physical activity (β = 0.52; 95% CI: −1.51 to 2.55); however, this association was lost in multivariable adjusted models. A clear association for anthocyanin intake on total cholesterol:HDL was initially observed across high physical activity; however, there was no clear difference between low (β = −0.16; 95% CI: −0.37 to 0.06) and high physical activity (β = −0.17; 95% CI: −0.32 to −0.01) in the age-, sex-, and energy intake-adjusted model. Nonetheless, statistical significance was lost after additional adjustments for total energy intake, mMDS, multivitamin use, supplement use, and sleep.

Table S1 shows physical activity was inversely associated with TG (β = −5.54; 95% CI: −10.40 to −0.67, p = 0.03), LDL:HDL ratio (β = −0.09; 95% CI: −0.14 to −0.03, p = 0.005), TG:HDL ratio (β = −0.18; 95% CI: −0.33 to −0.03, p = 0.02), and total cholesterol:HDL ratio (β = −0.10; 95% CI: −0.18 to −0.03, p = 0.005) after multivariable adjustments. Although not statistically significant, we observed an inverse association of physical activity with total cholesterol and LDL cholesterol; meanwhile, a non-significant positive association was observed with HDL cholesterol. Physical activity stratified by anthocyanin intake in Table 4 shows physical activity was associated with LDL:HDL among low anthocyanin intake (β = −0.10; 95% CI: −0.17 to −0.03) and high anthocyanin intake (β = −0.11; 95% CI: −0.19 to −0.03) in the least adjusted model. Similarly, the total cholesterol:HDL ratio among high anthocyanin intake (β = −0.14; 95% CI: −0.23 to −0.04) and low anthocyanin intake (β = −0.14; 95% CI: −0.23 to −0.05) observed similar associations when adjusted for age, sex, and energy intake. Overall, the associations observed in the stratified analysis were consistent across subgroups, suggesting the effect of physical activity was independent of anthocyanin intake. Sensitivity analysis for both exposures with additional exclusions for chronic disease, women, and supplement use further supported the robustness of our findings (Table S4).

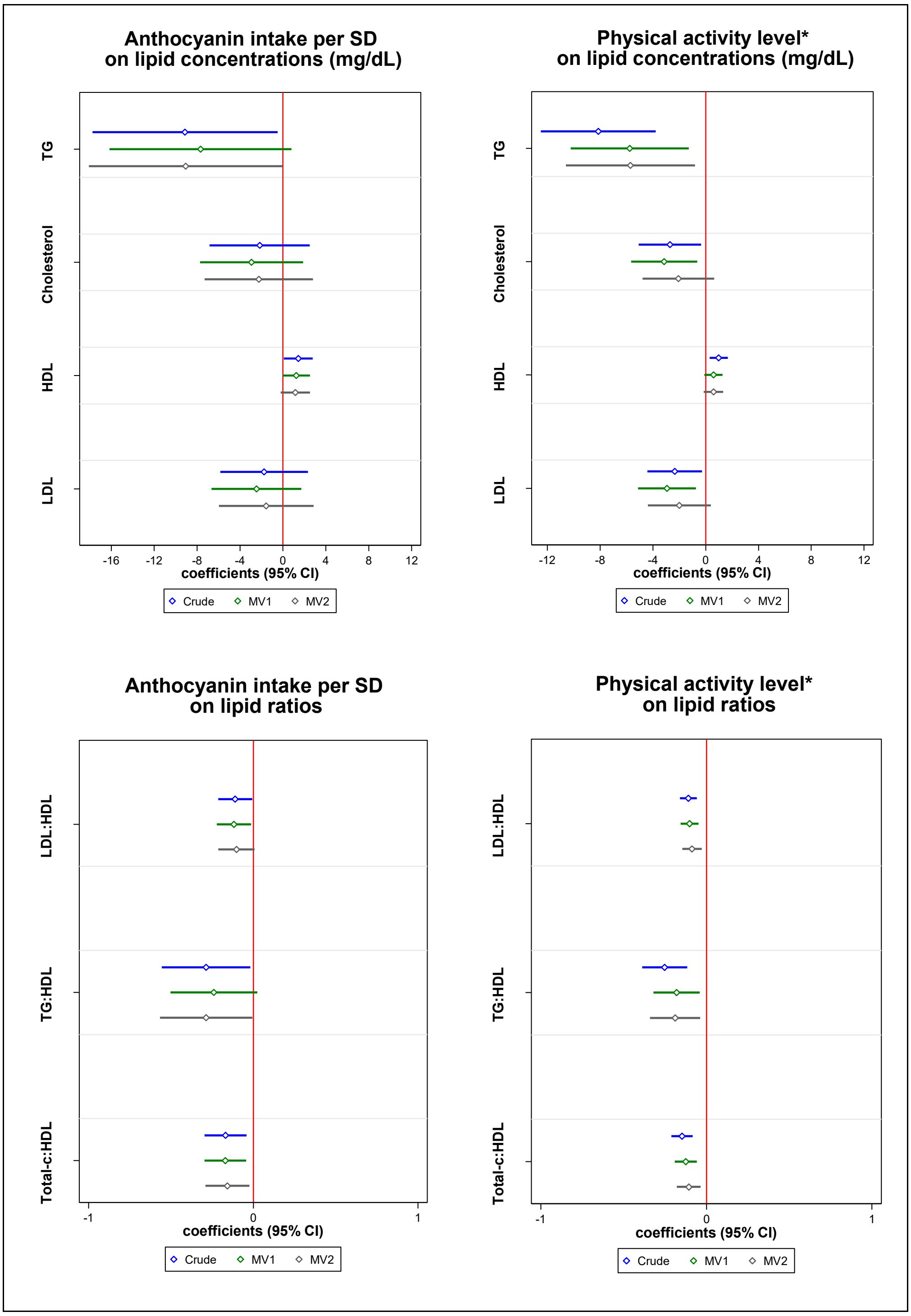

Figure 1. Independent associations (β, 95% CI) of anthocyanin intake (SD) and physical activity level with lipid concentrations (mg/dL) and lipid ratios. * Physical activity was assessed using a scale of 0–7 representing levels of physical activity ranging from none to running > 10 miles/wk or spending > 3 hrs/wk in comparable physical activity (Table S2). MV1: multivariable model 1 adjusted for age, sex, BMI, smoking status, education level, marital status, prevalent hypertension, dyslipidemia, and type 2 diabetes. MV2: multivariable model 2 adjusted for age, sex, BMI, total energy intake, mMDS, smoking status, education level, marital status, multivitamin use, supplement use, sleep, prevalent hypertension, dyslipidemia, and type 2 diabetes. CI: confidence intervals, HDL: high density lipoprotein cholesterol, LDL: low density lipoprotein cholesterol, SD: standard deviation, TG: triglycerides, Total-c: total cholesterol. Table S1 shows β coefficients (95% CI) and *p*-vales of independent associations for all linear regression models.

Table 3. Association (β, 95% CI) between anthocyanin intake (independent variable in SD units) and different lipid parameters (mg/dL) stratified by subgroups of low and high physical activity level.

Lipid Profile	Anthocyanin Intake (Per SD)	
	Low PA * (n = 95)	**High PA * (n = 154)**
Triglycerides		
Age, sex, and energy adjusted model (95% CI)	−8.47 (−22.38 to 5.45)	−8.95 (−19.64 to 1.74)
Multivariable adjusted model 1 (95% CI) [a]	−7.47 (−23.34 to 8.40)	−7.83 (−18.21 to 2.56)
Multivariable adjusted model 2 (95% CI) [b]	−11.29 (−27.19 to 4.61)	−6.57 (−18.32 to 5.19)
Total cholesterol		
Age, sex, and energy adjusted model (95% CI)	−3.75 (−11.07 to 3.56)	−0.97 (−6.94 to 5.00)
Multivariable adjusted model 1 (95% CI) [a]	−2.34 (−10.62 to 5.95)	−1.94 (−7.97 to 4.09)
Multivariable adjusted model 2 (95% CI) [b]	−4.38 (−12.74 to 3.97)	2.80 (−3.79 to 9.39)
HDL cholesterol		
Age, sex, and energy adjusted model (95% CI)	0.52 (−1.51 to 2.55)	1.97 (0.25 to 3.69)
Multivariable adjusted model 1 (95% CI) [a]	1.09 (−1.08 to 3.25)	1.55 (−0.07 to 3.17)
Multivariable adjusted model 2 (95% CI) [b]	0.83 (−1.39 to 3.05)	1.73 (−0.10 to 3.57)
LDL cholesterol		
Age, sex, and energy adjusted model (95% CI)	−2.16 (−8.61 to 4.30)	−1.31 (−6.59 to 3.97)
Multivariable adjusted model 1 (95% CI) [a]	−1.39 (−8.66 to 5.87)	−2.09 (−7.47 to 3.29)
Multivariable adjusted model 2 (95% CI) [b]	−2.66 (−10.17 to 4.86)	2.16 (−3.72 to 8.04)
LDL:HDL		
Age, sex, and energy adjusted model (95% CI)	−0.10 (−0.28 to 0.08)	−0.11 (−0.23 to 0.01
Multivariable adjusted model 1 (95% CI) [a]	−0.12 (−0.32 to 0.08)	−0.11 (−0.23 to 0.02)
Multivariable adjusted model 2 (95% CI) [b]	−0.13 (−0.33 to 0.07)	−0.04 (−0.18 to 0.09)
TG:HDL		
Age, sex, and energy adjusted model (95% CI)	−0.22 (−0.69 to 0.25)	−0.31 (−0.62 to 0.00)
Multivariable adjusted model 1 (95% CI) [a]	−0.23 (−0.75 to 0.30)	−0.27 (−0.57 to 0.03)
Multivariable adjusted model 2 (95% CI) [b]	−0.31 (−0.84 to 0.22)	−0.28 (−0.62 to 0.07)
Total cholesterol:HDL		
Age, sex, and energy adjusted model (95% CI)	−0.16 (−0.37 to 0.06)	−0.17 (−0.32 to −0.01)
Multivariable adjusted model 1 (95% CI) [a]	−019 (−0.42 to 0.05)	−0.15 (−0.30 to 0.00)
Multivariable adjusted model 2 (95% CI) [b]	−0.21 (−0.44 to 0.03)	−0.09 (−0.26 to 0.08)

Coefficients for triglycerides, total cholesterol, HDL and LDL cholesterol show the strength of the effect on mg/dL of lipid concentration per SD of anthocyanin intake, whereas coefficients for LDL:HDL, TG:HDL, and total cholesterol:HDL show the effect on the ratio of lipid concentrations per SD of anthocyanin intake. * Low PA ranged from none to regular recreation or work requiring modest physical activity, such as golf, horseback riding, calisthenics, gymnastics, table tennis, bowling, weight lifting, yard work (levels 0–3); High PA was defined as regularly participating in heavy physical exercise such as running or jogging, swimming, cycling, rowing, skipping rope, running in place or engaging in vigorous aerobic activity such as tennis, basketball, or handball (levels 4–7). [a] Adjusted for age, sex, BMI, smoking status, education level, marital status, prevalent hypertension, dyslipidemia, and type 2 diabetes. [b] Adjusted for age, sex, BMI, total energy intake, mMDS, smoking status, education level, marital status, multivitamin use, supplement use, sleep, prevalent hypertension, dyslipidemia, and type 2 diabetes. CI: confidence intervals, HDL: high density lipoprotein cholesterol, LDL: low density lipoprotein cholesterol, PA: physical activity, SD: standard deviation, TG: triglycerides.

2.4. The Combined Effect, Stratification, and Interaction Analyses between Anthocyanin Intake and Physical Activity on HDL Cholesterol <40 mg/dL

Table 5 shows the four combined effects created between low and high anthocyanin intake and physical activity on HDL cholesterol <40 mg/dL. The combined effects were as follows: low anthocyanins/high activity (RR = 1.46; 95% CI: 0.68 to 3.11, $p = 0.33$), high anthocyanins/low activity (RR = 2.36; 95% CI: 1.15 to 4.83, $p = 0.02$), and low anthocyanins/low activity (RR = 2.83; 95% CI: 1.42 to 5.67, $p = 0.003$). Consistent with the prevalence observed within each subgroup, low anthocyanin intake in combination with low physical activity was associated with the highest relative risk. Stratification analysis observed significant associations between physical activity among those with high anthocyanin intake (RR = 2.19; 95% CI: 1.07 to 4.49, p = 0.03) and low anthocyanin intake (RR = 1.99; 95% CI: 1.10 to 3.60, p = 0.02). Although neither interactions on the multiplicative scale (p = 0.72) nor the relative risk due to interaction on the additive scale (RERI: 0.02; 95% CI: −1.63 to 1.66; p = 0.98) were statistically significant, the joint effect analysis suggested the combination of low anthocyanin intake and low physical activity was associated with the greatest risk compared to the individual effects of the two exposures.

Table 4. Association (β, 95% CI) between physical activity (independent variable in level units) and different lipid parameters (mg/dL) stratified by subgroups of low and high anthocyanin intake.

Lipid Profile	Physical Activity Level (Per Unit)	
	Low Anthocyanin Intake * (n = 124)	**High Anthocyanin Intake * (n = 125)**
Triglycerides		
Age, sex, and energy adjusted model (95% CI)	−6.19 (−13.03 to 0.65)	−8.88 (−14.53 to −3.23)
Multivariable adjusted model 1 (95% CI) [a]	−4.33 (−11.34 to 2.68)	−5.34 (−11.50 to 0.81)
Multivariable adjusted model 2 (95% CI) [b]	−4.96 (−12.45 to 2.53)	−4.79 (−11.62 to 2.04)
Total Cholesterol		
Age, sex, and energy adjusted model (95% CI)	−2.69 (−6.01 to 0.64)	2.70 (−6.21 to 0.82)
Multivariable adjusted model 1 (95% CI) [a]	−3.39 (−6.93 to 0.15)	−2.79 (−6.75 to 1.16)
Multivariable adjusted model 2 (95% CI) [b]	−2.86 (−6.65 to 0.93)	−0.65 (−4.99 to 3.69)
HDL Cholesterol		
Age, sex, and energy adjusted model (95% CI)	0.82 (−0.15 to 1.80)	0.86 (−0.11 to 1.83)
Multivariable adjusted model 1 (95% CI) [a]	0.73 (−0.29 to 1.75)	0.11 (−0.89 to 1.11)
Multivariable adjusted model 2 (95% CI) [b]	0.65 (−0.44 to 1.74)	0.35 (−0.77 to 1.46)
LDL Cholesterol		
Age, sex, and energy adjusted model (95% CI)	−2.24 (−5.08 to 0.60)	−2.47 (−5.68 to 0.74)
Multivariable adjusted model 1 (95% CI) [a]	−3.34 (−6.28 to −0.20)	−2.63 (−6.25 to 0.98)
Multivariable adjusted model 2 (95% CI) [b]	−2.56 (−5.81 to 0.68)	−0.99 (−4.98 to 3.01)
LDL:HDL		
Age, sex, and energy adjusted model (95% CI)	−0.10 (−0.17 to −0.03)	−0.11 (−0.19 to −0.03)
Multivariable adjusted model 1 (95% CI) [a]	−0.12 (−0.19 to −0.04)	−0.08 (−0.17 to 0.01)
Multivariable adjusted model 2 (95% CI) [b]	−0.10 (−0.18 to −0.02)	−0.06 (−0.16 to 0.04)
TG:HDL		
Age, sex, and energy adjusted model (95% CI)	−0.18 (−0.38 to 0.02)	−0.29 (−0.48 to −0.09)
Multivariable adjusted model 1 (95% CI) [a]	−0.14 (−3.35 to 0.07)	−0.17 (−0.38 to 0.035)
Multivariable adjusted model 2 (95% CI) [b]	−0.15 (−0.37 to 0.06)	−0.18 (−0.41 to 0.05)
Total cholesterol:HDL		
Age, sex, and energy adjusted model (95% CI)	−0.14 (−0.23 to −0.05)	−0.14 (−0.23 to −0.04)
Multivariable adjusted model 1 (95% CI) [a]	−0.15 (−0.24 to −0.05)	−0.08 (−0.18 to 0.02)
Multivariable adjusted model 2 (95% CI) [b]	−0.13 (−0.23 to −0.03)	−0.06 (−0.18 to 0.05)

Coefficients for triglycerides, total cholesterol, HDL and LDL cholesterol show the strength of the effect on mg/dL of lipid concentration per unit of physical activity level, whereas coefficients for LDL:HDL, TG:HDL, and total cholesterol:HDL show the effect on the ratio of lipid concentrations per unit of physical activity level. * High and low anthocyanin intake were defined as standard deviations of anthocyanin intake above or below the median, respectively. [a] Adjusted for age, sex, BMI, smoking status, education level, marital status, prevalent hypertension, dyslipidemia, and type 2 diabetes. [b] Adjusted for age, sex, BMI, total energy intake, mMDS, smoking status, education level, marital status, multivitamin use, supplement use, sleep, prevalent hypertension, dyslipidemia, and type 2 diabetes. CI: confidence intervals, HDL: high density lipoprotein cholesterol, LDL: low density lipoprotein cholesterol, TG: triglycerides.

Table 5. Prevalence, joint effect relative risk (RR), stratification, and multiplicative and additive interactions of anthocyanin intake and physical activity on HDL cholesterol <40 mg/dL.

HDL Cholesterol < 40 mg/dL		Anthocyanin Intake (SD)				RR (95% CI) [c] for Anthocyanin Intake Stratified by Physical Activity
		High [a]		Low [b]		
		Prevalence (%)	RR (95% CI)[c]	Prevalence (%)	RR (95% CI) [c]	
Physical Activity	High [a]	12.66	1 Ref.	18.67	1.46 (0.68 to 3.11); $p = 0.33$	1.46 (0.67 to 3.18); $p = 0.34$
	Low [b]	32.61	2.36 (1.15 to 4.83); $p = 0.02$	38.78	2.83 (1.42 to 5.67); $p = 0.003$	1.21 (0.70 to 2.08); $p = 0.50$
RR (95% CI) [d] for physical activity stratified by anthocyanin intake			2.19 (1.07 to 4.49); $p = 0.03$		1.99 (1.10 to 3.60); $p = 0.02$	
Measure of interaction on multiplicative scale: Ratio of RR (95% CI)					$p = 0.72$	
Measure of interaction on additive scale: RERI (95% CI)					0.02 (−1.63 to 1.66); $p = 0.98$	

Regressions are adjusted for age, sex, and energy intake. [a] High anthocyanin intake: standard deviations above the median anthocyanin intake; High PA: regularly participating in heavy physical exercise such as running or jogging, swimming, cycling, etc. or engaging in vigorous aerobic activity such as tennis, basketball, or handball (levels 4–7). [b] Low anthocyanin intake: standard deviations below the median anthocyanin intake; Low PA: none to regular recreation or work requiring modest physical activity, such as golf, horseback riding, calisthenics, gymnastics, table tennis, bowling, weightlifting, yard work (levels 0–3). [c] Relative risks represent low vs high anthocyanin intake stratified by physical activity level. [d] Relative risks represent low vs high physical activity stratified by anthocyanin intake. CI: confidence intervals, HDL: high density lipoprotein, RERI: relative excess risk due to interaction, RR: relative risk.

3. Discussion

To our knowledge, this is the first study to assess both individual and joint exposures of anthocyanin intake and physical activity on lipid profile measures and test for multiplicative and additive measures of interaction. Independently, anthocyanins and physical activity were both inversely associated with total cholesterol:HDL, whereas only physical activity was inversely associated with triglycerides, LDL:HDL, and TG:HDL. Furthermore, the combined effect of a low anthocyanin intake and low physical activity more than doubled the relative risk of having HDL cholesterol <40 mg/dL, compared to the reference high anthocyanins/high activity joint exposure, although no statistically significant interaction was observed.

Berries, in particular, have been studied for their rich anthocyanin content; meanwhile, other studies have used anthocyanin supplementation or derived total anthocyanin intake from the habitual diet [11–15]. Some evidence suggests that the effect of polyphenols may depend on the form in which they are consumed in such a way that supplements may not offer the same synergistic effects and health benefits as food sources [16]. Our top five sources of anthocyanin-rich foods included blueberries, strawberries, red wine, apple or pears, and raisins or grapes, which accounted for 88% of total anthocyanin intake. The remaining 12% can be attributed to other sources, including peaches or plums, apricots, bananas, cantaloupe, and prunes, which contributed small amounts to the total anthocyanin intake in our study. Additional sources of anthocyanins, which were not collected in the study FFQ, include some red to purplish blue-colored vegetables and grains, such as purple corn, purple sweet potato, red cabbage, black carrot, black soybean, and some varieties of rice [17]. Our analysis on anthocyanin intake suggested the greatest between-person variability of anthocyanin intake was associated with the consumption of blueberries and strawberries, most likely due to the seasonality and affordability aspects of purchasing berries.

A variety of scientific studies, including in vitro studies, animal models, and human clinical trials, show that anthocyanins possess anti-inflammatory and antimicrobial activities, which improve cardiometabolic, visual, and neurological health. These protective effects have been explained by participation in different mechanisms and pathways, including the free-radical scavenging pathway, cyclooxygenase pathway, mitogen-activated protein kinase pathway, and inflammatory cytokines signaling, as well as some crucial cellular processes, including the cell cycle, apoptosis, autophagy, and biochemical metabolism [17,18]. Protective effects of anthocyanin intake previously associated with CVD risk determinants further support our findings. A meta-analysis of RCTs using purified anthocyanins or anthocyanins-rich foods as treatment compared with a placebo or nonexposed controls observed anthocyanins significantly reduced total cholesterol (standardized mean difference (SMD): −0.33; 95% CI: −0.62, −0.03; I2 = 86.9%), and LDL cholesterol (SMD: −0.35; 95% CI: −0.66, −0.05; I2 = 85.2%) in regards to the lipid profile [19]. A more recent meta-analysis of 45 randomized controlled trials stated that the consumption of berries and purified anthocyanins (2.2−1230 mg anthocyanins/day) significantly increased HDL cholesterol and reduced total cholesterol, LDL cholesterol, and TGs [14]. Similar to our findings for anthocyanin intake, which lost statistical significance in multivariable models, a systematic review noted that most of the potential effects observed across RCTs were nonsignificant. However, improvement of biomarkers were consistent across studies, particularly in those with elevated lipids at baseline [7].

Physical activity showed clear inverse associations with triglycerides, LDL:HDL, TG:HDL, and total cholesterol:HDL when measured with an ordinal scale capturing habitual weekly frequency and intensity of physical activity and exercise performed. Existing evidence supports aerobic exercise of adequate intensity, duration, and volume results in favorable and independent improvements of blood lipids and lipoproteins in individuals with and without dyslipidemia. Furthermore, the most consistent findings are shown for increases in HDL cholesterol [20–22]. A randomized controlled trial suggested improvements in lipids and lipoproteins were related to the amount of activity and not to the intensity of exercise or improvement in fitness [23]. In particular, a relatively high amount of regular exercise (equivalent to jogging 27.2 to 28.8 km/week at moderate pace) significantly improved the overall lipoprotein profile by decreasing LDL size, increasing HDL cholesterol concentration and size, and decreasing triglycerides, which was not observed for lower amounts of exercise [23]. Therefore, regular physical activity should be strongly promoted within lifestyle interventions for the prevention and treatment of dyslipidemia.

The US Preventive Services Task Force's (USPSTF) identifies dyslipidemia as a CVD risk factor, defined as LDL cholesterol >130 mg/dL or HDL cholesterol <40 mg/dL. Therefore, driven by our initial stratification analysis that suggested independent associations on HDL cholesterol, we studied the joint effect between anthocyanin intake and physical activity on HDL cholesterol <40 mg/dL followed by stratification and interaction analyses. While the stratification analysis tested for the possible effect modification of one factor on the causal pathway of another factor, the interaction analysis tested the potential synergism between two independent causal pathways to produce an effect greater than the sum or multiplication of the two individual effects. The rationale for studying interactions is to better understand which two exposures share inter-related biological mechanisms that create an observable synergistic effect, identifying to whom would an intervention be most advantageous [24]. For the correct calculation and interpretation of RERI, the combinations of anthocyanin intake and physical activity were created by presenting the variables as risk factors and the reference category determined as the category with the lowest risk when considered jointly [25]. The double exposed category for low anthocyanin intake and low physical activity showed the strongest association with HDL cholesterol <40 mg/dL; however, neither measures of multiplicative nor additive interaction were statistically significant.

Although our analysis lacked statistical significance, our results suggest a possible joint effect greater than the sum of the individual exposures most likely due to shared underlying mechanisms. A recent intervention in healthy adult males suggested anthocyanin intake duration affects metabolic responses, including fat and carbohydrate oxidation, during moderate-intensity walking exercise. This may be attributed to an enhanced bioavailability of anthocyanins-derived metabolites involved in mechanisms of oxidation during physical activity [26]. The highest concentrations of dietary anthocyanins derive from elderberries, chokeberries, bilberries, raspberries, black currants, blackberries, and blueberries, among others. However, currently, little is known on the bioavailability of anthocyanins and the concentration in these foods may vary greatly due to influences, such as genetic, environmental, and agronomic factors, including light, temperature, humidity, fertilization, food processing, and storage conditions [27]. Future randomized controlled trials should consider a combination of assessing dietary anthocyanin intake by a validated FFQ combined with biomarker assessment.

Limitations of this study include a possible misclassification bias, due to the self-reported nature of the data and unknown anthocyanin supplementation among participants. Although it is uncertain whether supplementation use would over or underestimate the effect of anthocyanin-rich foods, evidence suggests anthocyanin supplementation improves antioxidative and anti-inflammatory capacity in a dose–response manner and a greater effect would be observed for both subgroups with low and high intakes of anthocyanin-rich foods [13]. Nevertheless, the probable use of anthocyanin supplementation is very low considering these supplements were not indicated in the open-ended question on the type of supplements habitually consumed. Although physical activity was reported using a validated questionnaire, self-reported physical activity has been demonstrated to be less predictive of CVD risk than objective accelerometer measurements [22,23]. Likewise, the validated semiquantitative FFQ did not include all common food sources of anthocyanins and some items captured more than one food with varying anthocyanin content [28]. Due to the predominately male prevalence of the firefighter profession, our results must be extrapolated to women with precaution. Moreover, this study population of Midwestern US career firefighters is not representative of the general population; however, biological plausibility should be the basis for generalizations in epidemiology [29,30]. In addition, because of the cross-sectional design of this study, we cannot infer causality from our results but rather can generate viable hypotheses for future studies.

Although we studied the specific effect of anthocyanin intake, this plant-based bioactive compound is consumed within the bigger context of diet. It is the synergy between foods and nutrients that defines diet quality; therefore, our findings should be considered within the context of multiple possible pathways by which an inappropriate diet could lead to the development of CVD [31]. Although multivariable-adjusted models controlled extensively for potential lifestyle predictors of lipid parameters, including diet; measured with a previously validated mMDS score, residual confounding cannot be completely eliminated [32]. Nonetheless, an appropriate diet, such as the Mediterranean diet, rich in polyphenols through the frequent and abundant consumption of fruits, vegetables, wine, and extra virgin olive oil, may be a practical recommendation for achieving a high anthocyanin intake [31].

Considering multivariable analyses require a large sample size, interaction analyses require an even greater sample size. Nevertheless, the power to detect interactions tends to be greater on the additive scale than the multiplicative scale [33,34]. Due to our relatively small sample size, the statistical power is a limitation of our study; nonetheless, the joint effect and stratification analysis still offer substantial insight on the effect modification and potential interaction between the given exposures. Future studies with a larger sample size and longitudinal design are warranted to further study this hypothesis, while limiting the possibility of reverse causality [33]. In line with our findings across a diverse selection of lipid parameters, future studies may also consider focusing specifically on measurements of total cholesterol and HDL cholesterol, which have proven sufficient to capture the lipid-associated risk in CVD prediction [35].

The USPSTF currently recommends offering adults who are overweight or obese and have additional CVD risk factors intensive behavioral counseling interventions to promote a healthful diet and physical activity; meanwhile, evidence among those without known risk factors suggests a positive but small benefit for the prevention of CVD [36,37]. In regards to the promotion of anthocyanin-rich diets for healthy lipid profiles, previous studies have demonstrated berries, as main sources of anthocyanins, have greater effects on lipid concentrations in obese/overweight individuals (BMI >25 kg/m^2), individuals with cardiovascular risk factors, those ≥50 years, or who have metabolic syndrome compared to healthy individuals [11,14,38]. Future research in this line conducted among a large representative population with the inclusion of additive interaction analysis could be instrumental for identifying narrower CVD risk subgroups and offer greater efficacy among behavior change interventions for the promotion of ideal cardiovascular health.

4. Materials and Methods

4.1. Study Population

Feeding America's Bravest is a cluster-randomized diet intervention trial that included 44 fire stations from the Indianapolis Fire Department (IFD) and 6 fire stations from Fishers (IN) Fire Department. The primary objective of this RCT was to compare a Mediterranean Diet Nutritional Intervention (MDNI) with multiple behavior change strategies: diet/lifestyle education, discounted access to key Mediterranean diet foods, electronic education platforms and reminders, with a Midwestern-style diet or "usual care" group with a cross-over study design over a 2-year period.

Although 486 persons were enrolled, only 265 participants completed the baseline lifestyle questionnaire between November 28, 2016 and April 16, 2018; participants with missing FFQ or biochemical assessment (n = 3) and participants whose energy intake exceeded predefined levels (men: 800–5000 kcal/d, women: 500–3500 kcal/d) (n = 13) were excluded, leaving 249 participants for evaluation.

Full informed consent was received from participants who met the eligibility criteria at the time of enrollment. In accordance with the Declaration of Helsinki, all potential participants were informed of their right to refuse to participate or to withdraw from the study at any time without retribution. The study protocol was approved by the Harvard Institutional Review Board (IRB16–10170) and is registered at Clinical Trials (NCT029441757) [39]. More details on this study's objective, design, and methods have been previously published elsewhere [40].

4.2. Dietary Assessment

Dietary intake was assessed at baseline using a validated 131-item semi-quantitative FFQ, which reflected the previous year's habitual intake, and a lifestyle questionnaire with additional dietary information, including a 13-item modified Mediterranean diet score (mMDS) [28,41]. Flavonoid subclasses were calculated as the habitual daily intake (mg/day), estimated using the US Department of Agriculture (USDA) flavonoid content of foods database, according to previously described methods [42,43]. Commonly consumed US dietary sources of anthocyanins include fruits, such as berries, blackcurrants, red grapes, plums, and cherries, as well as red wine, fruit juices, and some vegetables, such as radishes [44,45]. Anthocyanins are further classified into six subclasses: pelargonidin, cyanidin, delphinidin, peonidin, petunidin, and malvidin.

4.3. Physical Activity

Physical activity level was collected at baseline using a validated physical activity questionnaire administered within the lifestyle questionnaire [46]. On a scale from 0–7, participants were asked to identify the statement which option best described their habitual level of physical activity over the past month: (0) Avoid walking or exertion (e.g., always use elevator, drive whenever possible instead of walking, biking, or rollerblading); (1) walk for pleasure, routinely use stairs, occasionally exercise sufficiently to cause heavy breathing or perspiration; (2) 10 to 60 min per week; (3) over one hour per week; (4) run less than 1 mile per week or spend less than 30 min per week in comparable physical activity; (5) run 1 to 5 miles per week or spend 30 to 60 min per week in comparable physical activity; (6) run 5 to 10 miles per week or spend 1 to 3 h per week in comparable physical activity; and (7) run over 10 miles per week or spend over 3 h per week in comparable physical activity (Table S2).

4.4. Outcome Assessment

Baseline lipid panels were collected in participants' biochemical assessments from the fire department medical examinations at Public Safety Medical (PSM) clinics. Blood samples were collected after an overnight fast at baseline and at follow-up. Plasma and serum were collected in 15-mL specific tubes and were aliquoted, frozen at −80 °C, and stored. Blood lipid profiles were determined using standardized automated high-throughput enzymatic analyses, which achieved coefficients of variation of ≤3% for cholesterol and ≤5% for triglycerides, using a cholesterol assay kit and reagents Ref:7D62–21 and triglyceride assay kit and reagents Ref:7D74–21 by ARCHITECT c System, Abbott Laboratories, IL, USA. Baseline measures were gathered from the PSM electronic medical record database within the last year from enrollment in the study. The primary outcomes of this study were lipid profile measures, specifically TGs, total cholesterol, HDL cholesterol, LDL cholesterol, and ratios for LDL:HDL, TG:HDL, and total cholesterol:HDL.

4.5. Covariate Assessment

Information on sociodemographic characteristics, dietary intake, lifestyle habits, anthropometric measurements, and medical history were collected at baseline through in-person data collection, an online lifestyle questionnaire, or medical record after informed consent was given. BMI was calculated by dividing weight by height squared (kg/m^2). Energy intake was calculated using the baseline FFQ. The mMDS score, described in Table S3, was replicated based on Yang et al. and Sotos-Prieto et al. [28,41]. Participants with dyslipidemia, hypertension, or type 2 diabetes were identified if they had a previous diagnosis of these conditions or were being treated with lipid-lowering, antihypertensive, or antidiabetic medications, respectively, within the previous year to enrollment.

4.6. Statistical Analysis

A continuous variable for total anthocyanin intake was transformed into units of SDs using the standardization method to obtain a normal distribution. In addition, a dichotomous variable of total anthocyanins was created to define high and low anthocyanin intake using the median as the cut-off point; the median intake was equivalent to 19.14 mg/day. Physical activity was used as a continuous variable for each unit (level) increase. As a dichotomous variable, high physical activity was defined as regularly participating in heavy physical exercise, such as running or jogging, swimming, cycling, etc., or engaging in vigorous aerobic activity, such as tennis, basketball, or handball (levels 4–7), whereas low physical activity represented none to regular recreation or work requiring modest physical activity, such as golf, horseback riding, calisthenics, gymnastics, table tennis, bowling, weight lifting, and yard work (levels 0–3).

Baseline characteristics of participants were presented according to low and high categories of anthocyanin intake and physical activity. Quantitative values were expressed as mean ± SD and qualitative variables as a percentage. Statistical significance of between-group variation between low and high categories for each exposure were tested using Student's t-test for quantitative variables and chi-squared test for qualitative variables.

To determine the contribution of each food source to the between-person variance of total anthocyanin intake, stepwise-selection regression analyses and nested least-squares linear regression models were conducted. The cumulative R^2 indicates the proportion of variability with the addition of each source, whereas the change in cumulative R^2 identifies each source's contribution to the total variability of anthocyanin intake. Moreover, the contribution of anthocyanins from each food source was presented as a percentage of the total anthocyanin intake. Additionally, subclasses of anthocyanins were presented as percentages of the total anthocyanin intake.

Multivariable linear regression models were used to determine the extent to which each continuous exposure of anthocyanin intake and physical activity level predicted lipid profile measures. Beta coefficients were reported with 95% confidence intervals (CIs) and *p*-values presented for each adjusted model. To control for potential confounding, multivariable adjusted models included age (years), sex (M/F), BMI (kg/m^2), total energy intake (kcal/d), mMDS (points), smoking status (never, current, or former), education level (technical school, some college, associate's degree/Bachelor's degree or higher), marital status (married/single), multivitamin use (yes/no), supplement use (yes/no), sleep (hours/day), prevalent hypertension, dyslipidemia, and type 2 diabetes (yes/no). Additionally, the independent multivariable linear regression models for anthocyanin intake were adjusted for physical activity level, total time spent sitting down (hours/week), and time spent in front of the television, computer and in the car (hours/week), whereas the fully adjusted models for physical activity were adjusted for anthocyanin intake. Total time sitting and sedentary behavior showed a correlation coefficient of 0.28, indicating these covariates measured different forms of inactivity. A sensitivity analysis considered additional exclusions for chronic diseases, women, and supplement use.

To assess the potential effect modification between anthocyanin intake and physical activity on HDL cholesterol, we followed the recommendations by Knol and Vanderweele [47]. First, the prevalence within each subgroup was presented as a percentage and the joint effect of the four possible combinations of low and high exposures of anthocyanin intake and physical activity on HDL <40 mg/dL as relative risks, adjusted for age, sex, and total energy intake. Relative risks were calculated using generalized linear models with Poisson distribution and robust standard errors [48]. A stratification analysis tested effect modification by assessing each dichotomous exposure stratified by the other. This was followed by a comprehensive interaction analysis by applying both multiplicative and additive interaction analyses. Multiplicative interaction was tested by comparing age-, sex-, and energy-adjusted models with and without the interaction term, whereas the RERI was assessed on the additive scale [49].

All analyses were conducted with Stata version 14.0 (StataCorp, College Station, TX, USA). All *p*-values are two-sided and were considered statistically significant at $p < 0.05$.

Supplementary Materials: The following are available online at . Table S1: Independent associations of anthocyanin intake (SD) and physical activity level with lipid profile measures (mg/dL), Table S2: Self-Reported Physical Activity (SRPA) questionnaire, Table S3: Description of the mMDS score developed using Feeding America's Bravest diet and lifestyle questionnaire, Table S4: Sensitivity analysis for anthocyanin intake (SD) and physical activity level on lipid profile measures (mg/dL).

Author Contributions: Conceptualization, M.R.-C. and M.S.-P.; methodology, M.R.-C.; formal analysis, M.S.H. and M.R.-C.; resources and data curation, M.S.-P., S.M. and A.C.; writing—original draft preparation, M.S.H.; writing—review and editing, M.S.H., M.S.-P., M.R.-C., M.A.M.-G., A.C. and S.N.K.; visualization, M.R.-C., M.S.-P. and M.A.M.-G.; supervision and project administration, S.M. and S.N.K.; funding acquisition, S.N.K. All authors had full access to all of the data in this study and take complete responsibility for the integrity of the data, the accuracy of the data analysis. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the US Department of Homeland Security, grant number EMW-2014-FP-00612.

Acknowledgments: The authors acknowledge the Advisory Board and the data advisory monitoring board (DAMB), Indianapolis Fire Department (IFD), Fishers Fire Department, Indianapolis Local 416 support, and the National Fire Organizations; International Association of Fire Fighters, National Volunteer Fire Council, National Fallen Firefighters' Foundation, The Fire Protection Research Foundation, and International Association of Fire Chiefs that support the research, as well as the firefighters and their families for their participation.

Conflicts of Interest: Commercial sponsors of this study are: Kroger Company (coupons and customer loyalty discounts); Barilla America (Barilla Plus Products), Arianna Trading Company, Innoliva and Molino de Zafra, Spain (extra virgin olive oil samples and discounts) and the Almond Board of California (free samples of roasted unsalted almonds). The sponsors had no role in the overall study design; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results. M.S.H., M.R.-C., M.S.-P., M.A.M.-G. and A.C. declare no conflict of interest. Kales reports grants from US Dept. of Homeland Security, non-financial support from Barilla America, non-financial support from California Almond Board, non-financial support from Arianna Trading Company, non-financial support from Innoliva/Molina de Zafra, during the conduct of the study; personal fees from Medicolegal Consulting, personal fees from Mediterranean Diet Roundtable, outside the submitted work.

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Sample Availability: Samples of the compounds are not available from the authors.

PUBLICATION 3

Hershey MS, Fernández-Montero A, Sotos-Prieto M, Kales S, Gea A, Ruiz-Estigarribia L, Sánchez-Villegas A, Díaz-Gutiérrez J, Martínez-González MA, & Ruiz-Canela M. The Association Between the Mediterranean Lifestyle Index and All-Cause Mortality in the Seguimiento Universidad de Navarra Cohort. *American Journal of Preventive Medicine.* 2020;59(6), e239-e248. doi:10.1016/j.amepre.2020.06.014

PUBLICATION 4

Hershey MS, Sanchez-Villegas A, Sotos-Prieto M, Fernández-Montero A, Pano O, Lahortiga F, Martínez-González MA, & Ruiz-Canela M. The Mediterranean Lifestyle and the Risk of Depression in Middle-Aged Adults. *The Journal of Nutrition*. 2021;131(2):604S-615S. doi:10.1093/jn/nxab333

PUBLICATION 5

Hershey MS, Sotos-Prieto M, Ruiz-Canela M, Christophi CA, Moffatt S, Martínez-González MA, & Kales SN. The Mediterranean lifestyle (MEDLIFE) index and metabolic syndrome in a non-Mediterranean working population. *Clinical Nutrition.* 2021;40(5):2494-2503. doi:10.1016/j.clnu.2021.03.026

DISCUSSION

This dissertation includes 3 longitudinal analyses conducted among Spanish university graduates from the "Seguimiento Universidad de Navarra" (SUN) prospective cohort (publications 1, 3, and 4). Research previously conducted in the SUN cohort has focused on the association of risk factors with disease and mortality risk. This has been studied by estimating measures of association, mainly the relative risk either as the HR or OR, and potential interactions have been assessed on the multiplicative scale. A novel aspect that this dissertation has contributed within this cohort has been the study of measures of interaction between the MedDiet and PA on the additive scale. This methodology, which estimated RERI, provided further insights towards the joint association between these risk factors mainly using an additive scale for comparing absolute risks in contrast with the previously reported evidence on relative risks using a multiplicative scale. Furthermore, we also estimated the APs of the joint effect aimed to quantify the effect of the interaction in absolute values, taking a first step towards future analyses on measures of population impact associated with this risk factor combination. Another key contribution of this dissertation includes evidencing the association of the previously validated MEDLIFE index, which comprehensively captures the traditional Mediterranean lifestyle, with mortality and depression.

In addition, this dissertation includes 2 cross-sectional studies that used baseline measurements from "Feeding America's Bravest", a two-year randomized trial aimed to test a nutritional intervention in US career firefighters (publications 2 and 5). This study design allowed us to apply our line of research to a non-Mediterranean population. Therefore, our findings have contributed new insights to the field of chronic disease epidemiology with regard to measures of interaction between modifiable lifestyle factors and the Mediterranean lifestyle among a non-Mediterranean population. In the following discussion we summarize the primary results of the five publications, state strengths and limitations of the studies, provide interpretations given the currently available evidence, discuss biological plausibility, generalizability, and the potential application along with future directions of our findings.

Summary of Key Results

Better together than apart for the prevention of premature mortality

In the SUN cohort, we dug deeper into the methodology for interaction analyses to present a practical article that discusses the main concepts of interaction, reasons for reporting measures of interaction on the additive scale, and an example of a comprehensive interaction analysis. Our comprehensive review on the combination of the MedDiet and PA on all-cause mortality demonstrated the current research gap for reporting interaction analyses, identifying only four studies that met our inclusion criteria.[74,95,177,178]

Only one study by Alvarez-Alvarez at el., previously conducted in the SUN cohort, reported *p*-values for interaction by using likelihood ratio tests on the multiplicative scale, but none on the additive scale. Yet, our analysis within the same study population of Spanish university graduates further demonstrated the interrelatedness between dietary and PA habits. Those with higher levels of PA exhibited slightly higher MDS, greater total daily energy intake with a greater percentage from carbohydrates, higher intakes of fiber, vegetables, fruits, cereals, fish, dairy, and nuts, as well as a lower percentage of total energy intake from fat and lower meat consumption compared to participants with a low PA level. On the other hand, those with higher MDS showed slightly higher PA scores, more frequent exercise, higher weekly energy expenditure, faster walking pace, more minutes walking per day, climbed more stairs, and spent fewer hours sitting per day compared to participants with lower MDS adherence. The significant differences for each exposure across categories of the opposite exposure suggested that a greater adherence to the MDS was associated with a greater PA level and vice versa.

Thereafter, our original interaction analysis in the SUN cohort for the conjoint effect of low MDS (0-3 points) in combination with a low PA score (0-3 points) observed an increased risk of all-cause mortality greater than the sum of the individual risk factors. These results suggested a potential additive interaction or synergism between both exposures. Furthermore, the absolute measures presented, RERI and the AP due to interaction, provided informative estimates regarding the impact of the joint effect. According to our data, which observed increased relative risks as lifestyle factor combinations worsened that surpassed the risks of the individual factors, we quantified the effect due to the interaction MDS*PA was 36% of the total joint effect.

Anthocyanin Intake and Physical Activity: Associations with the Lipid Profile of a US Working Population

As evidenced by previous studies, US career firefighters are a working population at high CVD risk, therefore, we studied the potential synergism between the dietary intake of anthocyanins, a cardioprotective polyphenol, and level of PA on lipid parameters. The US Preventive Services Task Force (USPSTF) identifies dyslipidemia as a CVD risk factor, defined as LDL >130 mg/dL or HDL cholesterol <40 mg/dL. Driven by our initial stratification analysis that showed independent associations on HDL cholesterol, we studied the joint effect between anthocyanin intake and PA on HDL cholesterol <40 mg/dL followed by stratification and interaction analyses. While the stratification analysis tested for the possible effect modification of one factor on the causal pathway of another factor, the interaction analysis tested the potential synergism between two independent causal pathways to produce an effect greater than the sum or multiplication of the two individual effects. Although neither interactions on the multiplicative scale nor RERI on the additive scale were statistically significant, the joint effect analysis suggested that the combination of low anthocyanin intake and low PA more than doubled the relative risk of having HDL cholesterol <40 mg/dL, compared to the reference category of the joint exposure to both high anthocyanins and high PA.

The Association Between the Mediterranean Lifestyle Index and All-Cause Mortality in the Seguimiento Universidad de Navarra Cohort

In a longitudinal analysis within the SUN cohort, the validated 28-item MEDLIFE index identified participants with low (2-10 points) and high (15-23 points) adherence to MEDLIFE. Compared with lower MEDLIFE adherence, participants with higher scores exhibited better adherence to the MedDiet, higher total energy intake, greater PA, and were more likely to be never or former smokers and have a chronic disease. Higher adherence to MEDLIFE, encompassing food consumption, dietary habits, PA, rest, social habits, and conviviality, was associated with a 41% relatively lower risk of all-cause mortality compared with lower adherence. In a stratified analysis, an association was found only among participants with age at last contact ≥50 years old, suggesting that the inverse association of the MEDLIFE index with CVD deaths was stronger in older participants. Inverse associations were found for the overall MEDLIFE score and lifestyle block, whereas no associations were observed for the dietary blocks among a Mediterranean adult population.

The Mediterranean Lifestyle and the Risk of Depression in Middle-Aged Adults

In this analysis, Spanish university graduates who volunteered for the SUN cohort had an average MEDLIFE score of 12 points. The individual MEDLIFE components; vegetables, PA, red meat, napping, and fruit showed the greatest contribution to the between-person variability of final MEDLIFE scores. Compared to the first quartile (3-10 points), the second and third quartiles (11-12 and 13-14 points) of MEDLIFE adherence were significantly associated with a 18% (95% CI: 69%-96%) and 26% (95% CI: 61%-89%) decreased relative risk for incident depression, respectively. The fourth quartile (15-23 points) did not show any significant association with incident depression, suggesting a L-shaped dose-response curve across MEDLIFE categories.

The Mediterranean lifestyle (MEDLIFE) index and metabolic syndrome in a non-Mediterranean working population

The prevalence of metabolic syndrome among our study population of US career firefighters was 17.7%. In this cross-sectional study, participants with higher MEDLIFE adherence (11-17 points) had 71% lower odds of having metabolic syndrome compared to those with lower MEDLIFE adherence (2-7 points). Higher MEDLIFE scores were significantly associated with lower levels of abdominal obesity and hypertriglyceridemia, and showed a more favorable lipid profile, including total cholesterol, LDL cholesterol, and total cholesterol:HDL cholesterol, compared to low MEDLIFE scores. Furthermore, inverse associations with metabolic syndrome were observed for the items; preference for whole grain products and watching television ≤4 hours per week, Block 2: Mediterranean dietary habits, and for each additional point of the MEDLIFE index in a non-Mediterranean working population.

Interpretation of Results

Interaction analyses

The rationale for studying interactions is to better understand which inter-related biological mechanisms are shared by two exposures that create an observable synergistic effect, identifying subgroups of the population which an intervention would be most advantageous.[153] We applied the recommendations by Knol and Vanderweele for presenting a comprehensive interaction analysis in both a longitudinal study among a sample of middle-aged Spanish university graduates in the SUN cohort and a cross-sectional study within the "Feeding America's Bravest" trial among a US working population.

In the SUN cohort, we first demonstrated the MedDiet and a physically active lifestyle have been studied in epidemiology as single or combined lifestyle factors, however interaction analyses were not commonly reported. We suspect that our review is not only indicative of the research gap on interaction analyses conducted for this particular combination of factors on all-cause mortality, but rather seems to be the case for most combined effects between different lifestyle factors on health outcomes in general. This research gap implies that the possibility of detecting a biological or mechanistic interaction on the additive scale is usually discarded when investigators explore joint effects and assess interactions exclusively with likelihood ratio tests to detect statistical interactions on the multiplicative scale. Therefore, we used the comprehensive methodology recommended by Knol and Vanderweele[84] which may provide necessary insights for understanding potential synergisms between lifestyle factors that support multicomponent lifestyle interventions as effective public health strategies.

Given the effect of an interaction varies according to the prevalence of the two exposures and the outcome within a given subgroup, the public health implications of the MedDiet and PA depend on the proportion of the population in which these factors occur jointly.[79] The greater the number of subgroups, the fewer the cases of mortality which correspond to each combination, the smaller the effect observed from the interaction. Hence, according to our original analysis, the subgroup with low MedDiet adherence and low PA would benefit from an intervention targeting both habits simultaneously to reduce the risk posed by this synergism. This subgroup received three or less points for both the MDS and PA scores, indicative of individuals with ample room for improvement in many possible aspects of both diet and PA compared to the rest of the study population. Similarly, from a more applicable perspective to public health, greater risk reductions were observed across combinations of MDS and PA on protective scales. Although quantifying the interaction requires transforming healthy lifestyle factors into their corresponding risk factors, by selecting the lowest risk category as the reference, the observed joint effect of adhering to the MedDiet and PA, greater than the sum of the effect of each individual lifestyle habit, offers a more translatable message to the public.

With regard to our interaction analysis in "Feeding America's Bravest", our results suggested a possible joint effect between a low anthocyanin intake (below the median intake) and a low PA score (0-3 points) on HDL cholesterol greater than the sum of the individual exposures. This interrelationship between food consumption, micronutrient intakes, and lifestyle factors has been recently demonstrated by the PREDIMED study, which observed participants who had a greater fruit and vegetables variety score had significantly higher intakes of fiber, vitamins, minerals and flavonoids, and were more likely to be physically active and non-smokers.[179]

The current scientific literature supports the beneficial effects of anthocyanin intake and PA, both individually and combined, on cardiovascular health biomarkers and outcomes. A meta-analysis of 45 randomized controlled trials demonstrated the independent association for the consumption of berries and purified anthocyanins (2.2−1230mg anthocyanins/day) significantly increased HDL cholesterol and reduced total cholesterol, LDL cholesterol, and TG.[180] Evidence on PA supports aerobic exercise of adequate intensity, duration, and volume results in favorable and independent improvements of blood lipids and lipoproteins in individuals with and without dyslipidemia. The most consistent findings were shown for increases in HDL cholesterol.[181–183] Furthermore, the potential synergism between anthocyanins and PA has also been speculated within a recent intervention study in healthy adult males that suggested anthocyanin intake duration affects metabolic responses, including fat and carbohydrate oxidation, during moderate-intensity walking exercise.[184] Furthermore, in the "Nurses' Health Study II", the combined effect of >3 servings per week of the two main food sources of anthocyanins, namely strawberries and blueberries, demostrated stonger inverse associations with miocardial infarction compared to ≤1 serving per month. However, stratification analyses across various risk factors, including PA, did not observe statistically significant *p*-values for interaction.[185] Our interaction analysis also lacked statistical significance (*p*=0.72; RERI: 0.02; 95% CI: -1.63 to 1.66; p=0.98), however, our study was novel in reporting measures of interaction on the additive scale, in addition to the multiplicative scale, for the potential synergism between two causal factors of dislipidemia.

Although we studied the specific effect of anthocyanin intake, this plant-based bioactive compound is consumed within the bigger context of the overall dietary pattern. It is the synergy between foods and nutrients that defines diet quality; therefore, our findings should be considered within the context of multiple possible pathways by which an inappropriate diet could lead to the development of CVD.[186] In this context, another study conducted within the same "Feeding America´s Bravest" baseline data observed that a 1-unit increase in the mMDS was associated with a decrease in total cholesterol:HDL cholesterol ratio and an increase in HDL-cholesterol.[173] In addition, a systematic review noted that most of the potential effects of anthocyanin intake observed across RCTs linked to CVD development and progression, including LDL cholesterol, HDL cholesterol, total cholesterol, TG, or blood pressure, were not statistically significant. Nevertheless, observed improvements in biomarkers were consistent across studies in this systematic review, particularly in those with elevated lipids at baseline.[187] Our findings support the rationale for promoting an anthocyanin-rich diet, such as the MedDiet, with an abundant intake of polyphenols through the frequent and customary consumption of fruits, vegetables, wine, and extra virgin olive oil.[188] These nutritional exposures in combination with PA may promote a more favorable lipid profile compared to either factor alone in a population at high CVD risk.

MEDLIFE analyses

Thereafter, the validated MEDLIFE index allowed us to study a combination of numerous lifestyle factors that characterize the unique way of living in the Mediterranean region, beyond the MedDiet and PA. We evidenced its association with all-cause mortality, cause-specific mortality, and incident depression in a Mediterranean population, meanwhile its association with the metabolic syndrome and its components were assessed in a non-Mediterranean population. The MEDLIFE index and MDS or mMDS were replicated for both study populations using the available baseline variables. Furthermore, the use of a simple lifestyle score allowed us to assess each item and block as a secondary analysis. This was of interest given that most individual MEDLIFE components were not independently associated with all-cause mortality, depression, or metabolic syndrome, yet the global MEDLIFE exposure showed clear associations. Although final MEDLIFE scores were lower among the non-Mediterranean population and the main drivers of MEDLIFE associations may be different compared to a Mediterranean population, the overall effect may provide a better understanding of the holistic MEDLIFE exposure across different geographical regions.

Our findings for the MEDLIFE index were supported by two other studies conducted in parallel. Mata-Fernández et al. studied the association between the MEDLIFE index and CVD, observing a 50% decreased risk of primary cardiovascular events (myocardial infarction, stroke or cardiovascular death) among participants with scores between 14-23 points compared to 0-9 points in the SUN cohort.[143] In addition, Sotos-Prieto et al. reported that a higher MEDLIFE score was associated with a 27% lower prevalence of metabolic syndrome, 37% lower prevalence of abdominal obesity, and 24% lower prevalence of low HDL cholesterol levels in the Study on Nutrition and Cardiovascular Risk in Spain (ENRICA) cohort. Moreover, MEDLIFE as a continuous variable was inversely associated with the CVD risk factors; HOMA-IR and high-sensitivity C-reactive protein. Lastly, the MEDLIFE index was associated with a 45% relatively lower risk of all-cause mortality and 69% relatively lower CVD mortality risk in this same study.[144] These findings among a representative sample of the adult Spanish population support our findings for a 41% decreased risk of all-cause mortality and 65% reduced risk of CVD death in the SUN cohort.

In light of this evidence for the Mediterranean lifestyle on improved cardiometabolic health and reduced mortality, we aimed to study its association with yet another highly prevalent chronic disease worldwide, depression. In a similar manner, only one MEDLIFE item was independently associated with depression, meanwhile the global MEDLIFE scores showed significant inverse associations for adherence levels of 11-14 points compared to 3-10 points. However, this potential synergism was limited to medium adherence categories. The lack of statistical significance and smaller magnitude of effect for 15-23 points suggested that a L-shaped curve may better describe the dose-response pattern for this association. Our results may be explained in part by similar nonlinear associations found for the individual components of the MEDLIFE index with depression, including the MedDiet and PA[189,190], as well as the smaller sample size and fewer cases found in the fourth quartile.

Beyond the MedDiet and PA, poor overall relationship quality, lack of social support, and social strain have been previously associated with increased risk of depression.[191] On the contrary, confiding in others has been inversely associated with incident depression.[192] Furthermore, recent studies have observed that exposure to longer television/screen time and non-optimal sleep/daytime napping were prospectively associated with an increase in the frequency of depressed mood, among those with and without depressive disorders, as well as incident depression.[103,192] Nonetheless, independent of the individual associations evidenced for each item, the MEDLIFE index measures a singular joint exposure that reflects the traditional Mediterranean lifestyle with a particular dose-response relationship.

With regard to a US working population and as evidenced by previous studies, US career firefighters often experience several lifestyle risk factors, including poor diet quality and suboptimal eating habits, physiological stress from strenuous PA at work, emotional stress, and environmental pollutants, in addition to low fitness.[193,194] Such lifestyle factors have led to a high prevalence of obesity and other chronic conditions, including hypertension, hypercholesterolemia, and high blood glucose, contributing to significant morbidity and disability among this working population.[194–197] Existing evidence on dietary workplace interventions for health promotion support healthy dietary changes, such as the current *Dietary Guidelines for Americans*, which includes the MedDiet (with its characteristic good palatability and frugality), for the long-term prevention of diet-related chronic disease.[198] On the other hand, evidence supports workplace PA interventions to improve body composition.[199] However, evidence of the joint effect of multicomponent lifestyle patterns, including interrelated factors such as PA and sedentary behavior within a general way of living, on metabolic syndrome is limited.[200,201]

Our findings on the Mediterranean lifestyle in the SUN cohort and "Feeding America's Bravest" suggest a potential synergism between MEDLIFE factors that may provide greater health benefits (in terms of mortality, depression, and metabolic syndrome) than any individual factor alone. Evidence in this line may contribute towards effective strategies for the primordial prevention of premature death, particularly among those over the age of 50 years at greater risk of CVD. Moreover, a recent meta-analysis evidenced the association between a healthy lifestyle and all-cause mortality was stronger in studies with longer follow-up or among younger participants, indicating larger benefits could be obtained if people adopt healthy lifestyles at an early age and follow for a long time.[112] Therefore, even if a threshold exists for the protective effect of the MEDLIFE index on incident depression, our studies support prevention strategies based on the promotion of Mediterranean lifestyle behaviors may reduce the burden of chronic disease and subsequent premature mortality.

Biological Plausibility

Interaction analyses

In the SUN cohort, the global effect of the MedDiet produced by the sum of its components may be attributed to its antioxidant and anti-inflammatory properties.[40,41] More specifically, accumulating evidence indicates the traditional MedDiet induces: lipid-lowering effects, protection against oxidative stress, inflammation and platelet aggregation, modification of hormones and growth factors involved in the pathogenesis of cancer, inhibition of nutrient sensing pathways by specific amino acid restriction, and gut microbiota-mediated production of metabolites influencing metabolic health.[202] With regard to PA, a randomized controlled trial has suggested that improvements in lipids and lipoproteins were related to the amount of activity and not the intensity of exercise or improvement in fitness. In particular, a relatively high amount of regular exercise (equivalent to jogging 27.2 to 28.8 km/week at moderate pace) significantly improved the overall lipoprotein profile by decreasing LDL cholesterol size, increasing HDL cholesterol concentration and size, and decreasing TG, which was not observed for lower amounts of exercise.[203] In addition to optimal nutrition, being physically active promotes healthy aging by counteracting the negative effects of inactivity, including oxidative stress and inflammation.[204,205] These effects of PA have been explained through the activation of mechanisms such as AMP-activated protein kinase that increase the AMP:ATP ratio that promotes the expression of several metabolic and regulatory proteins involved in glucose metabolism, endothelial nitric oxide synthase, antioxide synthase, antioxidant enzymes, and the downregulation of inflammation.

Although our interaction analysis in “Feeding America’s Bravest” lacked statistical significance, the joint effect suggested a potential synergism most likely due to shared underlying mechanisms.[206] This potential synergism may be attributed to an enhanced bioavailability of anthocyanin-derived metabolites involved in mechanisms of oxidation during PA.[184] It is known that anthocyanins possess anti-inflammatory and antimicrobial activities, which improve cardiometabolic, visual, and neurological health. These protective effects have been explained by different mechanisms and pathways, including the free-radical scavenging pathway, cyclooxygenase pathway, mitogen-activated protein kinase pathway, and inflammatory cytokines signaling, as well as some crucial cellular processes, including the cell cycle, apoptosis, autophagy, and biochemical metabolism.[207,208]

Furthermore, the combination of both a healthy diet and PA has demonstrated a decrease in fasting glucose, insulin resistance, blood pressure, and low-grade inflammation.[75] Thus, evidence of the effects of dietary anthocyanins, the MedDiet, and PA support the possible synergistic associations between the MedDiet and PA on all-cause mortality, as well as anthocyanin intake and PA on lipid profiles, are most likely due to both factors enhancing health through complementary metabolic and molecular mechanisms.

MEDLIFE analyses

The MEDLIFE index is a singular exposure that captures the combined effect of numerous Mediterranean lifestyle behaviors associated with CVD risk in addition to diet and leisure time activity.[124] Consequently, this comprehensive measure of an overall lifestyle pattern may capture both direct and indirect underlying effects that support the possible synergism of numerous modifiable factors. When considering the five components of metabolic syndrome and all-cause mortality, the overall effect of the Mediterranean lifestyle may be attributed to the biological mechanisms supporting healthy physiological pathways and molecular mechanisms that combat chronic stress and inflammation. In addition to the traditional MedDiet and PA, a traditional afternoon nap ≤30 minutes a day complements a good night's sleep (6–8 hours) to reduce mortality and CVD risk by combatting the effects of insufficient sleep, which triggers metabolic and endocrine hormones, altering appetite and glucose metabolism.[209–211] Although evidence is limited, stronger social relationships have been associated with increased survival as a result of multiple biological pathways that include better immune function.[212,213] In conclusion, the healthy functioning of systems impede disruptions in the autonomic nervous system, hypothalamic-pituitary-adrenal axis, cardiovascular, metabolic, and immune systems responsible for the biochemical changes characteristic of metabolic syndrome and, thus, determinants of healthy longevity.[214]

On the other hand, the L-shaped association for the MEDLIFE index on depression could be explained in part by the nature of some individual components which have shown similar non-linear associations.[189,190] It has been speculated the plateau effect observed between the MedDiet and incident depression could be explained by certain psychological elements of neurotic or obsessive traits in some participants with the highest MDS adherence.[189] The health benefits of the MedDiet on depression have been attributed to an elevated intake of phytochemicals, B vitamins and minerals (zinc and magnesium), poly- and mono- unsaturated fat (omega-3 fatty acids and oleic acid), and fiber. These components contribute to an adequate cortisol regulation, decreased oxidative stress markers, increased antioxidants, decreased inflammatory markers, modulation of gut-brain axis, mitochondrial function, and modulation of epigenetic state.[215,216]

The decreased risk associated with egg consumption is most likely attributed to its rich content in tryptophan, vitamin B12, folate, and choline, among other necessary micronutrients for the production of neurotransmitters, such as serotonin, dopamine, and norepinephrine, involved in the regulation of mood, appetite, and cognition.[217] Nevertheless, the criteria for 2-4 servings per week of eggs was only one of fifteen items within the MEDLIFE index that captured the overall Mediterranean food consumption.

The inverse association between PA and incident depressive symptoms has demonstrated a U- or J-shaped relationship, which may be due to increased oxidative stress and cortisol response caused by high PA levels.[190] With respect to the sociocultural components, mechanistic explanations are still needed to support the associations found for other lifestyle habits with depression. For instance, it has yet to be established if higher screen time is associated with depressed mood due to the sedentary nature of screen time activities or due to the potential 'direct' psychological impact of content absorbed from TV time/computer time itself, or even as an exacerbating mediator between poor sleep and increased risk of depression.[103] Hence, evidence is lacking on the nature of these individual associations to fully explain the dose-response curve of the global MEDLIFE exposure on incident depression.

Strengths and Limitations

Strengths

The main strength shared among the interaction analyses conducted in the SUN cohort and "Feeding America's Bravest" includes the estimation of measures of interaction on the additive scale, in addition to *p*-values for statistical interaction on the multiplicative scale. Testing for interaction on the multiplicative scale evaluated whether the association deviated from multiplicativity, according to the model, whereas testing for additive interactions or the departure from risk additivity evaluated whether the number of cases attributable to the combined effect of two risk factors was more or less than the sum of the cases that would be caused by each risk factor separately. An additive interaction would have indicated a greater absolute risk reduction from an intervention among certain subgroups more so than others, offering greater insight towards public health and clinical decision making.[87] Our study offered an additional strength by measuring PA, a multidimensional exposure, with a 8-item PA score, which captures the type, duration, and frequency of PA. Just as the preference for studying dietary patterns over isolated nutrients or foods has been established, researchers are advocating the same for PA.[218]

The primary strength shared among the MEDLIFE studies includes the aim of the MEDLIFE index to holistically capture the multifactorial etiology of chronic lifestyle diseases, which have risen with the cultural divergence from traditional ways of living.[72,119] Similar to holistic approaches in nutritional epidemiology that acknowledge mechanisms of action that associate diet with health outcomes are complex, multifaceted, and interacting, lifestyle scores aim to better capture the heterogeneity and multifactorial determinants of chronic diseases and mortality.[123,126,216] Such scores need to accommodate for concepts of interaction and synergism. Our comprehensive assessment of multifaceted habits that form an overall way of life, which share inter-related biological mechanisms, created an observable synergistic effect, whereas the assessment of each individual item lacked statistical power. Other lifestyle indices, such as a Revised LS7, have combined CVD risk factors; however, LS7 includes risk factors (high BMI, total cholesterol, blood pressure, and fasting blood glucose) which are largely preventable by means of a healthy lifestyle and require clinical and biochemical measurements.[219] Furthermore, in comparison with other simple lifestyle scores that address a general healthy lifestyle[112,220], the MEDLIFE index specifically measures the traditional Mediterranean lifestyle and can be easily self-assessed by the patient. Furthermore, a key aspect of this dissertation´s novelty has been the extrapolation of the MEDLIFE index to a non-Mediterranean population, namely US career firefighters, which mirrors the current dietary and PA guidelines for Americans.

Specific strengths of the SUN cohort include the prospective design with long follow-up, the large population size, high retention rate, adjustment for numerous potential confounders, validation of the methods in specific subgroups, and greater expected validity of self-reported data from an educationally homogenous population. These strengths are attributed to the only criterion for this cohort which requires participants to be university graduates, ensuring better internal validity due to the high literacy level in comparison with the general population. In addition, the FFQ, PA questionnaire, self-reported cases, the MEDLIFE index, and some covariables have been previously validated in Spanish populations.[124,136,142,146,148]

Some strengths of our studies within the "Feeding America's Bravest" included the standardized procedures for data collection by trained PSM clinical staff during work hours in private areas, which limited possible misclassification bias. The extensive baseline data collection from annual medical examinations for each consented IFD subject was a notable strength of this study. These variables allowed for replicating the MEDLIFE index, extensive control for potential confounding, and internal validation of some self-reported variables, such as PA, which we observed had a moderate correlation (r=0.41) with VO2max, a strong indicator of physical fitness. Some of these measurements retrieved from the PSM electronic system included at rest and maximal heart rate, systolic and diastolic blood pressure, heart rate recovery, electrocardiographic findings, hip and waist circumference, body composition, data regarding grip strength, leg-press and push-ups performed, and routine lab values (i.e., lipid panels, hs-CRP, and glucose). Furthermore, the study team's data collection of baseline anthropometrics included firefighters' body fat percentage and waist circumference, in addition to BMI, for more accurate measures of body composition. Lastly, an extensive nutrient dataset was derived from the FFQ by a trained professional according to previously described methods.[165,221] Thus, the comprehensive data collection was a primary strength of our studies, which allowed for appropriate study designs to conduct analyses of association and interaction.

In addition, the mMDS was created to address the firefighters' eating habits both at work and at home for greater precision of daily eating patterns. This previously validated mMDS created for US career firefighters assessed MedDiet adherence based on their lifestyle, eating habits, nature of work (meals at home and at work), type of drinks, and alcohol consumption. The 13-item mMDS questionnaire showed good correlation (r>0.75) with the previously validated 131-item FFQ and MEDAS, meanwhile specific self-reported nutrients derived from the FFQ were correlated with biomarkers in plasma, specifically omega-3, EPA and DHA.[172] Therefore, despite the questionable quality of MedDiet scores in different populations, this mMDS was tailored specifically to our study population.[162]

Limitations

Limitations shared across the five published observational studies included the potential misclassification of some exposures due to the self-reported nature of the questionnaires and lack of validation of some variables, including the MEDLIFE index in our study populations. However, this inaccuracy would most likely be non-differential by equally misclassifying participants with and without the given outcome and shift the estimate towards the null.[222] Although multivariable-adjusted models controlled extensively for potential confounders, the primary limitations posed by measuring long-term exposures from self-reported measurements include residual confounding due to variations in habits over time and suboptimal recall, which cannot be completely eliminated.[155,156] Furthermore, our analyses only employed baseline data, whereas analyses with repeated measures might have detected associations and interactions over time. However, limitations to conduct repeated measurements are inherent to the differences between the extensive and comprehensive baseline assessment in the SUN cohort and "Feeding America's Bravest" in comparison with the shorter follow-up questionnaires. Moreover, the differences between our MEDLIFE exposures and the original MEDLIFE index emphasize the research gap on the lack of standardized methods in assessing lifestyle factors among studies.[93]

The main limitation of the interaction analysis in the SUN cohort was the lack of statistical power most likely attributed to the distribution of cases and study population size. Considering that a multivariable analysis requires a large sample size, an interaction analysis requires an even greater sample size and, therefore, the AP due to interaction may add strength to the RERI estimate. Although we may not have had the sufficient statistical power for a more robust interaction analysis, the power to detect interactions tends to be greater on the additive scale than the multiplicative scale when the main effects are positive.[223,224] In addition, our results should be interpreted with caution due to the use of recoded variables as risk factors (i.e. nonadherence to the MedDiet rather than high adherence and physical inactivity rather than PA), which may not infer the same results for exposures in their preventive form.[152] Moreover, similar to a previous concern within the GBD Study, the broader issue of comparability of risk factors may also apply to other lifestyle studies, such as ours. This concern is the effect of each lifestyle factor may vary according to the choice of measurement of the exposures within a given population.[218] Therefore, our categorization of exposures may be debatable given the irregular distribution of participants, nonetheless, the biological relevance and case distribution were considered to present the most appropriate analysis.[225]

Our articles in the SUN cohort on the associations of the Mediterranean lifestyle with mortality and depression acknowledged a possible misclassification, due to the self-reported data and lack of validation of some variables, the MEDLIFE index, and its psychometric properties. In addition, the studies would have gained statistical power if the sample size of healthy individuals were larger and had a longer follow-up time, capturing more incident cases of depression and deaths from CVD. Finally, the items and scoring criteria may be debatable; nevertheless, the MEDLIFE index relied on the independent recommendations from the MedDiet Foundation for intake cut off points and existing evidence on lifestyle factors in the Mediterranean region.

Limitations specific to our studies conducted in the "Feeding America's Bravest" trial mainly include the cross-sectional study design, which hinders the ability to infer causality from our results, since the temporal sequence is not well defined. Instead, we have generated viable hypotheses for future studies within larger longitudinal cohorts or intervention trials, while limiting the possibility of reverse causality.[226] In addition, completion of the lifestyle questionnaire by all enrolled participants would have increased sample size and provided greater statistical power. Nonetheless, our results were in line with previous research, suggesting that a larger sample size would have observed statistical significance.

In the interaction analysis conducted in "Feeding America's Bravest", non-differential misclassification is possible due to the indirect measurement of total daily energy intake and anthocyanin intake derived from each participant's baseline FFQ. Furthermore, the validated semiquantitative FFQ did not include all common food sources nor supplementation of anthocyanins, meanwhile some items captured more than one food with varying anthocyanin content. Currently little is known on the bioavailability of anthocyanins and the concentration in different food sources may vary greatly due to influences such as genetic, environmental, and agronomic factors, including light, temperature, humidity, fertilization, food processing, and storage conditions.[227] In addition, although PA was reported using a validated questionnaire, self-reported PA has been demonstrated to be less predictive of CVD risk than objective accelerometer measurements.[183,203] Nonetheless, our measure of PA showed a moderate correlation with VO2max (r=0.41) in this study population.[169–171] With regard to the interaction analysis, due to our relatively small sample size, statistical power was an important limitation of our study.

In our study on MEDLIFE and metabolic syndrome, the observed prevalence of metabolic syndrome was almost 18%, therefore the ORs may have overestimated the relative risks. In addition, MEDLIFE items, block classification, and scoring criteria may be debatable in a non-Mediterranean population, nonetheless, they relied on the independent recommendations from the MedDiet Foundation for intake and behavioral cut off points. Moreover, since 1995 a consumer-friendly MedDiet pyramid developed by the Harvard School of Public Health, the World Health Organization, and Oldways, a nonprofit food think tank in

Boston, has offered guidelines tailored to the American population and as of 2015 the *Dietary Guidelines for Americans* have nudged US nutritional policy towards a traditional MedDiet.[71,132,228,229] Due to the cross-sectional study design, it is possible that part of the effect observed was a consequence of metabolic syndrome itself. However, given previous knowledge, less healthy lifestyle behaviors seem to be more likely causes than consequences of metabolic syndrome. Nevertheless, prospective longitudinal cohort studies and intervention studies, such as the PREDIMED-Plus trial, are needed to assess the temporal sequence.[230] A more comprehensive data collection may have allowed for greater reproducibility of the original MEDLIFE index (i.e., herbs and spices, socializing with friends, collective sports). Lastly, a common concern with metabolic syndrome outcomes includes dichotomizing the continuous components, criticized as being arbitrary and possibly resulting in false positives, further misclassifying at risk populations of CVD and T2D. However, it is arguable any individual with any metabolic syndrome component may benefit from a healthy lifestyle.[25]

Generalizability

Interaction analyses

In the SUN cohort studies the primarily young and educated study population is not representative of the general population; however, biological plausibility should be the basis for generalizations in epidemiology.[231,232] Our interaction analysis in this cohort of Spanish university graduates focused on quantifying the interaction between the MedDiet and PA, however more studies are needed to study other dietary patterns for greater generalizability and a meta-analysis of the effect attributed to the interaction would provide stronger evidence.

Similar to studying an overall dietary pattern as a cumulative effect of several individual components, lifestyle indices are used to study the cumulative effect of individual behaviors. Nevertheless, the mechanism by which these individual components interact is complex, suggesting the use of interaction analysis as an essential statistical method to complement frequently reported joint effects. While quantifying the synergism between the MedDiet and PA focuses on one interaction among many possible lifestyle interactions, this methodology for interactions on the additive scale of >2 risk factors and network analysis may be advantageous towards understanding the potential synergism between multiple lifestyle factors.[233,234] More studies on interactions are needed to fill this gap in nutritional epidemiology and provide high-quality evidence as interest grows in studying overall lifestyle patterns on the general population health.

MEDLIFE analyses

Due to the predominately male prevalence of the firefighter profession and, thus, our study population of Midwestern US career firefighters from the "Feeding America's Bravest" trial, 81% of which identified as Caucasian, is not representative of the general US population. Our results must be extrapolated to women with precaution, however, biological plausibility should be the basis for generalizations in epidemiology.[231,232] Although cultural relevance of the MedDiet and Mediterranean lifestyle in non-Mediterranean populations may be debated, our findings are robust and the MEDLIFE items support the proposed shifts to improve food, beverage, PA and other lifestyle factors in the US for general health promotion.[132] Furthermore, the association between lifestyle and health outcomes may vary according to sex and cultural differences, particularly through social components when comparing collectivistic and individualistic cultures.[214,235] Moreover, even though our participants were primarily healthy middle-aged men, MEDLIFE criteria may not adequately consider lifestyle behaviors recommended to individuals with health conditions.[124]

The MEDLIFE index was not designed with the aim of assessing each block separately but rather to evaluate the comprehensive Mediterranean lifestyle reflecting culture and tradition. Therefore, the main drivers of the association could be different in each population, but the overall effect may provide a better understanding of the whole Mediterranean lifestyle, tradition, and culture in a holistic approach, and therefore add the novelty of going beyond the exclusive focus on the foods consumed.[144] Although our results for the MEDLIFE index suggested this population of US career firefighters at high CVD risk may benefit from such recommendations at the public health level, more evidence is needed in other workplace populations as well as the general population. For our study on depression, although our prevalence of depression at 6% was similar to the Spanish population (5.3%), our results should be extrapolated to the general population with caution, since our primarily young and educated population is not representative of the general population.[236] Moreover, the MEDLIFE index and our study inclusion criteria aim to represent a healthy, young and educated population, whereas individuals at higher CVD risk or of greater age are known to have higher rates of depression and vice versa.[237–239]

Application and Future Directions

Interaction analyses

Our findings for the potential interaction between the MedDiet and PA underline the important public health message of adhering to both modifiable lifestyle factors for the prevention of premature mortality. Additionally, we anticipate our reported interaction analysis may serve as an example for future studies to report complete interaction analyses on the multiplicative and additive scales after presenting the individual, joint, and stratified effects of the combined exposures. Both follow-up duration and sample size should be considered when conducting additive interaction analyses, provided that the detection of causal interactions may depend on the progression of time and more precise estimates may require very large study populations.[240] This frequent concern may be solved in part by calculating the AP due to interaction, which does not solely rely on statistical significance, making it a valuable measure of interaction.[88] However, despite poor statistical significance, the joint effect and stratification analysis still offer substantial insight on the effect modification and potential interaction between the given exposures. Provided that chronic disease affects all aspects of health and the combination of poor diet and PA may play a greater role in the burden from chronic disease, more so than overall mortality, future research should study cause-specific mortality, premature mortality, and death free of chronic disease, including CVD, diabetes, cancer, and metabolic syndrome.[241,242] Not only the MedDiet and PA, but other lifestyle factors as well should be studied in combination with each other to understand the interaction between multifactorial causes of disease and mortality and create effective guidelines for general, at risk, and diseased populations.

Translating the findings of an interaction analysis into a public health message is difficult. Future strategies will require educating health professionals to understand results from studies analyzing how to best communicate the synergism between lifestyle factors to communicate the synergistic health benefits to patients who are healthy or have cardiometabolic risk.[243] There are considerable limitations when asking a dietitian to deliver interventions on PA or a PA expert to deliver nutritional interventions, let alone other lifestyle factors. Thus, clinicians should be specifically trained to discuss lifestyle factors as proposed by Frates et al.[244] This issue is similar at the public health level, we need to better combine the dietary guidelines with PA guidelines in a more integrative manner, such as the *Dietary*

Guidelines for Americans[228] and the *Physical Activity Guidelines for Americans*.[245] Multi-component lifestyle interventions must be supported by long-term policies, communication, and implementation strategies across sectors.[103,239,246] Studying potential synergisms between lifestyle factors, such as the MedDiet and PA[67], is particularly relevant given the current research gaps in studying the complexities of combined aspects of lifestyle in comparison to isolated behaviors. Measures of additive interaction in particular provide key insights into the nature of joint effects and help target lifestyle interventions more effectively.

The USPSTF currently recommends offering adults who are overweight or obese and have additional CVD risk factors intensive behavioral counseling interventions to promote a healthful diet and PA; meanwhile, evidence among those without known risk factors suggests a positive but small benefit for the prevention of CVD.[247,248] Understanding the intercorrelation and combined effect of dietary bioactive components, such as anthocyanins, with PA on CVD risk parameters may contribute towards better primordial prevention strategies for CVD. With regard to the promotion of anthocyanin-rich diets for healthy lipid profiles, previous studies have demonstrated berries, as main sources of anthocyanins, have greater effects on lipid concentrations in obese/overweight individuals (BMI >25 kg/m^2), individuals with cardiovascular risk factors, those ≥50 years, or who have metabolic syndrome compared to healthy individuals.[180,249,250] Future studies with a larger sample size and longitudinal design are warranted, while limiting the possibility of reverse causality.[223] Moreover, future randomized controlled trials should consider assessing dietary anthocyanin intake with biomarker assessment in addition to a validated FFQ or dietary recall. In line with our findings across a diverse selection of lipid parameters, future studies may also consider focusing specifically on measurements of total cholesterol and HDL cholesterol, which have proven sufficient to capture the lipid-associated risk in CVD prediction.[251] Future research in this line conducted among a large representative population with the inclusion of additive interaction analysis could be instrumental for targeting narrower CVD risk subgroups and offer greater efficacy among behavior change interventions for the promotion of ideal cardiovascular health.

MEDLIFE analyses

As evidenced by our present findings, the MEDLIFE index is a suitable tool intended for nutritional epidemiology to study the healthy traditional Mediterranean lifestyle as a general manner of living, beyond diet and PA.[123] Future research may consider some additional components of the Mediterranean lifestyle such as culinary techniques; conviviality and wine consumption during meals, i.e. the MADP; use of biodiverse, local, and seasonal products; and limited consumption of ultraprocessed foods.[72] Smoking, although frequently considered a risk factor, is a lifestyle habit that has shown an association with mortality stronger than any other low-risk factor.[95] Additional lifestyle factors for an even greater comprehensive lifestyle assessment include financial stability, time in nature, pet ownership, materialistic values, stress management, and those closely tied to social connections, gratitude, forgiveness, confidence, and self-esteem, such as substance abuse, problematic internet, social media, gaming, and smartphone use.[252,253]

Future studies with a greater sample size than "Feeding America's Bravest" should also include greater sociodemographic (i.e. geographical, ethnic, gender, etc.) and cultural diversity, including the roles of extended and immediate family on lifestyle behaviors. In addition, longer follow-up and ample data collection should allow using repeated measures of MEDLIFE to avoid reverse causation and better understand the temporal sequence of potentially bidirectional associations.[217] Other studies on metabolic syndrome in particular should consider measures of visceral adiposity or body composition and explore the role of metabolically healthy obese. While most of the emphasis of public health has been focused on the main causes of death with life-saving interventions, disability becomes an increasingly large component of disease burden and health expenditure. More research is needed to identify new and more effective evidence-based lifestyle education programs and interventions strategically targeted at individuals at high risk of chronic diseases.[254–256]

With a rapidly ageing global population, sedentary lifestyle, poor diet quality, reduced quality of life, and rising obesity rates, the demands on health systems to address disability, which increases with age, will require policy makers to anticipate these changes.[7] Holistic lifestyle medicine programs are already pointed in the right direction to improve patients' well-being, confidence, motivation, importance, and health perceptions to make healthier lifestyle choices through group consultations. Such strategies at a public health level offer greater cost and time efficiency, in addition to efficacy.[254] The "Fifty-Fifty Trial", a one-year peer-group-based intervention, has evidenced the efficacy of health-expert-led workshops focused on both lifestyle behavior change and risk factor education related to motivation to change, the health benefits of PA, healthy diet, smoking cessation, stress management, and self-control of blood pressure.[257] Moreover, lifestyle exposures should be studied within the context of positive epidemiology for a more holistic picture of the distribution and determination of population health.[2] Currently, epidemiologic research focuses on disease and risk factors for disease, neglecting positive health assets as a broader range of health.

A final application includes the estimation of measures of impact, such as the risk difference, joint effect APs, and the population attributable fraction, associated with the Mediterranean and other traditional lifestyle patterns. For instance, in 2008 Van Dam et al. estimated within the “Nurses' Health Study” the population attributable risks of mortality were 28% for cigarette smoking, 14% for being overweight, 17% for lack of PA (<30 minutes of PA per day), and 13% for low diet quality (scored <40 percentile of the cohort for a healthy eating index (HEI)), meanwhile 55% of total deaths were attributable to the combination of smoking, being overweight, lack of PA and a low diet quality. The investigators concluded their results indicated that diet, exercise and other lifestyle factors have additive influences on the risk of premature mortality.[109] More studies of this nature complemented with an interaction analysis should be conducted to pursue a variety of effective lifestyle prevention strategies across culturally different populations. Other absolute measures of population impact, such as cardiometabolic disease-free life expectancy[38] and cost-per-QALY[254] may provide even more useful findings for stakeholders and policy makers regarding primordial prevention strategies for the reduction of chronic disease burden and premature mortality worldwide.

CONCLUSIONS

1. The joint association of low adherence to the MedDiet and low level of a physically active lifestyle was associated with an increased risk of all-cause mortality greater than the sum of the individual risk factors, suggesting a potential interaction or synergism between both exposures, most likely beyond additivity but below multiplicativity, with 36% of the joint effect due to their interaction, in the SUN cohort.

2. Among US career firefighters, anthocyanins and PA were both inversely associated with the ratio of total cholesterol:HDL cholesterol, whereas only PA was inversely associated with triglycerides (TG), LDL cholesterol:HDL cholesterol, and TG:HDL cholesterol. Furthermore, the combined effect of a low anthocyanin intake and low PA more than doubled the relative risk of having low HDL cholesterol (<40 mg/dL), compared to the reference category of high anthocyanins/high activity joint exposure, although no statistically significant interaction was observed.

3. In a study population of Spanish university graduates ≥50 years of age, a higher adherence to the Mediterranean lifestyle, as measured by the MEDLIFE index encompassing food consumption, dietary habits, PA, rest, social habits, and conviviality, relatively reduced the risk of all-cause mortality by 41% compared to lower adherence.

4. Among Spanish university graduates, those with medium MEDLIFE adherence showed a statistically significant decreased relative risk for incident depression compared to participants with poor MEDLIFE adherence. Although greater MEDLIFE adherence did not show any significant association with depression, the decreased relative risk was not greater than that observed for medium adherence, suggesting an L-shaped dose-response curve between the Mediterranean lifestyle and incident depression.

5. In a cross-sectional study with US career firefighters, those with better adherence to the MEDLIFE index exhibited 71% relatively lower odds of metabolic syndrome compared to participants with poorer adherence. The MEDLIFE index was inversely associated with abdominal obesity and hypertriglyceridemia, as well as total cholesterol, LDL cholesterol, and total cholesterol:HDL cholesterol, suggesting a more favorable lipid profile.

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APPENDIX

1 - SUN cohort

Published Supplementary Material:

2 - Feeding America's Bravest

Published Supplementary Material:

1.A. Invitation to enroll in the "Seguimiento Universidad de Navarra" study

Universidad de Navarra
Facultad de Medicina
Departamento de Medicina Preventiva
y Salud Pública

PROYECTO
SUN

Pamplona, 11 de febrero de 2020

Estimada:

Tras tu paso por la universidad, te invitamos a participar en el Proyecto SUN (Seguimiento Universidad de Navarra), un proyecto que ya ha cumplido 20 años, pues se inició en 1999 en el departamento de Medicina Preventiva y Salud Pública de la Universidad de Navarra, y en el que colaboran profesores de Harvard T.H. Chan School of Public Health y otras universidades con experiencia en estudios similares.

Te adjuntamos un folleto con toda la información sobre el proyecto. Como verás, este proyecto ha permitido que desde la Universidad de Navarra se contribuya -con liderazgo científico y gran presencia internacional- a mejorar muchos aspectos relevantes de la dieta y el estilo de vida y así lograr beneficios para la salud pública. Si quieres ayudarnos formando parte de este estudio, tu participación altruista y voluntaria consistiría en rellenar el cuestionario que te adjuntamos y devolvérnoslo usando el sobre de franqueo en destino (sin coste adicional). Cada dos años recibirás un cuestionario de seguimiento pero mucho más breve. Por eso, te rogamos que cumplimentes la hoja donde te solicitamos los datos de contacto puesto que solo así podremos ponernos en contacto contigo*. Quizás te resulte más cómodo contestar el cuestionario a través de Internet. Para ello, puedes solicitarnos en sun@unav.es tu contraseña personal indicándonos: nombre, apellidos, fecha de nacimiento, dirección postal y el n? de identificación que está en la parte superior del cuestionario. Para más información puedes consultar la página web: www.proyectosun.es. Al participar en este estudio, además de recibir nuestra gratitud personal, has de saber que estarás colaborando a mejorar la salud de nuestra sociedad.

Agradeciéndote de antemano tu tiempo, atención y colaboración, recibe un cordial saludo,

Dr. Miguel Ángel Martínez-González D. Sergi Molas Giner en nombre de todo el equipo de investigación del Proyecto SUN Director de Alumni Universidad de Navarra Director del Dpto. Medicina Preventiva y Salud Pública

* El Responsable del Tratamiento de los datos Universidad de Navarra, en cumplimiento del Reglamento (UE) 2016/679 del Parlamento Europeo y del Consejo, de 27 de abril de 2016, relativo a la protecciÓn de las personas fisicas en lo que respecta al tratamiento de datos personales y a la libre circulación de éstos, en

adelante RGPD, le informa que si participa en este Estudio sus datos serán tratados por el equipo investigador para extraer conclusiones del tratamiento empleado. También podrán acceder a los datos las autoridades sanitarias y los miembros del comité ético si lo considerasen necesario. Todos los datos personales incluidos los clínicos serán tratados conforme a las leyes actuales de protección de datos, especialmente conforme al RGPD. No será poslble identificarle a usted a través de las comunicaciones que pudiera generar este estudio. Usted es el responsable de la veracidad y corrección de los datos que nos entrega y tiene la facultad de ejercer los derechos de acceso, rectificación, supresión, limitación del tratamiento, portabflidad y de oposición de sus datos de acuerdo con lo dispuesto en la normativa en materia de protección de datos. Para ejercerlos, deberá dirigirse por escrito al Delegado de Protección de Datos de la Universidad de Navarra a la siguiente dirección postal Irunlarrea 1, 31008 Pamplona, Navarra (Universidad de Navarra) 0 a la dirección de correo electrÓnico sun@unav.es, en cualquier caso deberá adjuntar una fotocopia de Su documento nacional de identidad 0 equivalente.
Asimismo, se le informa su derecho a, en caso de no estar de acuerdo con el tratamiento realizado por nuestra Entidad 0 considerar vulnerados sus derechos, presentar una reclamación en todo momento ante la Agencia Española de Protección de datos.

Irunlarrea 1. 31080 Pamplona. España Tel. +34 948 425 600 Ext 806511 • 9717 716 511
Pueden ampliar la información acerca del proyecto SUN a través de sun@unav.es y www.proyectosun.es

1.B. Personal data sheet

Datos Personales

3485552

Estos son los datos que tenemos de usted:

- Si son correctos, NO tiene que hacer nada
- Si ha habido cambios, rellene y reenvie

Luis Javier

Sánchez

Soto

1 687 111 222

2 91 1111 222

LJSanchezSoto@gmail.com

1 C/ Jose Ortega 7 5B. Túnez. 38877

2 Sin datos. No rellene si no es necesario.

3 Sin datos. No rellene si no es necesario.

Proyecto SUN

1.C. Baseline questionnaire

1. Sexo

- Varón ...
- Mujer ...

2. Ciudad donde vives

Código postal ciudad donde vives

0 1 2 3 4 5 6 7 8 9

¿Desde hace cuántos años?

0 1 2 3 4 5 6 7 8 9

3. Fecha de Nacimiento

Día Mes Año

4. Peso actual (Kg.)

5. Talla (altura) actual (cm.)

6. ¿Has cambiado de peso en los últimos 5 años?

- No he cambiado de peso
- Perdí peso: 1 - 2 Kg
- Perdí peso: 3 - 4 Kg
- Perdí peso: 5 - 10 Kg
- Perdí peso: > 10 Kg
- Gané peso: 1 - 2 Kg
- Gané peso: 3 - 4 Kg
- Gané peso: 5 - 10 Kg
- Gané peso: > 10 Kg
- He ganado peso por un embarazo

7. Estado civil:

- Soltero/a
- Casado/a
- Viudo/a
- Separado/a
- Otros

(Dobla por esta línea)

8. Número de hijos:

Ninguno 1 2 3 4 5 6 7 8 9 10 ó más

9. ¿Cuántas personas en total viven actualmente en tu hogar incluyéndote tú?

1 2 3 4 5 6 7 8 9 10 ó más

10. ¿Cuál es el nivel más alto de estudios que has completado?

- No he acabado ninguna diplomatura ni licenciatura
- Doctorado
- Master
- Licenciatura - Escuela técnica superior
- Diplomatura (ingeniería técnica - escuelas universitarias)

¿Has terminado alguna de estas carreras?

- Medicina
- Enfermería
- Farmacia
- Dietética
- Biológicas
- Otra carrera biosanitaria
- Ninguna carrera biosanitaria

11. ¿Cuál es tu situación laboral?

- Trabajo tiempo completo
- A tiempo parcial
- Ama de casa
- Paro
- Jubilado/a
- Estudiante

12. ¿Has fumado 100 cigarrillos o más en toda tu vida?

- No (Pasa a pregunta 13)
- Sí, y sigo fumando
- Sí, pero ya no fumo

N.º cigarrillos / día en promedio fumados a cada edad

Cigarrillos / día →	Ning.	1-4	5-14	15-24	25-34	35-44	45 +
< 15 años							
15 - 19 años							
20 - 29 años							
30 - 39 años							
40 - 49 años							
50 - 59 años							
60 + años							

¿Cuánto hace que dejaste completamente de fumar?

- < 1 año
- 1 - 2 años
- 3 - 5 años
- 6 - 9 años
- 10 + años

MARCA DE CIGARRILLOS (Ejemplo: Marlboro Lights)

No debes marcar esta zona sombreada

10 20 30 40 50 60 / 1 2 3 4 5 6 7 8 9

SUMCO 9416-6-K (1)

Página 1

ESTE N.º DEBERÁS MARCARLO EN SU COLUMNA CORRESPONDIENTE

marca así — así no marques

13. ¿Fumas actualmente en pipa o fumas puros?

- SÍ
- NO

14. ¿Has convivido HABITUALMENTE con algún fumador?. Si es así, señala, por favor, los años en que has estado pasivamente expuesto al humo "de segunda mano" del tabaco y el número aproximado de horas en que has estado expuesto cada día por término medio.

En el hogar: exposición pasiva

Nunca he estado expuesto	Años de exposición	Horas al día expuesto
Esposo/a fumador/a	< 1 año	< 1 hora
	1 - 2 años	1 - 2 horas
	3 - 5 años	3 - 5 horas
	6 - 9 años	6 - 9 horas
	10 + años	10 + horas
Otro/s fumador/es en casa	< 1 año	< 1 horas
	1 - 2 años	1 - 2 horas
	3 - 5 años	3 - 5 horas
	6 - 9 años	6 - 9 horas
	10 + años	10 + horas

En el trabajo: exposición pasiva

Nunca he estado expuesto	Años de exposición	Horas al día expuesto
Un sólo colega fumaba en la misma habitación	< 1 año	< 1 hora
	1 - 2 años	1 - 2 horas
	3 - 5 años	3 - 5 horas
	6 - 9 años	6 - 9 horas
	10 + años	10 + horas
Varios colegas fumaban en la misma habitación	< 1 año	< 1 horas
	1 - 2 años	1 - 2 horas
	3 - 5 años	3 - 5 horas
	6 - 9 años	6 - 9 horas
	10 + años	10 + horas

15. Por término medio en una semana típica, ¿cuántos días/semana bebes alcohol (vino, cerveza o licores destilados), incluyendo el que tomas en las comidas?

Nunca o casi nunca 1 2 3 4 5 6 7 días

16. ¿Cuántos días/semana bebes vino en la comida?

Nunca o casi nunca 1 2 3 4 5 6 7 días

17. ¿Cuál fue el máximo número de bebidas alcohólicas (sumando vino, cerveza y licor) que tomaste un día entre semana?

Ninguna 1 - 2 3 - 5 6 - 9 10 - 14 15 ó más

¿Y un día de fin de semana?

Ninguna 1 - 2 3 - 5 6 - 9 10 - 14 15 ó más

¿Y un día especial (celebración, boda, festividad)?

Ninguna 1 - 2 3 - 5 6 - 9 10 - 14 15 ó más

18. ¿Conduces cuando has bebido algo de alcohol?

- No sé conducir
- Sí, a veces
- Casi nunca
- No, absolutamente nunca

19. ¿Usas cuando vas en un coche?

Cinturón de seguridad: Sí / No siempre / Casi nunca

Bolsa de aire (airbag): Sí / No

20. Cuando haces ejercicio o deporte siguiendo tu modo típico de hacerlo, ¿cuál crees que es tu grado de intensidad en el esfuerzo?. Puntúalo de 0 (el mínimo posible) a 10 (el máximo).

- ☐ Nunca hago deporte
- ☐ 0 ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10

21. Habitualmente, ¿cuánto tiempo andas al día?

- ☐ < 10 minutos
- ☐ 21 - 30 minutos
- ☐ 1 - 2 horas
- ☐ 10 - 20 minutos
- ☐ 1/2 - 1 hora
- ☐ > 2 horas

22. Tu paso habitual al andar por la calle es…

- ☐ Lento
- ☐ Normal, medio
- ☐ Rápido
- ☐ Muy rápido

23. ¿Cuántos pisos subes al día por escaleras en total?

- ☐ 2 ó menos
- ☐ 3 - 4
- ☐ 5 - 9
- ☐ 10 - 14
- ☐ 15 ó más

24. Por término medio, ¿cuántos kilómetros haces al año en coche, ya sea conduciendo tú o conduciendo otro?

- ☐ < 1.000
- ☐ 10.001 - 20.000
- ☐ > 50.000
- ☐ 1.001 - 10.000
- ☐ 20.001 - 50.000

¿Y en moto?

- ☐ Ninguno
- ☐ 1.000 - 5.000
- ☐ > 10.000
- ☐ < 1.000
- ☐ 5.001 - 10.000

25. Nivel de colesterol (mg/dl) (sólo análisis hechos hace < 5 años)

- ☐ No me he hecho análisis
- ☐ No recuerdo
- ☐ < 180 (bajo)
- ☐ 201 - 240 (algo alto)
- ☐ > 300 (muy alto)
- ☐ 180 - 200 (normal)
- ☐ 241 - 300 (alto)

¿Y de HDL (mg/dl)?

- ☐ No me he hecho análisis
- ☐ No recuerdo
- ☐ < 35 (bajo)
- ☐ 36 - 60 (normal)
- ☐ > 60 (alto)

26. Pulso en reposo (latidos/minuto, frecuencia cardiaca)

- ☐ < 50
- ☐ 71 - 80
- ☐ 101 - 105
- ☐ 50 - 60
- ☐ 81 - 90
- ☐ 106 - 110
- ☐ 61 - 70
- ☐ 91 - 100
- ☐ > 110

27. Medicación actual. Marcar el uso HABITUAL:

- ☐ No tomo ninguna medicación habitualmente
- ☐ Aspirina ≥ 2 veces/semana
- ☐ Otros analgésicos
- ☐ Reductores de colesterol
- ☐ Insulina
- ☐ Antidiabéticos orales
- ☐ Digoxina-digitálicos
- ☐ Diuréticos
- ☐ Beta-bloqueantes (Sumial, Tenormin…)
- ☐ Antagonistas del Calcio (Adalat, Manidón…)
- ☐ Nitritos (parches, Isoket, cafinitrina…)
- ☐ Otros antihipertensivos
- ☐ Para controlar el peso
- ☐ Antidepresivos
- ☐ Tranquilizantes o inductores del sueño
- ☐ Otros

Por favor, si tomas habitualmente otra medicación, adjunta DOSIS, FRECUENCIA Y DURACIÓN en un papel aparte.

28. ¿Haces ejercicio?

- ☐ No
- ☐ Sí

29. ¿Cuánto tiempo por término medio dedicaste a las siguientes actividades en el último año?

	FRECUENCIA MEDIA DURANTE LA SEMANA										MESES AL AÑO		
	NUNCA	MINUTOS / SEMANA			HORAS / SEMANA								
		1 - 4	5 - 19	20 - 59	< 1	1 - 1,5	2 - 3	4 - 6	7 - 10	≥ 11	< 3	3 - 6	> 6
Andar o pasear fuera de casa (incluye golf)	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Correr o hacer jogging despacio	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Correr más competitivo y rápido (atletismo, etc.)	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Pasear en bicicleta	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Bicicleta estática	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Nadar	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Tenis, frontón, squash, otros de raqueta o pala	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Fútbol, futbito	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Otros de equipo (baloncesto, balonmano…)	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Baile, danza, aerobic	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Excursiones al monte, escalada	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Gimnasia	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Cuidado del jardín y/o piscina, bricolaje, etc.	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Esquí, patinaje	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Judo, karate u otras artes marciales	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Vela	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐
Otras actividades físicas-deporte no mencionadas	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐	☐

30. Tiempo por término medio en las siguientes actividades en el último año. Distingue y contesta ENTRE SEMANA y FIN DE SEMANA

DÍA TÍPICO DE TRABAJO ENTRE SEMANA

TIEMPO AL DÍA	NUNCA	< 30 MIN.	30 - 60 MIN.	HORAS / DÍA								
Ver televisión-video	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Sentado ante pantalla ordenador	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Conduciendo	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Estar sentado (en total)	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Dormir por las noches	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Dormir la siesta	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Tomando el sol (verano)	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Tomando el sol (invierno)	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Salir con los amigos	☐	☐	☐	1	2	3	4	5	6	7	8	9+
De pie en el trabajo	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Tareas domésticas	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Actividad en el trabajo más intensa que estar de pie	☐	☐	☐	1	2	3	4	5	6	7	8	9+

DÍA TÍPICO DE FIN DE SEMANA

TIEMPO AL DÍA	NUNCA	< 30 MIN.	30 - 60 MIN.	HORAS / DÍA								
Ver televisión-video	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Sentado ante pantalla ordenador	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Conduciendo	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Estar sentado (en total)	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Dormir por las noches	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Dormir la siesta	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Tomando el sol (verano)	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Tomando el sol (invierno)	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Salir con los amigos	☐	☐	☐	1	2	3	4	5	6	7	8	9+
De pie en el trabajo	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Tareas domésticas	☐	☐	☐	1	2	3	4	5	6	7	8	9+
Actividad en el trabajo más intensa que estar de pie	☐	☐	☐	1	2	3	4	5	6	7	8	9+

31. Tensión arterial actual (mmHg) (sólo si fue tomada hace < 2 años)

Sistólica (máxima)

- No me la he tomado
- Me la tomaron, pero no recuerdo

< 100	111 - 120	131 - 140	151 - 160	> 175
101 - 110	121 - 130	141 - 150	161 - 175	

Diastólica (mínima)

< 60	71 - 80	91 - 100	111 - 120	> 130
61 - 70	81 - 90	101 - 110	121 - 130	

32. ¿Te has sometido a alguna de las siguientes exploraciones o intervenciones preventivas, SIN NECESITAR DE DICHAS PRUEBAS POR ENFERMEDAD?. Señala cada vez que se ha realizado la intervención (edad al realizarla)

INTERVENCIÓN	EDAD (AÑOS) AL REALIZARLA				
	NUNCA	< 25	25 - 44	45 - 64	≥ 65
Revisión médica general					
Electrocardiograma					
Prueba de esfuerzo coronaria					
Radiografía de tórax					
Sangre oculta en heces					
Colonoscopia/Sigmoidoscopia					
Revisión dental					
Presión intraocular					
(Sólo mujeres) Citología cuello uterino (Papanicolau)					
(Sólo mujeres) Mamografía					
(Sólo para varones) Detección cáncer de próstata:					
Tacto rectal					
Ecografía					
Antígeno prostático					

(Dobla por esta línea)

33. ¿Algún profesional te ha diagnosticado alguna vez alguna de las siguientes enfermedades?.

ENFERMEDAD	EDAD (AÑOS) AL DIAGNÓSTICO				
	NUNCA	< 25	25 - 44	45 - 64	≥ 65
Diabetes					
Hipertensión					
Colesterol alto					
Triglicéridos altos					
Infarto de miocardio					
Angina de pecho					
Cirugía coronaria ("by-pass")					
Angioplastia coronaria					
Accidente cerebro-vascular (trombosis-embolia-hemorragia cerebral)					
Taquicardia paroxística					
Fibrilación auricular					
Aneurisma de aorta					
Insuficiencia cardiaca					
Embolia pulmonar					
Trombosis venosa periférica					
Claudicación intermitente (insuficiencia arterial periférica)					
Accidente de tráfico con fractura u hospitalización de > 24 horas					
Fractura de cadera					
Artritis reumatoide					
Pólipos en colon o recto					
Úlcera gástrica o duodenal					
Asma bronquial					
Bronquitis crónica-Enfisema					

SUMCO

Página 3

En este cuadro debes repetir en las 6 casillas superiores el número que figura en el cuadro de la 1ª página y a continuación marcarlo igual que lo has hecho anteriormente.

marca así — así no marques

NÚMERO

0	0	0	0	0	0
1	1	1	1	1	1
2	2	2	2	2	2
3	3	3	3	3	3
4	4	4	4	4	4
5	5	5	5	5	5
6	6	6	6	6	6
7	7	7	7	7	7
8	8	8	8	8	8
9	9	9	9	9	9

(Continúa pregunta 33)

ENFERMEDAD	EDAD (AÑOS) AL DIAGNÓSTICO				
	NUNCA	< 25	25 - 44	45 - 64	≥ 65
Cálculos (piedras) en la vesícula					
Cálculos renales o cólico nefrítico					
Depresión					
Cataratas					
Obesidad					
Apnea del sueño					
Cáncer o tumores (señala el tipo)					
Otras (1)					
Otras (2)					

No debes marcar estas tres zonas sombreadas

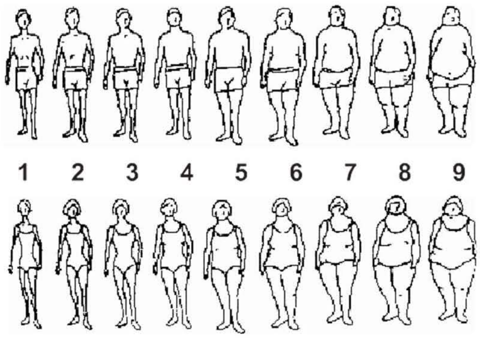
	CÓDIGO									
Cáncer o tumores	0	1	2	3	4	5	6	7	8	9
	0	1	2	3	4	5	6	7	8	9
Otras (1)	0	1	2	3	4	5	6	7	8	9
	0	1	2	3	4	5	6	7	8	9
Otras (2)	0	1	2	3	4	5	6	7	8	9
	0	1	2	3	4	5	6	7	8	9

34. ¿Padeció algún pariente tuyo alguna de las siguientes enfermedades?

ENFERMEDAD		EDAD (AÑOS) AL DIAGNÓSTICO				
		NUNCA	< 25	25 - 44	45 - 64	≥ 65
Infarto de miocardio/muerte súbita cardiaca	Padre					
	Madre					
Cáncer de mama	Madre					
	Hermana					
	Abuela materna					
	Abuela paterna					

ENFERMEDAD	NINGUNO	PADRE	MADRE	HERMANO
Hipertensión				
Diabetes				
Melanoma				
Cáncer de pulmón				
Cáncer de colon o recto				
Pólipos en colon o recto				
Obesidad				

35. ¿Qué diagrama representa mejor cómo era tu silueta corporal a cada edad?



FIGURAS →	1	2	3	4	5	6	7	8	9
Actualidad									
A los 5 años									
A los 20 años									
A los 30 años									
A los 40 años									

36. Habitualmente cuando tomas el sol, ¿usas cremas protectoras?

No tomo el sol · Sí · No

37. En la niñez o adolescencia, después de tomar el sol durante un rato largo (>2 horas) sin crema protectora, ¿qué tipo de reacción tenías en la piel?

Prácticamente ninguna · Sólo un ligero enrojecimiento · Quemarme · Quemaduras graves, dolorosas · Quemaduras graves, dolorosas, con ampollas

38. Entre los 15 y los 20 años ¿cuántas veces tuviste quemaduras graves por el sol, con ampollas?

Nunca · 1 Vez · 2 Veces · 3-4 Veces · 5-9 Veces · 10 + Veces

39. Por favor, ¿puedes contar cuántos lunares tienes desde las rodillas hasta los tobillos, sumando ambas piernas?

Me resulta incómodo contarlos · Ninguno · 1-2 · 3-5 · 6-9 · 10-14 · 15-20 · ≥ 21

40. ¿Te consideras una persona competitiva, inconformista, luchadora, que se exige todo lo que puede en su trabajo, incluso se pide más de lo que puede? Puntúate de 0 (lo más conformista) a 10 (lo más competitivo).

Conformista 0 1 2 3 4 5 6 7 8 9 10 Competitivo

41. ¿Te consideras una persona tensa, agresiva, que se preocupa demasiado de las cosas, o eres una persona que suele estar relajada y tranquila? Puntúate de 0 (lo más relajado) a 10 (lo más tenso).

Relajado 0 1 2 3 4 5 6 7 8 9 10 Tenso

42. ¿Te consideras con suficientes recursos, preparación y autonomía para resolver los problemas que se plantean en tu trabajo, o dependes exclusivamente de otros para ello?. Puntúate de 0 (lo más autónomo) a 10 (lo más dependiente).

Autonomía 0 1 2 3 4 5 6 7 8 9 10 Dependencia

43. Excluyendo tareas domésticas, ¿cuántas horas trabajas a la semana?

No trabajo · < 20 · 20-24 · 25-29 · 30-34 · 35-39 · 40-44 · 45-49 · 50-54 · 55-59 · 60-64 · 65-69 · 70-74 · 75-79 · 80-84 · ≥ 85

44. ¿Cuántos días a la semana vas a comer a casa al medio día?

0 1 2 3 4 5 6 7

45. ¿Padeces o has padecido alguna vez insomnio?

Nunca · Rara vez · Sí, y sigo padeciéndolo · Sí, anteriormente, pero ya no lo padezco

46. ¿Roncas por la noche?

No lo sé · Nunca · Rara vez · Sí

LAS PREGUNTAS 47 A 54 SÓLO DEBEN CONTESTARLAS LAS MUJERES (Zona sombreada suave)

47. Edad de la primera regla

AÑOS: 0 1 2 / 0 1 2 3 4 5 6 7 8 9

48. Si han desaparecido las reglas. ¿A qué edad desaparecieron?

No han desaparecido, sigo teniéndolas

AÑOS: 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

¿Cuál fue la causa de cesar las reglas?

Natural · Cirugía de órganos → EXTIRPACIÓN: Útero y ovarios · Útero nada más · Ovarios nada más · Quimioterapia o Radioterapia · Otras

No debes marcar esta zona sombreada oscura

CÓDIGO: 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

49. ¿Has tomado alguna vez terapia sustitutiva para la menopausia con estrógenos (hormonas sexuales)?

Nunca · Anteriormente · Ahora

Si las tomaste o las tomas actualmente:

¿Durante cuánto tiempo (años)? <1 1 2 3 4 5 6 7 8 9 ó más

Por favor, especifica la marca

No debes marcar esta zona sombreada oscura

CÓDIGO: 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

50. ¿Has sido diagnosticada de enfermedad fibroquística mamaria u otra enfermedad benigna de la mama?

No · Sí → ¿Se confirmó por biopsia? → No · Sí

51. Número de embarazos

Ninguno · 1 2 3 4 5 6 7 8 9 ó más

(Pasa a la página siguiente)

52. Embarazos múltiples (mellizos/gemelos)

No · Sí → Edad a la que lo tuviste:

AÑOS: 1 2 3 4 5 6 / 0 1 2 3 4 5 6 7 8 9

53. Edad de los embarazos:
marca para cada año de edad si se completó un embarazo de 6 ó más meses a esa edad, marca en la otra columna si fue un embarazo de menos de 6 meses, incluyendo pérdidas fetales y abortos.

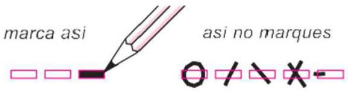
Edad (años)	≥ 6 meses	< 6 meses	Edad (años)	≥ 6 meses	< 6 meses	Edad (años)	≥ 6 meses	< 6 meses
≥15			27			39		
16			28			40		
17			29			41		
18			30			42		
19			31			43		
20			32			44		
21			33			45		
22			34			46		
23			35			47		
24			36			48		
25			37			≥49		
26			38					

54. Como promedio, ¿cuánto ha durado la lactancia materna de tus hijos?

Nada · < 1 mes · 1-3 meses · > 3 meses

SUMCO

NÚMERO



En este cuadro debes repetir en las 6 casillas superiores el número que figura en el cuadro de la 1ª página y a continuación marcarlo igual que lo has hecho anteriormente.

0	0	0	0	0	0
1	1	1	1	1	1
2	2	2	2	2	2
3	3	3	3	3	3
4	4	4	4	4	4
5	5	5	5	5	5
6	6	6	6	6	6
7	7	7	7	7	7
8	8	8	8	8	8
9	9	9	9	9	9

ENCUESTA DIETÉTICA

Por favor, marca una única opción para cada alimento.

Para cada alimento, marca el recuadro que indica la frecuencia de consumo **por término medio** durante el **año pasado**. Se trata de tener en cuenta también la variación verano/invierno. Por ejemplo si tomas helados 4 veces/semana sólo durante los 3 meses de verano, el uso promedio al año es 1/semana

I LACTEOS	CONSUMO MEDIO DURANTE EL AÑO PASADO								
	NUNCA O CASI NUNCA	AL MES	A LA SEMANA			AL DÍA			
		1 - 3	1	2 - 4	5 - 6	1	2 - 3	4 - 6	6 +
Leche entera (1 taza, 200 cc)									
Leche semidesnatada (1 taza, 200 cc)									
Leche desnatada (1 taza, 200 cc)									
Leche condensada (1 cucharada)									
Nata o crema de leche (1/2 taza)									
Batidos de leche (1 vaso, 200 cc)									
Yogurt entero (1, 125 gr)									
Yogurt descremado (1, 125 gr)									
Petit suisse (1, 100 gr)									
Requesón o cuajada (1/2 taza)									
Queso en porciones o cremoso (1, porción)									
Otros quesos: curados, semicurados (Manchego, Bola, Emmental...) (50 gr)									
Queso blanco o fresco (Burgos, cabra...) (50 gr)									
Natillas, flan, puding (1 taza, 200 cc.)									
Helados (uno)									

(Dobla por esta línea)

Por favor, marca una única opción para cada alimento.

II HUEVOS, CARNES, PESCADOS — Un plato o ración de 100-150 gr, excepto cuando se indica otra cosa	NUNCA O CASI NUNCA	AL MES	A LA SEMANA			AL DÍA			
		1 - 3	1	2 - 4	5 - 6	1	2 - 3	4 - 6	6 +
Huevos de gallina (uno)									
Pollo o pavo CON piel									
Pollo o pavo SIN piel									
Carne de ternera o vaca									
Carne de cerdo									
Carne de cordero									
Conejo o liebre									
Hígado									
Otras vísceras (sesos, corazón, mollejas)									
Jamón serrano o paletilla									
Jamón York, jamón cocido (1 loncha)									
Embutidos (chorizo, salchichón, mortadela, 50 gr)									
Salchichas (50 gr)									
Patés, foie-gras (25 gr)									
Morcilla (50 gr)									
Hamburguesa (unidad)									
Sobrasada (50 gr) / albóndigas (3 unidades)									
Tocino, bacon, panceta (50 gr)									
Pescado blanco: pescadilla, merluza, besugo, mero, lenguado (1 plato, pieza o ración)									
Pescado azul: sardinas, atún, bonito, caballa, salmón (1 plato, pieza o ración)									
Bacalao									
Pescados salados y/o ahumados: arenques, salmón									
Ostras, almejas, mejillones, etc. (6 unidades)									
Gambas, langostinos, cigalas, etc.									
Pulpo, calamares, chipirones, jibia...									

Por favor, marca una única opción para cada alimento.

III VERDURAS Y HORTALIZAS

Un plato o ración de 250 grs, excepto cuando se indica	NUNCA O CASI NUNCA	AL MES 1 - 3	A LA SEMANA 1	A LA SEMANA 2 - 4	A LA SEMANA 5 - 6	AL DÍA 1	AL DÍA 2 - 3	AL DÍA 4 - 6	AL DÍA 6 +
Acelgas, espinacas									
Col, coliflor, brócoles									
Lechuga, endibias, escarola									
Tomate crudo (1, 150 gr)									
Zanahoria, calabaza									
Judías verdes									
Berenjenas, calabacines, pepinos									
Pimientos									
Espárragos									
Gazpacho andaluz									
Otras verduras (borraja, cardo...)									
Patatas fritas (caseras, bolsa, 1 ración, 150 gr)									
Patatas asadas o cocidas (1 ración, 150 gr)									

Por favor, marca una única opción para cada alimento.

IV FRUTAS

Una pieza o ración	NUNCA O CASI NUNCA	AL MES 1 - 3	A LA SEMANA 1	A LA SEMANA 2 - 4	A LA SEMANA 5 - 6	AL DÍA 1	AL DÍA 2 - 3	AL DÍA 4 - 6	AL DÍA 6 +
Naranja, pomelo (una), o mandarina (dos)									
Plátano									
Manzana, pera									
Fresas/fresones (6 unidades, plato postre)									
Melocotón, albaricoque, nectarina									
Cerezas, picotas, ciruelas (1 plato de postre)									
Higos, brevas									
Sandía (1 tajada, 200-250 gr)									
Melón (1 tajada, 200-250 gr)									
Uvas (un racimo, un plato postre)									
Frutas en almíbar (2 unidades)									
Frutas en su jugo (2 unidades)									
Dátiles, higos secos, pasas, ciruelas-pasas (150 gr)									
Almendras, cacahuetes, avellanas, nueces (50 gr)									
Aceitunas (10 unidades)									
Aguacates									
Mangos, papaya									
Kiwi									

	0	1	2	3	4	5	6	7
¿Cuántos días a la semana tomas fruta como postre?								

Por favor, marca una única opción para cada alimento.

V LEGUMBRES Y CEREALES

Un plato o ración de 60 gr en seco	NUNCA O CASI NUNCA	AL MES 1 - 3	A LA SEMANA 1	A LA SEMANA 2 - 4	A LA SEMANA 5 - 6	AL DÍA 1	AL DÍA 2 - 3	AL DÍA 4 - 6	AL DÍA 6 +
Lentejas									
Garbanzos									
Alubias (pintas, blancas o negras)									
Guisantes									
Pan blanco (3 rodajas, 60 gr)									
Pan negro integral (3 rodajas, 60 gr)									
Cereales desayuno (30 gr en seco)									
Arroz blanco (60 gr en seco)									
Pasta: fideos, macarrones, espaguetis (60 gr en seco)									
Pizza (1 ración, 200 gr)									

SUMCD 9418-9-4 (Ret. 3)

No debes marcar estas 4 zonas sombreadas

CÓDIGO											
	0	1	2	3	4	5	6	7	8	9	(1)
	0	1	2	3	4	5	6	7	8	9	
	0	1	2	3	4	5	6	7	8	9	(2)
	0	1	2	3	4	5	6	7	8	9	
	0	1	2	3	4	5	6	7	8	9	(3)
	0	1	2	3	4	5	6	7	8	9	
	0	1	2	3	4	5	6	7	8	9	(4)
	0	1	2	3	4	5	6	7	8	9	

Página 7

En este cuadro debes repetir en las 6 casillas superiores el número que figura en el cuadro de la 1ª página y a continuación marcarlo igual que lo has hecho anteriormente.

marca así — así no marques

NÚMERO

0	0	0	0	0	0
1	1	1	1	1	1
2	2	2	2	2	2
3	3	3	3	3	3
4	4	4	4	4	4
5	5	5	5	5	5
6	6	6	6	6	6
7	7	7	7	7	7
8	8	8	8	8	8
9	9	9	9	9	9

Por favor, marca una única opción para cada alimento.

VI ACEITES Y GRASAS

CONSUMO MEDIO DURANTE EL AÑO PASADO

Una cucharada o porción individual *Para untar, mojar en el pan, para aliñar, o para ensaladas, utilizas en total.*	NUNCA O CASI NUNCA	AL MES	A LA SEMANA			AL DÍA			
		1 - 3	1	2 - 4	5 - 6	1	2 - 3	4 - 6	6 +
Mantequilla (porción individual)									
Margarina (porción individual)									
Aceite de oliva (una cucharada)									
Aceite de girasol									
Aceite de maíz									
Manteca de cerdo									
Otros: (1)									
¿CON QUÉ FRECUENCIA CONSUMES?:									
Alimentos fritos en casa									
Alimentos fritos fuera de casa									

EN TU CASA, PARA FREIR SE UTILIZA:

Aceite de oliva	Mantequilla
Aceite de girasol	Margarina
Aceite de maíz	Otros (2)

Marca de aceite de oliva que usas habitualmente (3)

(Dobla por esta línea)

Por favor, marca una única opción para cada alimento.

VII BOLLERÍA Y PASTELERÍA

CONSUMO MEDIO DURANTE EL AÑO PASADO

	NUNCA O CASI NUNCA	AL MES	A LA SEMANA			AL DÍA			
		1 - 3	1	2 - 4	5 - 6	1	2 - 3	4 - 6	6 +
Galletas tipo María (4-6 unidades, 50 gr)									
Galletas con chocolate (4-6 unidades, 50 gr)									
Magdalenas comerciales (1-2 unidades)									
Donuts (uno)									
Ensaimada, croissant u otra bollería **industrial comercial**... (uno, 50 gr)									
Bollería, repostería **casera**									
Pasteles (uno, 50 gr)									
Churros, porras y similares (ración, 100 gr)									
Chocolates y bombones (30 gr)									
Turrón (1/8 de barra)									
Pastas de té, mantecados, mazapán (ración, 90 gr)									

Por favor, marca una única opción para cada alimento.

VIII BEBIDAS

Vaso de vino tinto									
Vaso de otro tipo de vino									
Vaso de vino en las comidas									
Cerveza (1 jarra, 330 cc)									
Licores, destilados: whisky, ginebra, coñac, anís... (1 copa, 50 cc)									
Bebidas carbonatadas con azúcar: Coca-Cola, naranjadas... (1 botellín, 200 cc)									
Idem, pero bajas en calorías, Bebidas light... (1 botellín, 200 cc)									
Zumo de naranja natural (1 vaso, 200 cc)									
Zumos naturales de otras frutas (1 vaso, 200 cc)									
Zumos de frutas o de verduras en botella o enlatados (200 cc)									
Café descafeinado (1 taza, 50 cc)									
Café (1 taza, 50 cc)									
Agua (del grifo) (1 vaso, 200 cc)									
Agua embotellada (1 vaso, 200 cc)									

Marca de agua embotellada que bebes más habitualmente (4)

SUMCO 9416-9-K (4)

Por favor, marca una única opción para cada alimento.

IX MISCELÁNEA

	CONSUMO MEDIO DURANTE EL AÑO PASADO								
	NUNCA O CASI NUNCA	AL MES	A LA SEMANA			AL DÍA			
		1 - 3	1	2 - 4	5 - 6	1	2 - 3	4 - 6	6 +
Croquetas, buñuelos, empanadillas	☐	☐	☐	☐	☐	☐	☐	☐	☐
Sopas y cremas de sobre	☐	☐	☐	☐	☐	☐	☐	☐	☐
Salsa de tomate frito, ketchup (1 cucharadita)	☐	☐	☐	☐	☐	☐	☐	☐	☐
Mayonesa (1 cucharadita)	☐	☐	☐	☐	☐	☐	☐	☐	☐
Picante: tabasco, pimienta	☐	☐	☐	☐	☐	☐	☐	☐	☐
Sal (una pizca)	☐	☐	☐	☐	☐	☐	☐	☐	☐
Azúcar (1 cucharadita)	☐	☐	☐	☐	☐	☐	☐	☐	☐
Sacarina	☐	☐	☐	☐	☐	☐	☐	☐	☐
Mermeladas (1 cucharadita)	☐	☐	☐	☐	☐	☐	☐	☐	☐
Miel	☐	☐	☐	☐	☐	☐	☐	☐	☐
Otros alimentos de frecuente consumo:									
(1) ________	☐	☐	☐	☐	☐	☐	☐	☐	☐
(2) ________	☐	☐	☐	☐	☐	☐	☐	☐	☐
¿Con qué frecuencia haces comidas fuera de casa?	☐	☐	☐	☐	☐	☐	☐	☐	☐

(1) No debes marcar esta zona sombreada — CÓDIGO: 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

(2) No debes marcar esta zona sombreada — CÓDIGO: 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

¿Tomaste vitaminas y/o minerales (incluyendo calcio) habitualmente durante el año pasado?

☐ No ☐ Sí

Si las tomaste, por favor indica la marca:

Marcas de los suplementos de vitaminas o minerales	NUNCA O CASI NUNCA	AL MES	A LA SEMANA			AL DÍA			
		1 - 3	1	2 - 4	5 - 6	1	2 - 3	4 - 6	6 +
(1) ________	☐	☐	☐	☐	☐	☐	☐	☐	☐
(2) ________	☐	☐	☐	☐	☐	☐	☐	☐	☐
(3) ________	☐	☐	☐	☐	☐	☐	☐	☐	☐

(1) No debes marcar esta zona sombreada — CÓDIGO: 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

(2) No debes marcar esta zona sombreada — 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

(3) No debes marcar esta zona sombreada — 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

Habitualmente, ¿qué haces con la grasa de la carne? 1 La como ☐ 2 Se la quito ☐

	SÍ	NO		SÍ	NO
¿Procuras tomar mucha fibra?	☐	☐	¿Evitas el consumo de mantequilla?	☐	☐
¿Procuras tomar mucha fruta?	☐	☐	¿Procuras reducir el consumo de grasa?	☐	☐
¿Procuras tomar mucha verdura?	☐	☐	¿Procuras reducir el consumo de carne?	☐	☐
¿Procuras tomar mucho pescado?	☐	☐	¿Limitas la sal en las comidas?	☐	☐
¿Sueles comer entre comidas (picotear)?	☐	☐	¿Le añades azúcar a algunas bebidas?	☐	☐
¿Sigues una dieta especial?	☐	☐	¿Procuras reducir el consumo de dulces?	☐	☐

Si has contestado SÍ, señala el tipo de dieta: ____________________

(3) No debes marcar esta zona sombreada — 0 1 2 3 4 5 6 7 8 9 / 0 1 2 3 4 5 6 7 8 9

SUMCO 9416-9-K (Ref. 4)

¿Dispones de correo electrónico (e-mail)? ☐ No ☐ Sí,

¿Dispones de acceso a Internet? ☐ No ☐ Sí

1.D. Table of modified criteria for 1 point in the SUN cohort from the original MEDLIFE index designed and validated by Sotos-Prieto et. al.

Original MEDLIFE items	Original components for Aragon Worker's Health Study cohort	SUN cohort MEDLIFE items	Components derived from baseline FFQ in the SUN cohort (serving size)	Original criteria for 1 point	SUN criteria for 1 point
Block 1: Mediterranean food consumption					
1. Sweets	Candy (1 serving=1 unit or 50g), chocolates (1 serving=30g), biscuits (1 serving=4–6 units), turron (1 serving=40g)	1. Sweets	Cookies, chocolate cookies, pastries, donuts, homemade baked goods, store-bought baked goods (50g), muffins (25-50g), tea biscuits (90g), chocolates (30g), churros (100g), turrón (35g)	≤ 2 servings/wk	≤ 2 servings/wk
2. Red meat	Beef, pork, lamb (1serving=100–150g)	2. Red meat	Beef, pork, lamb (100-150g)	< 2 servings/wk	< 2 servings/wk
3. Processed meat	Ham (1serving=1 slice or 30g), sausage, soft spicy sausage, bacon (1 serving=50g), hamburger (1 serving=1 unit), liver (1 serving=100–150g), pâté (1serving=25g)	3. Processed meat	Sausage, soft spicy sausage, bacon (50g), cured ham (60g), cooked ham (30g), hamburger (150g), liver, organ meats (100-150g), pâté (25g)	≤ 1 serving/wk	≤ 1 serving/wk
4. Eggs	Eggs (1egg)	4. Eggs	Eggs (1 unit)	2-4 units/wk	2-4 servings/wk
5. Legumes	Lentils, beans, peas, chickpeas (1 serving=1 plate or 150g)	5. Legumes	Lentils, beans, chickpeas, peas (60g uncooked)	≥ 2 servings/wk	≥ 2 servings/wk
6. White meat	Poultry and rabbit (1serving=100–150g)	6. White meat	Chicken/turkey with skin, chicken/turkey without skin, rabbit (100-150g)	2 servings/wk	2 servings/wk
7. Fish/seafood	White/oily fish (1 serving=100–150g), canned fish (1 serving=1can or 50g), seafood (1 serving=200g)	7. Fish/seafood	White fish, fatty fish, codfish, salted or smoked fish, shrimp, octopus, calamari (100-150g), oysters and shellfish (6 units)	≥ 2 servings/wk	≥ 2 servings/wk
8. Potatoes	Roast/boiled potatoes, French fries (1 serving=150–200g)	8. Potatoes	Baked or boiled potatoes (150g)	≤ 3 servings/wk	≤ 3 servings/wk
9. Low-fat dairy products	Skimmed dairy milk (1 serving=1 cup or 200ml), soft cheese	9. Low-fat dairy products	Skim milk, low-fat milk (200cc), low fat yogurt (125g), fresh soft cheese (50g)	2 servings/d	2 servings/d
10. Nuts and olives	Walnuts, almonds, hazelnuts (1 serving=1 handful or 30g), olives (1 serving=10 units)	10. Nuts and olives	Almonds, peanuts, hazelnuts, walnuts (50g), olives (10 units)	1-2 servings/d	1-2 servings/d
11. Herbs, spices, and garnish	Onion, garlic, herbs (parsley, oregano)	11. Sofrito	Olive oil, pepper, other vegetables (250g), tomato (150g)	≥ 1 serving/d	>2/4 ingredients above the median
12. Fruit	All fruit and fresh fruit-based juices (1 serving=150–200g)	12. Fruit	Orange, banana, apple, pear, kiwi, mango, avocado, peach, apricot, nectarine (1 unit), clementine (2 units), strawberry (6 units), cherries, plums, figs, grapes (1 dessert plate), watermelon, melon (200-250g), dates and dried fruits (150g)	3-6 servings/d	3-6 servings/d
13. Vegetables	All vegetables except potatoes (1 serving=150–200g)	13. Vegetables	Spinach, cauliflower, broccoli, lettuce, carrot, squash, green beans, eggplant, zucchini, cucumber, pepper, asparagus, gazpacho, garden salad, other vegetables (250g), tomato (150g) *(excludes potatoes)*	≥ 2 servings/d	≥ 2 servings/d
14. Olive oil	Olive oil, virgin olive oil (1 serving=1tbsp)	14. Olive oil	Olive oil (1Tbsp)	≥ 3 servings/d	≥ 3 servings

15. Cereals	White and whole grain bread (1 serving=40g), cereals (1 serving= 1 plate) and derivatives	15. Cereals	White bread, whole-grain bread (3 slices), white rice, pasta (60g uncooked), pizza (200g), breakfast cereal (30g)	3-6 servings/d	3-6 servings/d
Block 2: Mediterranean dietary habits					
16. Water or infusions	Water or infusions (1 serving=1 glass)	16. Water and coffee	Tap water, bottled water (200 cc), coffee, decaffeinated coffee (50cc)	6-8 glasses of water/d or ≥ 3 servings of infusions/wk	≥ 6 servings/d
17. Wine (red and white)	White/red wine(1 serving=1 cup)	17. Wine (red and white)	Red/white wine (1 glass 100 cc)	1-2 servings/d	women: >0≤0.5 serving/d men: >0≤1 serving/d
18. Limit salt at meals		18. Limit salt at meals	Do you limit salt at meals?	Yes	Yes
19. Preference for whole grain products		19. Preference for whole grain products	Do you try to consume a lot of fiber? + fiber from grains	Yes, preference for whole grain products/ fiber >25g/d	Yes + >6g/d fiber from grains
20. Snacks	Potato chips, popcorn (1 serving=1 bag or 50g)	20. Snacks	Potato chips (150g)	≤ 2 servings/wk of potato chips, popcorn (50g)	< 1 serving/wk
21. Limit nibbling between meals	Nibbling outside five main meals	21. Limit snacking between meals	Do you tend to snack in between meals?	Yes, limit nibbling between meals	No
22. Limit sugar in beverages (including sugar-sweetened beverages)		22. Limit sugar in beverages (including sugar-sweetened beverages)	Do you add sugar to some beverages? + sugar-sweetened beverages + bottled juice (200cc)	Yes, limit sugar in beverages	No + ≤ 1/wk + ≤ 1/wk
Block 3: Physical activity, rest, social habits, and conviviality					
23. Physical activity (>150 min/wk or 30 min/d)	Jogging, walking quickly, dance, aerobics, gardening	23. Physical activity	Brisk walking, jogging, running, climbing stairs, bicycling, stationary cycling, swimming, dance, aerobic exercise, martial arts, gymnastics, gardening, tennis, soccer, skiing, ice skating, team sports, and other physical activities or sports	Yes >150 min/wk, 500-1000 MET-min/wk, or 30 min brisk walking	> 300 min/wk
24. Siesta/nap	During weekends	24. Siesta/nap	Napping throughout the week	Yes, during weekends	≤ 30 min/d
25. Hours of sleep	During weekdays	25. Hours of sleep	Sleeping at night throughout the week	6-8 h/d during weekdays	6-8 h/d
26. Watching television	During weekdays	26. Watching television	Watching TV/videos throughout the week	< 1 h/d during weekdays	≤ 2 h/d
27. Socializing with friends	During week	27. Socializing with friends	Socializing throughout the week	≥ 2h during weekend	> 1 h/d
28. Collective sports	During week	28. Collective sports	Playing soccer, tennis, squash or other racket sports, basketball, or other team sports	≥ 2 h/wk	≥ 1 h/wk

0 points are assigned if any of these criteria are not met; 1 point is assigned for each criterion that is met. Highlighted items identify changes made to score.

(11) *Sofrito:* herbs, spices, and garnish were replaced by sofrito, defined as consuming more than two out of four ingredients above the median (olive oil, tomato, peppers, and other vegetables), a typical base for many Mediterranean dishes made by stir-frying onion and garlic in olive oil.[258]

(16) *Water and coffee:* infusions were substituted by regular and decaffeinated coffee (≥ 6 servings/d including water and coffee) since infusions or tea are not included in the baseline FFQ and coffee was considered an appropriate beverage for assessing adequate hydration and its given health benefits.[259]

(17) *Wine:* light wine consumption was given sex-specific cut-off points, women: 0.1-5g/d and men: 0.1-10g/d, instead of 1-2 servings/d. This criteria was defined by considering the grams of alcohol specifically from wine rather than all alcoholic beverages,[260,261] Trichopoulou's low alcohol consumption range,[262] and the Mediterranean alcohol drinking pattern previously studied in this cohort of relatively young Spanish university graduates.[263]

(19) *Preference for whole grain products:* 'yes' to preference for whole grain products or fiber >25g/d was redefined as stating 'yes' to "Do you try to consume a lot of fiber?" and >6g/d of fiber from cereals. This combination of criteria was created to capture the original item's intention to collect information on the consumption of whole grain products, which is not a direct question asked in our cohort baseline questionnaire. 6 grams of fiber from cereals was derived from the recommendation to consume half of one's total cereal intake (~6 serv/d) in the form of whole grains (~3 serv/d)[132] A full serving of whole grains (16 grams) will contain from just over a half gram of fiber to around 3 grams of fiber (wheat contains 2 grams), therefore, 3 servings*2g fiber=6g/d of fiber from cereals.[264]

(20) *Snacks:* snacks were defined as potato chips since it was the only snack item collected in the SUN cohort's baseline FFQ designed in 1999 and is commonly consumed as a snack in Spain.

(21) *Limit snacking between meals:* 'yes' to limit nibbling between meals was replaced with stating 'no' to "Do you tend to eat in between meals (snacking)?" to best match the original question collected in the SUN cohort's baseline questionnaire.[265]

(22) *Limit sugar in beverages:* 'yes' to limiting sugar in beverages (including sugar-sweetened beverages) requires stating 'no' to "Do you add sugar to some beverages?" and consuming sugar-sweetened beverages <1 serving/wk and bottled juices <1 serving/wk. The first criteria was modified to best fit the SUN cohort's baseline data. In addition, criteria were assigned to evaluate the intake of sugar-sweetened beverages and bottled juices as sugary drinks for their high content of added sugars.

(23) *Physical activity:* >150 min/wk or 30 min/d of physical activity was replaced with >300 min/wk across all reported activities and sports in the baseline physical activity questionnaire. All items were accounted for, with the exception of sailing, which is not moderate intensity (3-6 METs). Walking was only considered if participants reported their habitual walking pace to be fast or very fast.[74,266]

(24) *Siesta/nap:* yes, to napping on the weekend was replaced with napping ≤ 30 min/d throughout the week to define healthy habitual napping. Evidence indicates the effect is different depending on the duration of the nap and its context within daily hours of sleep. A J-curve association exists between sleep and mortality and a linear dose-response relationship between nap time and the risk of all-cause mortality, indicating a need to define this lifestyle habit.[209–211]

(25) *Hours of sleep:* 6-8 h/d of sleep on weekdays was replaced with 6-8 hours/d throughout the week to not discriminate weekdays from weekends.[209,267]

(26) *Watching TV:* watching TV <1 h/d on weekdays was replaced with ≤2 h/d throughout the week.[268]

(27) *Socializing with friends:* socializing with friends ≥2 h/d on the weekend was modified to >1 h/d throughout the week to account for the high prevalence of socializing within our study population.[220,269,270]

(28) *Collective sports:* collective sports ≥2 h/wk was modified to ≥1 h/wk which included tennis, squash or other racket sports, soccer, basketball, and other sports not mentioned on the baseline questionnaire.

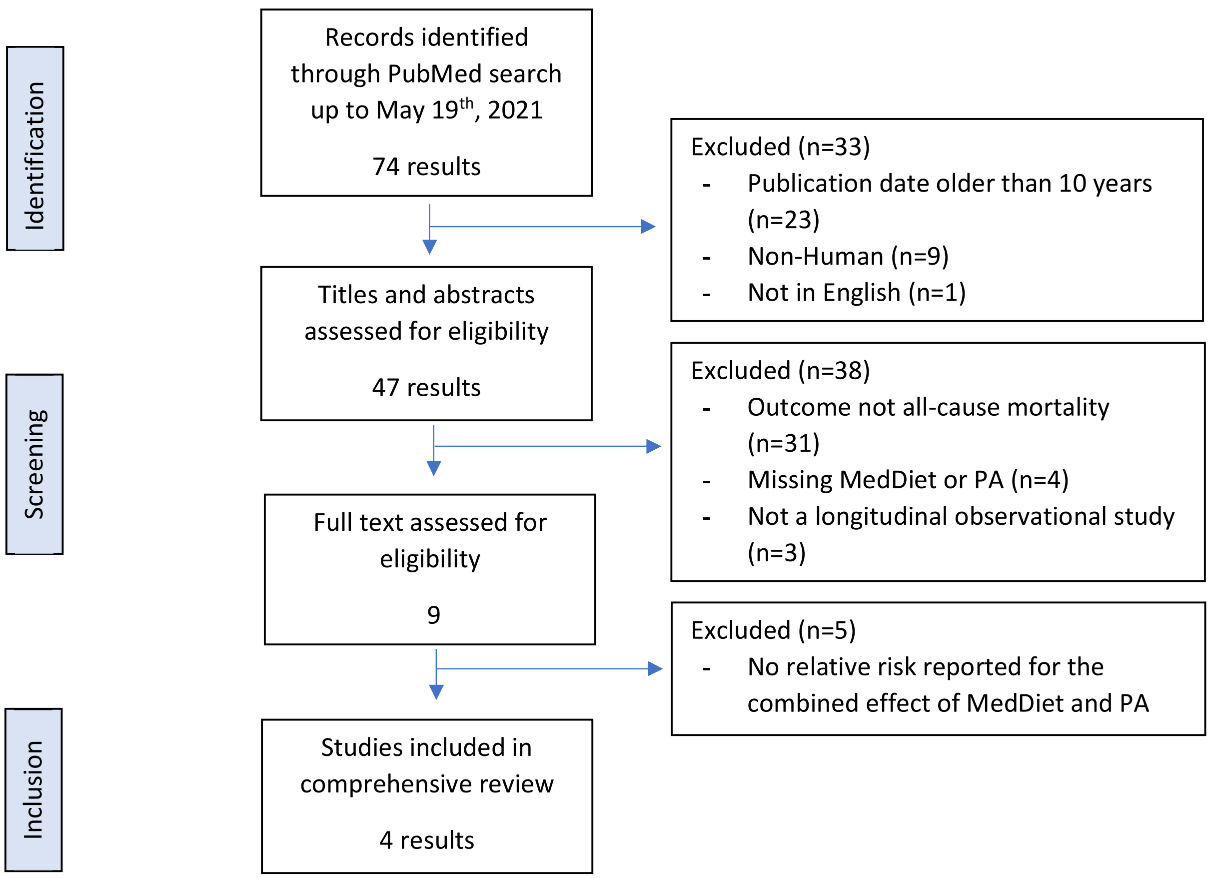
Supplementary Material 1.1

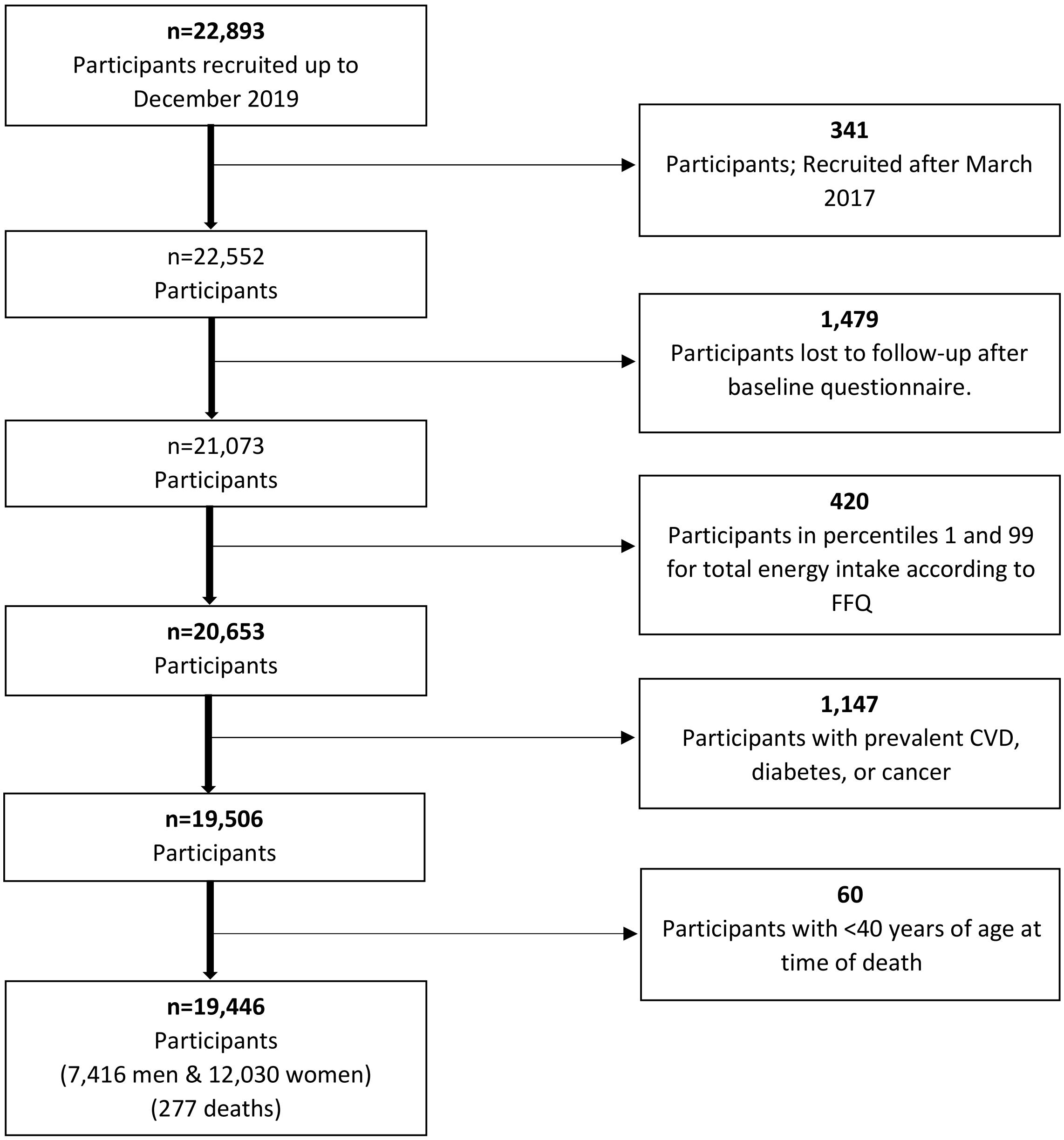
Hershey MS, Martínez-González MA, Álvarez-Álvarez I, Martínez-Hernández JA, & Ruiz-Canela M. The Mediterranean diet and physical activity: Better together than apart for the prevention of premature mortality. *British Journal of Nutrition*. 2021;1-12. doi:10.1017/S0007114521002877

Supplementary Table 1. Search strategy, database searched until May 19th, 2021.

Database	Search strategy
PubMed	TOPIC: ((“Mediterranean diet” OR “Mediterranean dietary pattern” OR “Mediterranean diets” OR “Diets, Mediterranean”)) AND ((“Physical Activity” OR “Physical Activities” OR “Activity, Physical”)) AND ((“survival" OR “survive" OR “mortality" OR “fatal" OR “death")) AND (("follow up" OR "longitudinal studies" OR "cohort studies" OR "prospective studies"))

Supplementary Figure 1. Flow chart of eligible studies for comprehensive review.





Supplementary Figure 2. Flowchart for selection of participants in the SUN cohort

Supplementary Table 2: Baseline characteristics according to adherence to Trichopoulou's Mediterranean diet score and an 8-item physical activity score in the SUN cohort.

	Mediterranean Diet				Physical Activity	
	Q1 MDS (0-3 pts.)	**Q2 MDS (4 pts.)**	**Q3 MDS (5-6 pts.)**	**Q4 MDS (7-9 pts.)**	**Low PA (0-3 pts.)**	**High PA (4-8 pts.)**
N (%)	6,772 (34.8)	3,968 (20.4)	6,527 (33.6)	2,179 (11.2)	6,840 (35.2)	12,606 (64.8)
PA score (points)	4.0 (1.8)	4.2 (1.8)	4.4 (1.8)	4.7 (1.8)	2.2 (0.9)	5.3 (1.1)
MDS (points)	2.3 (0.8)	4.0 (0.0)	5.4 (0.5)	7.3 (0.5)	4.0 (1.8)	4.4 (1.8)
Women (%)	62.6	62.6	61.5	59.4	67.0	59.1
Age (years)	34.6 (10.6)	37.0 (11.6)	39 (12.1)	42.6 (12.7)	37.7 (11.7)	37.3 (11.9)
BMI (kg/m^2)	23.1 (3.5)	23.3 (3.4)	23.6 (3.5)	23.8 (3.5)	23.8 (3.9)	23.2 (3.2)
Masters or doctorate (%)	17.7	17.4	17.8	18.7	16.6	18.5
Smoking status (%)						
Never	65.5	67.3	70.4	41.4	45.0	50.9
Current	23.0	24.0	21.1	20.7	26.9	19.8
Former	23.2	26.7	31.0	37.2	27.4	28.5
Family history of CVD (%)	11.6	12.9	14.3	17.3	14.1	13.0
Hypertension at baseline (%)	4.6	5.1	7.7	9.9	7.1	5.9
Hypercholesterolemia (%)	11.9	15.0	18.0	24.9	16.5	15.8
Depression (%)	10.5	11.6	11.3	12.7	12.6	10.5
Energy intake (kcal/d)	2,357 (741)	2,499 (783)	2,614 (802)	2,704 (724)	2,473 (784)	2,531 (775)
Carbohydrate intake (%E)	41.3 (7.1)	43.2 (7.3)	44.6 (7.2)	46.8 (7.0)	42.7 (7.5)	43.8 (7.3)
Protein intake (%E)	18.1 (3.3)	18.0 (3.3)	18.0 (3.2)	17.7 (3)	18.0 (3.3)	18.0 (3.2)
Fat intake (%E)	39.0 (6.0)	36.8 (6.3)	35.3 (6.3)	32.9 (6.4)	37.4 (6.6)	36.2 (6.5)
Saturated fat (g/d)	38.0 (15.7)	35.7 (15.4)	33.4 (14.4)	29.1 (11.5)	35.3 (14.9)	34.9 (15.1)
Polyunsaturated fat (g/d)	14.2 (6.9)	14.6 (7.3)	15.1 (7.2)	15.4 (6.9)	14.9 (7.3)	14.6 (6.9)
Monounsaturated fat (g/d)	42.3 (16.3)	43.9 (17.5)	45.0 (18.0)	45.1 (17.2)	44.1 (17.6)	43.7 (17.0)
Monounsaturated:saturated fat (%E)	1.14 (0.2)	1.3 (0.3)	1.4 (0.3)	1.6 (0.4)	1.3 (0.3)	1.31 (0.4)
Fiber intake (g/d)	21.5 (9.0)	28.0 (11.3)	34.6 (14.2)	42.2 (15.8)	27.6 (13.3)	30.6 (14.4)
Vegetables (g/d)	377 (240)	520 (318)	659 (398)	800 (431)	513 (351)	567 (377)
Fruits (g/d)	230 (218)	342 (326)	453 (359)	575 (408)	327 (319)	387 (346)
Legumes (g/d)	19.3 (18.0)	23.3 (19.4)	26.1 (21.1)	30.4 (18.9)	23.1 (20.4)	23.9 (19.5)
Cereals (g/d)	87 (69)	108 (82)	122 (84)	144 (81)	106 (80.0)	111 (81.0)
Meat (g/d)	197 (90)	188 (89)	178 (89)	152 (75)	188 (93.0)	181 (87.0)
Fish (g/d)	74 (59)	98 (71)	118 (68)	142 (76)	98 (66.0)	103 (73.0)
Dairy products (g/d)	284 (236)	220 (212)	168 (192)	92 (107)	203 (204)	214 (221)
Nuts (g/d)	5.3 (8.3)	6.9 (12.5)	9.6 (15.6)	14.7 (22.8)	7.0 (13.8)	8.7 (14.6)
Olive oil (g/d)	14.7 (13.1)	19.0 (15.4)	22.4 (16.5)	25.9 (16.6)	19.7 (16.3)	19.3 (15.4)
Alcohol consumption (g/d)	5.0 (9.8)	6.4 (10.4)	7.5 (10.3)	9.9 (10.3)	6.3 (11.0)	6.8 (9.8)
Do exercise (%)	65.5	67.3	70.4	74.1	36.2	86.0
Intensity (METs/h)	4.9 (1.1)	4.9 (1.1)	4.9 (1.1)	4.9 (1.1)	4.3 (1.0)	5.2 (1.1)
Energy expenditure (METs-h/wk)	19.0 (20.9)	21.0 (21.6)	23.5 (24.7)	27.7 (26.8)	7.3 (9.3)	29.8 (24.6)
Walking pace: brisk or very fast (%)	55.7	55.4	56.6	59.3	34.6	68.1
Walking (min/d)	35.7 (31.0)	37.3 (31.2)	39.4 (32.0)	42.2 (32.3)	25.1 (22.5)	44.9 (33.6)
Climbing stairs (floors/d)	3.6 (3.9)	3.6 (3.8)	3.6 (3.8)	3.8 (3.9)	2.3 (2.8)	4.3 (4.1)
Watching television (h/d)	1.6 (1.2)	1.6 (1.2)	1.6 (1.1)	1.6 (1.1)	1.9 (1.3)	1.5 (1.1)
Sitting down (h/d)	5.4 (2.1)	5.3 (2.1)	5.2 (2.1)	5.0 (2.0)	5.8 (2.0)	5.0 (2.0)

MDS: Trichopoulou's Mediterranean diet score, PA: physical activity, N: population size, BMI: body mass index, CVD: cardiovascular disease, kcal: kilocalories, d:day, E: total energy intake, g: grams, METs: metabolic equivalent of task, h: hours, wk: week, min: minutes

Values for continuous variables are presented as means ± (SD) and categorical variables are specified as a percentage (%).

Supplementary Table 3. Frequency of each item and comparison across Mediterranean diet and physical activity scores in the SUN cohort.

MDS items	n (%) with point			
1-point criteria	All	Low PA	High PA	*p-value*
Vegetables				
≥ median (473.8 g/d)	9,694 (49.9)	3,019 (44.1)	6,675 (53.0)	*<0.001*
Legumes				
≥ median (20.6 g/d)	9,201 (47.3)	3,113 (45.5)	6,088 (48.3)	*<0.001*
Fruits				
≥ median (286.9 g/d)	9,722 (50.0)	2,953 (43.2)	6,769 (53.7)	*<0.001*
Dairy products				
< median (139.8 g/d)	9,725 (50.0)	3,464 (50.6)	6,261 (49.7)	*0.193*
Cereals				
≥ median (85.7g/d)	9,709 (49.9)	3,264 (47.7)	6,445 (51.1)	*<0.001*
Meat				
< median (174.3 g/d)	9,738 (59.5)	3,312 (48.4)	6,426 (51.0)	*0.001*
Fish				
≥ median (88.3 g/d)	13,300 (68.4)	4,504 (46.8)	8,796 (51.6)	*<0.001*
Monounsaturated:saturated fat				
≥ median (1.23)	9,723 (50.0)	3,355 (49.1)	6,368 (50.5)	*0.051*
Alcohol				
10-50 g/d men & 5-25 g/d women	5,612 (28.9)	1,802 (26.4)	3,810 (30.2)	*<0.001*
Total MDS (mean ±SD)	4.26 (1.8)	4.02 (1.8)	4.39 (1.8)	*<0.001*

PA score items	n (%) with point					
1-point criteria	All	Q1 MDS	Q2 MDS	Q3 MDS	Q4 MDS	*p-value*
Do you exercise?						
yes	13,320 (68.5)	4,438 (65.5)	2,671 (67.3)	4,597 (70.4)	1,614 (74.1)	*<0.001*
Exercise intensity[a]						
Vigorous (≥ 6)	12,184 (62.7)	4,118 (60.8)	2,430 (61.2)	4,145 (63.5)	1,491 (68.4)	*<0.001*
Energy expenditure						
≥ 16.1 METs-h/wk	9,571 (49.2)	2,887 (42.6)	1,934 (48.7)	3,425 (52.5)	1,325 (60.8)	*<0.001*
Walking pace[b]						
Brisk or very fast	10,954 (56.3)	3,773 (55.7)	2,197 (55.4)	3,693 (56.6)	1,291 (59.3)	*0.017*
Walking time						
≥ 0.5 h/d	8,071 (41.5)	2,556 (37.7)	1,610 (40.6)	2,857 (43.8)	1,048 (48.1)	*<0.001*
Climbing upstairs						
≥ 3 floors/d	9,449 (48.6)	3,222 (47.6)	1,922 (48.4)	3,152 (48.3)	1,153 (52.9)	*<0.001*
Television viewing time						
< 1.5 h/d	10,266 (52.8)	3,537 (52.2)	2,031 (51.2)	3,505 (53.7)	1,193 (54.8)	*0.015*
Sitting time[c]						
< 5 h/d	8,498 (43.7)	2,709 (40.0)	1,686 (42.5)	3,007 (46.1)	1,096 (50.3)	*<0.001*
Total PA score (mean ±SD)	4.23 (1.8)	4.02 (1.8)	4.15 (1.8)	4.35 (1.8)	4.69 (1.8)	*<0.001*

MDS: Trichopoulou's Mediterranean diet score, PA: physical activity, n: number of participants, g: grams, d:day, h: hour, METs: metabolic equivalent of task, wk: week

[a]Exercise intensity was measured on a scale from 0 (minimum intensity) to 10 (maximum intensity)

[b]Responses included slow, normal/average, brisk, or very fast

[c]Includes working and leisure time

Supplementary Table 4. Methods for interaction analyses on multiplicative and additive scales.

Multiplicative Interaction:

Likelihood ratio test – Comparison of cox regression models using the likelihood ratio test.

STATA code:

```
generate I_A=g*e
stcox e g   c1 c2 c3
est store A
stcox I_A e g   c1 c2 c3
lrtest A .
```

g = 1; low Mediterranean diet score (Q1: 0-3 points)
g = 0; reference (Q4: 7-9 points)
c = covariables

e = 1; low physical activity (0-3 points)
e = 0; reference (4-8 points)

Additive Interaction:

Relative excess risk due to interaction (RERI) – Proportion of the *effect* of both exposures on the additive scale that is due to their interaction.

$$RERI_{HR} = HR_{11} - HR_{10} - HR_{01} + 1$$

HR_{11} is the adjusted hazard rate ratio comparing the doubly exposed higher risk combination to the reference combination with the lowest risk HR_{00}.[1] RERI = 0 means no interaction or exactly additivity; RERI > 0 means positive interaction or more than additivity; RERI < 0 means negative interaction or less than additivity; RERI can go from – infinity to + infinity. [2]

STATA code:

```
stcox I_A e g   c1 c2 c3
nlcom (exp(_b[I_A] + _b[g] + _b[e]) - exp(_b[g]) - exp(_b[e]) + 1)
```

Attributable proportions due to the joint effect – proportion of the joint effect (total hazard) that is due to each component among those who present both exposures.

Portion of the effect attributable to the MedDiet alone: $(HR_{10} - 1) / (HR_{11} - 1)$
Portion of the effect attributable to PA alone: $(HR_{01} - 1) / (HR_{11} - 1)$
Portion of the effect attributable to their interaction: $(RERI_{HR} - 1) / (HR_{11} - 1)$

STATA CODE:

```
nlcom (exp(_b[g])-1)/(exp(_b[I_A]+_b[g]+_b[e])-1)
nlcom (exp(_b[e])-1)/(exp(_b[I_A]+_b[g]+_b[e])-1)
nlcom (exp(_b[I_A]+_b[g]+_b[e])- exp(_b[g]) - exp(_b[e]) + 1)/(exp(_b[I_A]+_b[g]+_b[e])-1)
```

References:

1. Li R, Chambless L. Test for Additive Interaction in Proportional Hazards Models. *Ann Epidemiol*. 2007;17(3):227-236. doi:10.1016/j.annepidem.2006.10.009
2. Rothman KJ. *Modern Epidemiology*. 1st ed. Boston: Little, Brown; 1986.
3. VanderWeele TJ. Causal interactions in the proportional hazards model. *Epidemiology*. 2011;22(5):713-717. doi:10.1097/EDE.0b013e31821db503

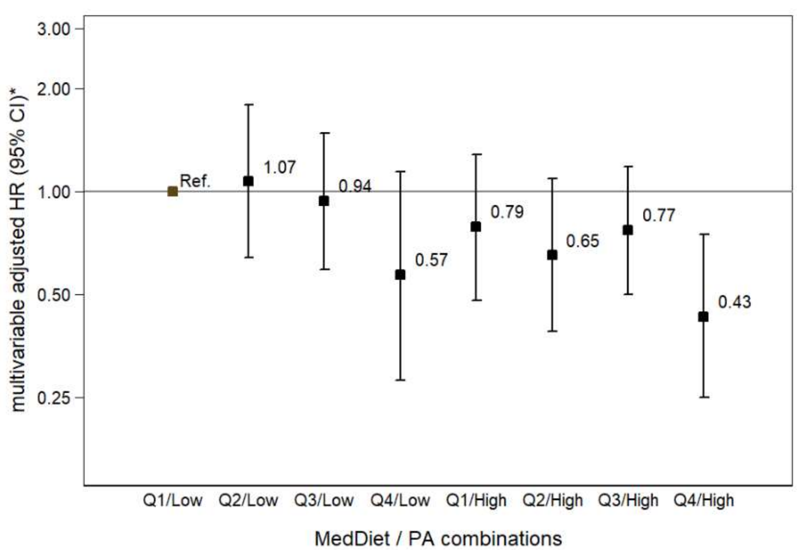
Supplementary Table 5. Individual and joint effects (HR) between protective factors for adherence to the Mediterranean diet and physical activity on all-cause mortality.

	N	Deaths *(%)*	Time at risk (person-years)	Multivariable adjusted HR*	95% CI	
					Lower limit	Upper limit
Continuous exposures						
9 item MDS	19,446	277 *(1.42)*	225,057	0.90	0.84	0.97
8 item PA score	19,446	277 *(1.42)*	225,057	0.88	0.82	0.94
Individual effects						
Q1 MDS *(0-3 pts.)*	6,772	71 *(1.05)*	80,629	1 Ref.		
Q2 MDS *(4 pts.)*	3,968	58 *(1.46)*	46,437	0.94	0.65	1.35
Q3 MDS *(5-6 pts.)*	6,527	110 *(1.69)*	74,063	0.98	0.71	1.35
Q4 MDS *(7-9 pts.)*	2,179	38 *(1.74)*	23,929	0.59	0.38	0.91
Low PA *(0-3 pts.)*	6,840	121 *(1.77)*	79,189	1 Ref.		
High PA *(4-8 pts.)*	12,606	156 *(1.24)*	14,5869	0.76	0.59	0.98
4x2 Joint effects						
Q1 MDS-low PA	2,705	35 (1.29)	32,007	1 Ref.		
Q2 MDS-low PA	1,442	29 (2.01)	16,929	1.07	0.64	1.79
Q3 MDS-low PA	2,121	45 (2.12)	23,941	0.94	0.59	1.48
Q4 MDS-low PA	572	12 (2.10)	6,312	0.57	0.28	1.14
Q1 MDS-high PA	4,067	36 (0.89)	48,622	0.79	0.48	1.28
Q2 MDS-high PA	2,526	29 (1.15)	29,508	0.65	0.39	1.09
Q3 MDS-high PA	4,406	65 (1.48)	50,122	0.77	0.50	1.18
Q4 MDS-high PA	1,607	26 (1.62)	17,617	0.43	0.25	0.75

HR: hazard ratio: CI: confidence intervals, MDS: Mediterranean diet score, N: sample population size, PA: physical activity, pts: points

*Adjusted for sex, BMI, education level, smoking status, cigarettes smoked, alcohol, total energy intake, family history of CVD, prevalent hypertension, hypercholesterolemia, depression, and stratified by year entering the cohort and age in decades. Individual exposures were additionally adjusted for the remaining lifestyle factor.

Supplementary Figure 3. Hazard ratios (95%CI) for the combinations of adherence to the MedDiet and PA levels on all-cause mortality.

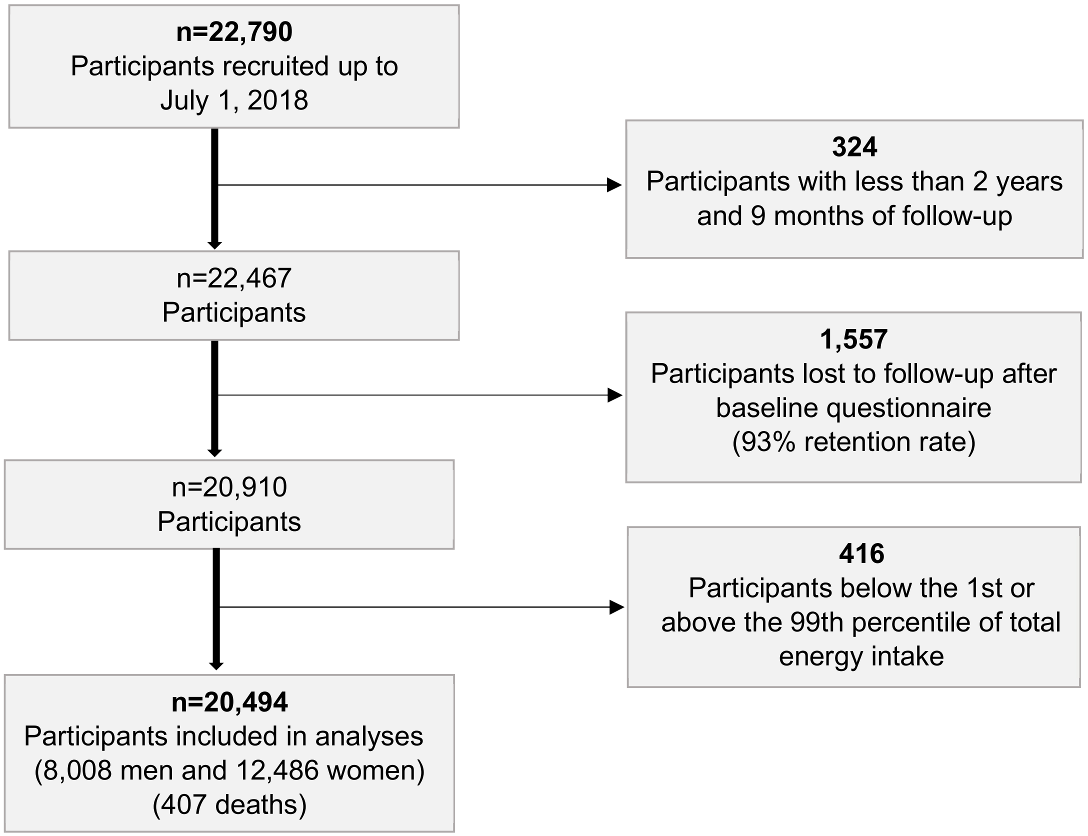


HR: hazard ratio: CI: confidence intervals, MedDiet: Mediterranean dietary pattern, PA: physical activity
*Multivariable Cox proportional hazard models adjusted for sex, BMI, education level, smoking status, cigarettes smoked, alcohol, total energy intake, family history of CVD, prevalent hypertension, hypercholesterolemia, depression, and stratified by year entering the cohort and age in decades.

Supplementary Material 1.2

Hershey MS, Fernández-Montero A, Sotos-Prieto M, Kales S, Gea A, Ruiz-Estigarribia L, Sánchez-Villegas A, Díaz-Gutiérrez J, Martínez-González MA, & Ruiz-Canela M. The Association Between the Mediterranean Lifestyle Index and All-Cause Mortality in the Seguimiento Universidad de Navarra Cohort. *American Journal of Preventive Medicine.* 2020;59(6), e239-e248. doi:10.1016/j.amepre.2020.06.014

Appendix Figure 1. Flowchart of eligible participants in the SUN cohort 1999–2018.



Appendix Table 1. Description of the Mediterranean Lifestyle (MEDLIFE) Index Modified for the SUN Cohort

Score items	**Components (serving size)**	**Criteria for 1 point**
Block 1: Mediterranean food consumption		
1. Sweets	Cookies, chocolate cookies, pastries, donuts, homemade baked goods, store-bought baked goods (50 g), muffins (25–50 g), tea biscuits (90 g), chocolates (30 g), churros (100 g), turrón (35 g)	≤2 servings/wk
2. Red meat	Beef, pork, lamb (100–150 g)	<2 servings/wk
3. Processed meat	Sausage, soft spicy sausage, bacon (50 g), cured ham (60 g), cooked ham (30 g), hamburger (150 g), liver (100–150 g), organ meats (100–150 g), pâté (25 g)	≤1 serving/wk
4. Eggs	Eggs (1 unit)	2–4 servings/wk
5. Legumes	Lentils, beans, chickpeas, peas (60 g uncooked)	≥2 servings/wk
6. White meat	Chicken/turkey with skin, chicken/turkey without skin, rabbit (100–150 g)	2 servings/wk
7. Fish/seafood	White fish, fatty fish, codfish, salted or smoked fish, shrimp, octopus, calamari (100–150 g), oysters and shellfish (6 units)	≥2 servings/wk
8. Potatoes	Baked or boiled potatoes (150 g)	≤3 servings/wk
9. Low-fat dairy products	Skim milk (200 cc), low-fat milk (200 cc), low fat yogurt (125 g), fresh soft cheese (50 g)	2 servings/d
10. Nuts and olives	Almonds, peanuts, hazelnuts, walnuts (50 g), olives (10 units)	1–2 servings/d
11. Sofrito	Olive oil, pepper, other vegetables (250 g), tomato (150 g)	>2/4 ingredients above the median
12. Fruit	Orange, banana, apple, pear, kiwi, mango, avocado, peach, apricot, nectarine (1 unit), clementine (2 units), strawberry (6 units), cherries, plums, figs, grapes (1 dessert plate), watermelon, melon (200–250 g), dates and dried fruits (150 g)	3–6 servings/d
13. Vegetables	Spinach, cauliflower, broccoli, lettuce, carrot, squash, green beans, eggplant, zucchini, cucumber, pepper, asparagus, gazpacho, garden salad, other vegetables (250 g), tomato (150 g) *(excludes potatoes)*	≥2 servings/d
14. Olive oil	Olive oil (1 tbsp)	≥3 servings/d
15. Cereals	White bread, whole-grain bread (3 slices), white rice, pasta (60 g uncooked), pizza (200 g), breakfast cereal (30 g)	3–6 servings/d
Block 2: Mediterranean dietary habits		
16. Water and coffee	Tap water, bottled water (200 cc), coffee, decaffeinated coffee (50 cc)	≥6 servings/d

17. Wine	Red/white wine (1 glass)	women: 0.1–5g/d men: 0.1–10g/d
18. Limit salt at meals	Do you limit salt at meals?	Yes
19. Preference for whole grains	Do you try to consume a lot of fiber? + fiber from grains	Yes + >6g/d fiber from grains
20. Snacks	Potato chips (150 g)	<1 serving/wk
21. Limit snacking in between meals	Do you tend to eat in between meals?	No
22. Limit sugar in beverages (including sugar-sweetened beverages)	Do you add sugar to some beverages? + soda + bottled juice (200 cc)	No + ≤1/wk + ≤1/wk
Block 3: Physical activity, rest, social habits, and conviviality		
23. Physical activity	Walking, jogging, running, climbing stairs, bicycling, stationary cycling, swimming, dance, aerobic exercise, martial arts, gymnastics, gardening, tennis, soccer, skiing, ice skating, team sports like basketball, other physical activities or sports	>300 min/wk
24. Nap	Napping throughout the week	≤30 min/d
25. Hours of sleep	Sleeping throughout the week	6–8 h/d
26. Watching television	Watching TV/videos throughout the week	≤2 h/d
27. Socializing with friends	Socializing throughout the week	>1 h/d
28. Collective sports	Playing soccer, tennis, squash, basketball and other team sports	≥1 h/wk

min, minutes; h, hours; d, day; wk, week; cc, cubic centimeter; g, grams; tbsp., tablespoon.

Appendix Table 2. Adjusted MEDLIFE Characteristics According to MEDLIFE Quartiles at Baseline in the SUN Cohort, 1999–2015

Characteristic	**MEDLIFE adherence**			
	Q1 (3–10 pts)	**Q2 (11–12 pts)**	**Q3 (13–14 pts)**	**Q4 (15–23 pts)**
N	6,390	5,783	4,820	3,501
Sweets (serv/wk)	9.19 (8.66)	8.31 (7.98)	7.50 (7.61)	6.54 (7.39)
Red meat (serv/wk)	4.07 (2.39)	3.78 (2.24)	3.45 (2.17)	2.84 (2.30)
Processed meat (serv/wk)	6.82 (5.10)	6.81 (5.18)	6.70 (5.16)	6.34 (5.33)
Eggs (serv/wk)	2.84 (2.33)	2.79 (1.97)	2.73 (1.88)	2.74 (1.57)
Legumes (serv/wk)	2.37 (2.10)	2.76 (2.41)	2.99 (2.57)	3.18 (2.07)
White meat (serv/wk)	2.10 (1.84)	2.22 (1.88)	2.31 (1.76)	2.48 (2.02)
Fish (serv/wk)	4.35 (3.01)	5.14 (4.10)	5.66 (3.40)	6.38 (3.88)
Potatoes (serv/wk)	1.37 (1.65)	1.34 (1.41)	1.36 (1.45)	1.49 (1.61)
Dairy products (serv/d)	1.13 (1.33)	1.32 (1.41)	1.59 (1.55)	1.85 (1.68)
Nuts and olives (serv/d)	0.26 (0.35)	0.30 (0.37)	0.35 (0.50)	0.45 (0.57)
Sofrito (%)	4.32	10.14	19.69	35.04
Fruit intake (serv/d)	1.90 (1.74)	2.47 (2.13)	2.96 (2.36)	3.75 (2.61)
Vegetable intake (serv/d)	1.76 (1.11)	2.31 (1.39)	2.80 (1.60)	3.56 (2.21)
Olive oil (serv/d)	1.24 (1.18)	1.46 (1.34)	1.74 (1.49)	2.15 (1.66)
Cereals (serv/d)	1.75 (1.31)	1.93 (1.41)	2.09 (1.48)	2.51 (1.61)
Water and coffee (serv/d)	5.05 (2.84)	5.92 (2.98)	6.52 (3.02)	7.14 (2.94)
Wine (g/d), median (IQR)	0 (0–1.84)	0.59 (0–3.77)	0.59 (0–3.77)	1.17 (0–3.77)
Men, median (IQR)	0.59 (0–4.34)	1.26 (0–5.03)	1.26 (0.59–6.91)	1.84 (0.59–5.03)
Women, median (IQR)	0 (0–1.17)	0 (0–1.26)	0.59 (0–1.26)	0.59 (0–1.84)
Limit salt at meals (%)	29.28	44.14	54.45	67.49
Try to consume a lot of fiber (%)	44.59	58.05	68.97	82.50
Fiber intake from cereals (g/d)	3.51 (2.64)	3.92 (3.06)	4.41 (3.41)	5.76 (4.38)
Snacks (serv/wk)	1.67 (1.78)	1.35 (1.58)	1.08 (1.35)	0.84 (1.21)
Tend to snack in between meals (%)	48.65	34.22	27.91	18.31
Add sugar to some beverages (%)	38.29	27.92	22.49	15.40
Sugar-sweetened beverages (serv/d)	0.29 (0.59)	0.20 (0.40)	0.18 (0.34)	0.13 (0.29)
Bottled juice (serv/d)	0.15 (0.40)	0.13 (0.34)	0.12 (0.36)	0.11 (0.34)

Physical activity (min/wk)	258.98 (311.41)	399.00 (385.33)	499.02 (426.28)	639.43 (479.91)
Siesta/napping ≤30 min/d (%)	38.79	52.91	62.32	71.83
Sleeping (h/d)	7.20 (0.98)	7.21 (0.83)	7.21 (0.71)	7.20 (0.68)
Watching TV (h/d)	1.83 (1.47)	1.61 (1.20)	1.51 (1.06)	1.47 (0.98)
Socializing (h/d)	1.25 (1.19)	1.40 (1.16)	1.48 (1.11)	1.60 (1.09)
Collective sports (h/wk)	0.29 (1.03)	0.53 (1.43)	0.74 (1.73)	1.07 (2.06)

Notes: Characteristics are age and sex adjusted using the inverse probability weighting method. Continuous variables are expressed as mean and SD, unless otherwise stated.

d, day; g, grams; h, hour, min, minute; pts, points; serv, serving; wk, week.

Appendix Table 3. Sources of Variability for the MEDLIFE Index in the SUN Cohort, 1999–2015

MEDLIFE item	Criteria for 1 point	Cumulative R^2	Change in R^2
Vegetables	≥2 servings/d	0.1943	–
Physical activity	>300 min/wk	0.2982	0.1039
Red meat	<2 servings/wk	0.3682	0.0700
Siesta/nap	≤30 min/d	0.4212	0.0530
Water and coffee	≥6 servings/d	0.4728	0.0516
Limit salt at meals	Yes	0.5241	0.0513
Limit snacking in between meals	Tend to snack between meals (no)	0.5662	0.0421
Legumes	≥2 servings/wk	0.6074	0.0413
Snacks	<1 serving/wk	0.6437	0.0363
Fruit	3–6 servings/d	0.6788	0.0351
Wine	women: ≤0.5 serving/d men: ≤1 serving/d	0.7125	0.0338
Preference for whole grain products	Try to consume a lot of fiber (yes) + >6g/d fiber from cereals	0.7452	0.0327
Eggs	2–4 servings/wk	0.7765	0.0313
Limit sugar in beverages	Add sugar to some beverages (no) + Sugar-sweetened beverages ≤1/wk + Bottled juice ≤1/wk	0.8057	0.0292
Socializing with friends	>1 h/d	0.8336	0.0279
Collective sports	≥1 h/wk	0.8528	0.0192
Sofrito	>2/4 ingredients above the median	0.8713	0.0185
Watching television	≤2 h/d	0.8889	0.0175
Sweets	≤2 servings/wk	0.9067	0.0178
White meat	2 servings/wk	0.9226	0.0160
Cereals	3–6 servings/d	0.9385	0.0159
Fish/seafood	≥2 servings/wk	0.9514	0.0129
Low-fat dairy products	2 servings/d	0.9635	0.0121
Olive oil	≥3 servings/d	0.9738	0.0104
Processed meat	≤1 serving/wk	0.9819	0.0080
Nuts and olives	1–2 servings/d	0.9891	0.0073

Hours of sleep	6–8 h/d	0.9955	0.0063

Notes: Cumulative R^2 indicates the percentage of variability with the addition of each item to form the total MEDLIFE score. The change in R^2 indicates the variability corresponding to each item, indicating the most to least influential items for the difference in low and high scores. Potatoes are not featured because the stepwise-selection regressions and nested least-squares linear regressions require the exclusion of the least correlated item.

d, day; g, grams; h, hour, min, minute; pts, points; wk, week.

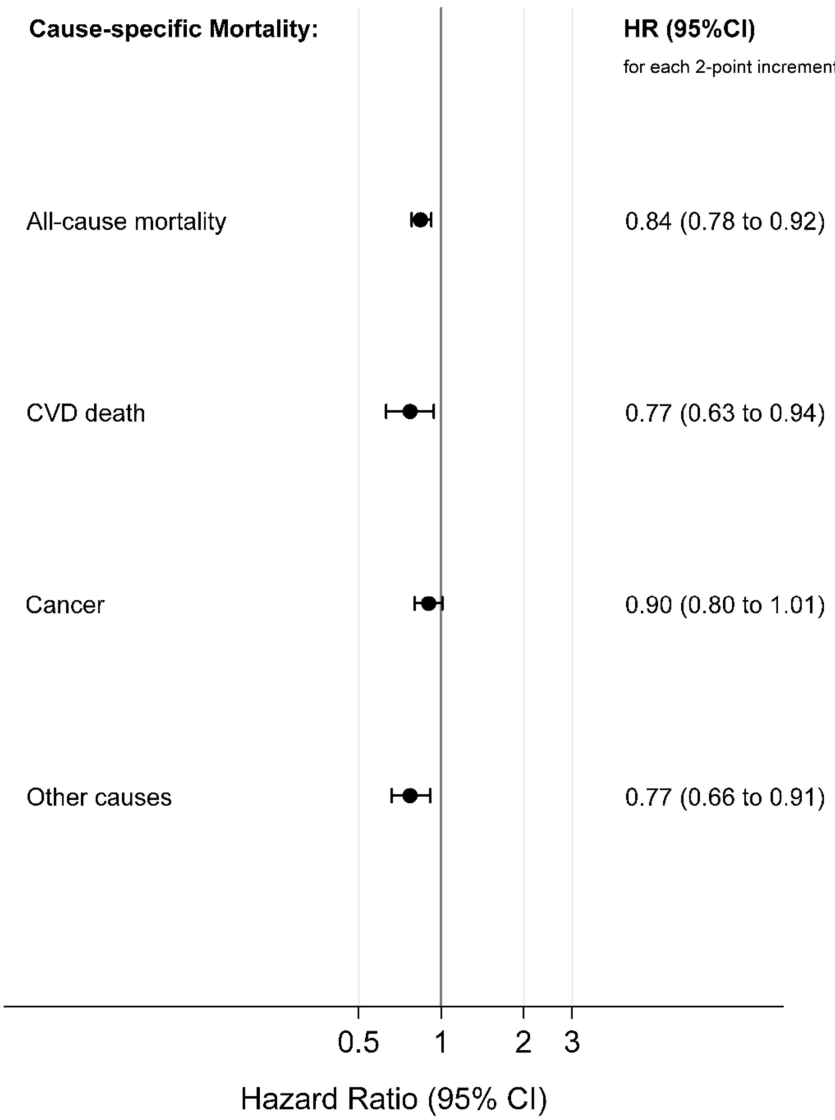
Appendix Table 4. Frequency of Adherence to Individual Items of the MEDLIFE Index in the SUN Cohort, 1999–2015

Score items	**Criteria for 1 point**	**% with 1 point**
Block 1: Mediterranean food consumption		
1. Sweets	≤2 servings/wk	18.0
2. Red meat	<2 servings/wk	24.4
3. Processed meat	≤1 serving/wk	11.6
4. Eggs	2–4 servings/wk	60.9
5. Legumes	≥2 servings/wk	57.9
6. White meat	2 servings/wk	13.8
7. Fish/seafood	≥2 servings/wk	89.3
8. Potatoes	≤3 servings/wk	96.4
9. Low-fat dairy products	2 servings/d	10.5
10. Nuts and olives	1–2 servings/d	6.0
11. Sofrito	>2/4 ingredients above the median	14.9
12. Fruit	3–6 servings/d	24.0
13. Vegetables	≥2 servings/d	54.6
14. Olive oil	≥3 servings/d	8.7
15. Cereals	3–6 servings/d	15.7
Block 2: Mediterranean dietary habits		
16. Water and coffee	≥6 servings/d	52.8
17. Wine	women: ≤0.5 serving/d (0.1–5 g/d) men: ≤1 serving/d (0.1–10 g/d)	48.5
18. Limit salt at meals	Yes	45.9
19. Preference for whole grain products	Try to consume a lot of fiber (yes) + >6g/d fiber from cereals	14.0
20. Snacks	<1 serving/wk	48.2
21. Limit snacking in between meals	Tend to snack between meals (no)	65.5
22. Limit sugar in beverages	Add sugar to some beverages (no) + Sugar-sweetened beverages ≤1/wk + Bottled juice ≤1/wk	67.4
Block 3: Physical activity, rest, social habits, and conviviality		

23. Physical activity	>300 min/wk	53.0
24. Siesta/nap	≤30 min/d	53.9
25. Hours of sleep	6–8 h/d	94.8
26. Watching television	≤2 h/d	84.3
27. Socializing with friends	>1 h/d	37.7
28. Collective sports	≥1 h/wk	20.4

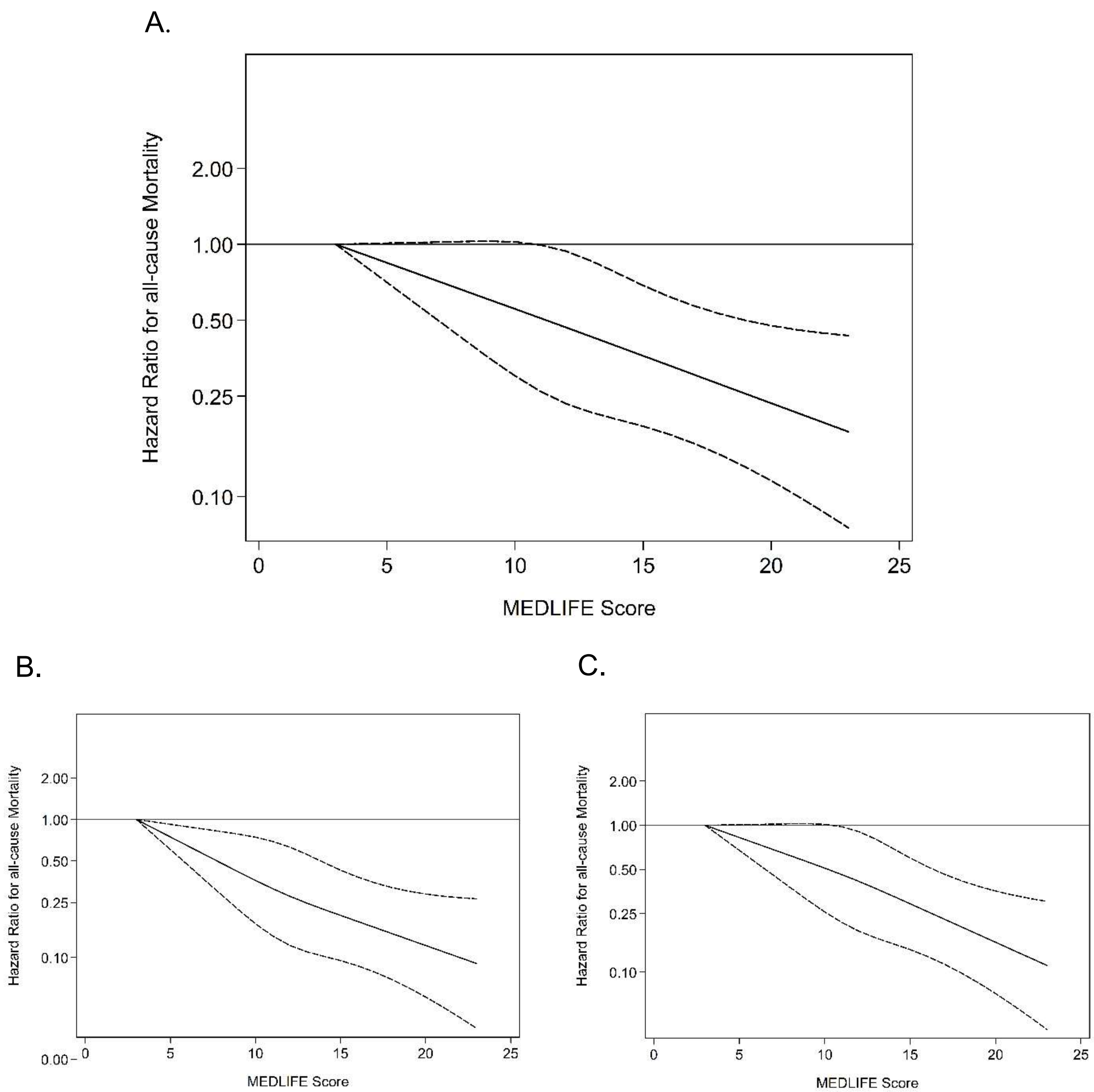
d, day; g, grams; h, hour, min, minute; wk, week.

Appendix Figure 2. HR and 95% CI for each 2-point increment in MEDLIFE on all-cause and cause-specific mortality.



Notes: Age was the underlying time variable and the model was stratified by age group and year of recruitment. Adjusted for sex, BMI, total energy intake, special diets, alcohol intake (not including wine), smoking status, cigarette pack-years, postgraduate education, family history of cardiovascular disease (CVD), prevalent hypercholesterolemia, hypertension, diabetes, cancer, and cardiovascular disease.

Appendix Figure 3. Restricted cubic spline for each 1-point increment of MEDLIFE and risk of all-cause mortality.



Notes: Age was the underlying time variable and the model was stratified by age group and year of recruitment. Adjusted for sex, BMI, total energy intake, special diets, alcohol intake (not including wine), smoking status, cigarette pack-years, postgraduate education, family history of cardiovascular disease, prevalent hypercholesterolemia, hypertension, diabetes, cancer, and cardiovascular disease. Three splines represent HR and 95% CI for (A) all participants (B) participants ≥ 50 years old at baseline (C) participants ≥ 50 years old at last contact.

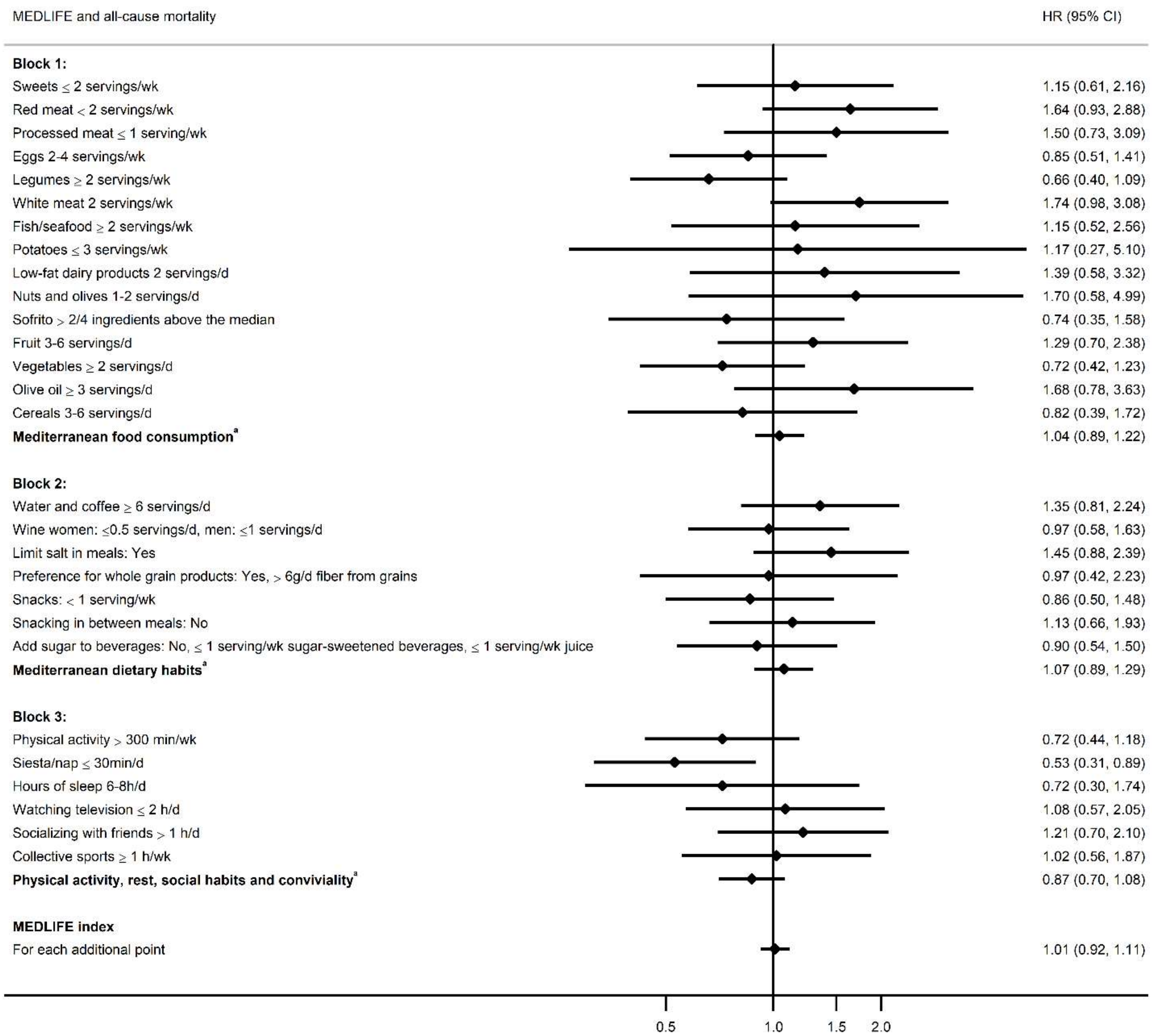
Appendix Table 5. Subgroup analyses for the association between MEDLIFE quartiles and risk of all-cause mortality.

All-cause mortality Subgroup analysis - MEDLIFE	**N**	**Cases**	**Multivariable adjusted HR (95% CI)[a]**				***p* for interaction**
			Q1 (3–10 pts)	**Q2 (11–12 pts)**	**Q3 (13–14 pts)**	**Q4 (15–23 pts)**	
Sex							0.712
Men	8,008	312	1 ref	0.77 (0.58, 1.0)	0.69 (0.51, 0.95)	**0.59 (0.36, 0.88)**	
Women	12,486	95	1 ref	0.84 (0.51, 1.38)	0.54 (0.29, 1.00)	0.66 (0.35, 1.25)	
Smoking status							0.076
Non-smokers	15,953	311	1 ref	0.85 (0.64, 1.13)	0.70 (0.51, 0.98)	**0.52 (0.34, 0.78)**	
Current smokers	4,541	96	1 ref	0.70 (0.41, 1.18)	0.38 (0.20, 0.74)	1.03 (0.53, 1.99)	
BMI							0.706
<25 kg/m^2	14,395	182	1 ref	0.76 (0.52, 1.11)	0.52 (0.33, 0.82)	**0.59 (0.36, 0.96)**	
>25 kg/m^2	6,099	225	1 ref	0.77 (0.54, 1.09)	0.69 (0.48, 1.01)	0.62 (0.37, 1.02)	
Age at baseline							**0.022**
<50 years old	16,728	116	1 ref	1.36 (0.87, 2.14)	0.93 (0.55, 1.58)	1.02 (0.56, 1.87)	
≥50 years old	3,766	291	1 ref	0.62 (0.46, 0.83)	0.53 (0.38, 0.74)	**0.44 (0.29, 0.68)**	
Age at last contact							**<0.001**
<50 years old	11,505	71	1 ref	0.96 (0.52, 1.77)	1.02 (0.51, 2.01)	1.15 (0.54, 2.44)	
≥50 years old	8,989	336	1 ref	0.76 (0.58, 0.99)	0.57 (0.42, 0.79)	**0.50 (0.34, 0.74)**	

Notes: Boldface indicates statistical significance (p<0.05).

[a]Adjusted for sex, BMI, total energy intake, special diets, alcohol intake (not including wine), smoking status, cigarettes pack-years, postgraduate education, family history of cardiovascular disease, prevalent hypercholesterolemia, hypertension, diabetes, cancer, cardiovascular disease, and stratified by age group and year of recruitment. Age was used as the underlying time variable in all models.

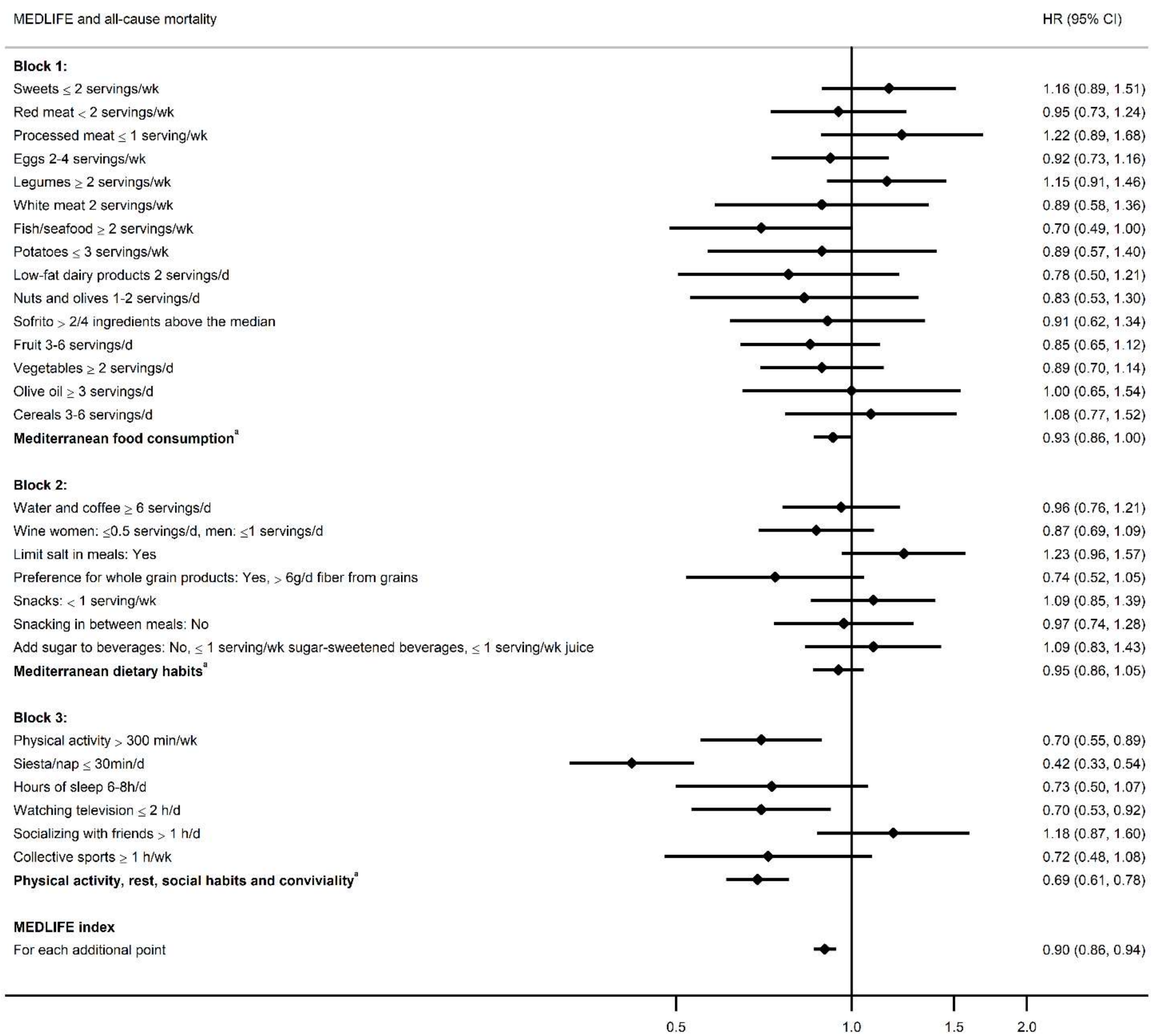
Appendix Figure 4. HR and 95% CI for each MEDLIFE item and block in participants <50 years old at last contact.



Notes: Age was the underlying time variable and the model was stratified by age group and year of recruitment. All models were adjusted for sex, BMI, total energy intake, special diets, alcohol intake (not including wine), smoking status, cigarette pack-years, postgraduate education, family history of cardiovascular disease, prevalent hypercholesterolemia, hypertension, diabetes, cancer, cardiovascular disease, and the remaining MEDLIFE items or blocks, respectively.

[a]Mediterranean food consumption is comprised of block 1 items, Mediterranean dietary habits is comprised of block 2 items, physical activity, rest, social habits, and conviviality comprises block 3 items of the MEDLIFE index.

Appendix Figure 5. HR and 95% CI for each MEDLIFE item and block in participants ≥50 years old at last contact.



Notes: Age was the underlying time variable and the model was stratified by age group and year of recruitment. All models were adjusted for sex, BMI, total energy intake, special diets, alcohol intake (not including wine), smoking status, cigarette pack-years, postgraduate education, family history of cardiovascular disease, prevalent hypercholesterolemia, hypertension, diabetes, cancer, cardiovascular disease, and the remaining MEDLIFE items or blocks, respectively.

[a]Mediterranean food consumption is comprised of block 1 items, Mediterranean dietary habits is comprised of block 2 items, physical activity, rest, social habits, and conviviality comprises block 3 items of the MEDLIFE index.

Appenidx Table 6. Sensitivity Analyses for the Association Between MEDLIFE Quartiles and the Risk of All-Cause Mortality

All-cause mortality	**N**	**Deaths**	**Q1 (3–10 pts)**	**Multivariable adjusted HR (95% CI)[a] Q2 (11–12 pts)**	**Q3 (13–14 pts)**	**Q4 (15–23 pts)**	***p* for trend**
Sensitivity analysis							
1 extra point for non-smokers	20,494	407	1 ref	0.80 (0.63, 1.02)	0.62 (0.46, 0.83)	**0.54 (0.38, 0.77)**	**<0.001**
1 extra point for nondrinkers in addition to light wine consumers	20,494	407	1 ref	0.77 (0.60, 0.98)	0.57 (0.43, 0.77)	**0.53 (0.37, 0.75)**	**<0.001**
1-point deduction if ultraprocessed foods ≥3 servings/d[1]	20,494	407	1 ref	1.12 (0.86, 1.45)	0.74 (0.55, 0.99)	**0.61 (0.44, 0.86)**	**0.002**
Exclusions							
Willet's energy intake limit[2]	18,935	385	1 ref	0.83 (0.64, 1.07)	0.66 (0.49, 0.88)	**0.60 (0.40, 0.87)**	**0.001**
Missing ≥30 FFQ items	19,897	359	1 ref	0.77 (0.59, 1.00)	0.60 (0.45, 0.81)	**0.60 (0.42, 0.86)**	**<0.001**
Chronic disease at baseline[b]							
Total deaths	15,728	145	1 ref	0.91 (0.59, 1.41)	0.87 (0.54, 1.41)	0.99 (0.58, 1.69)	**<0.001**
CVD deaths	15,728	19	1 ref	0.24 (0.04, 1.60)	0.86 (0.20, 3.73)	1.18 (0.20, 6.94)	0.2384
Cancer deaths	15,728	81	1 ref	1.19 (0.67, 2.09)	1.06 (0.57, 1.97)	0.91 (0.44, 1.91)	**<0.001**
Deaths within first 2 years of follow-up	20,445	358	1 ref	0.84 (0.65, 1.10)	0.69 (0.51, 0.93)	**0.61 (0.42, 0.88)**	**0.002**
Substitutions							
High adherence (7–10 pts) to MADP[c] instead of wine	20,494	407	1 ref	0.88 (0.69, 1.12)	0.68 (0.48, 0.96)	**0.58 (0.42, 0.78)**	**<0.001**
Abstinence from all alcohol instead of wine	20,494	407	1 ref	0.79 (0.63, 1.01)	0.55 (0.37, 0.82)	**0.59 (0.44, 0.80)**	**<0.001**
Physical activity (>150 min/d)	20,494	407	1 ref	0.77 (0.60, 0.99)	0.65 (0.49, 0.86)	**0.57 (0.40, 0.80)**	**<0.001**
Siesta/nap on weekends (yes)	20,494	407	1 ref	0.93 (0.73, 1.20)	0.81 (0.61, 1.07)	**0.67 (0.47, 0.95)**	**0.020**
Hours of sleep (6–8 h/d weekdays)	20,494	407	1 ref	0.79 (0.62, 1.01)	0.61 (0.46, 0.82)	**0.60 (0.42, 0.85)**	**<0.001**
Watching television (<1 h/d weekdays)	20,494	407	1 ref	0.85 (0.62, 1.15)	0.74 (0.57, 0.96)	**0.57 (0.41, 0.78)**	**<0.001**

Socializing with friends (≥2 h/d weekends)	20,494	407	1 ref	0.79 (0.62, 1.03)	0.72 (0.55, 0.95)	**0.53 (0.37, 0.74)**	**<0.001**
Collective sports (≥2 h/wk)	20,494	407	1 ref	0.76 (0.59, 0.98)	0.64 (0.49, 0.85)	**0.56 (0.39, 0.80)**	**<0.001**
Trichopoulou's Mediterranean diet score (0–8 pts) instead of block 1 of MEDLIFE[3]	20,494	407	1 ref	0.81 (0.63, 1.04)	0.65 (0.49, 0.86)	**0.51 (0.36, 0.71)**	**<0.001**
Original criteria used by Sotos-Prieto[4] for block 3: physical activity, rest, and social interaction	20,494	407	1 ref	0.91 (0.71, 1.17)	0.88 (0.66, 1.15)	0.71 (0.50, 1.00)	0.053

Notes: Boldface indicates statistical significance ($p<0.05$).

[a]Adjusted for sex, BMI, total energy intake, special diets, alcohol intake (not including wine), smoking status, cigarettes pack-years, postgraduate education, family history of cardiovascular disease, prevalent hypercholesterolemia, hypertension, diabetes, cancer, cardiovascular disease, and stratified by age group and year of recruitment. Age was used as the underlying time variable in all models.

[b]Chronic diseases included prevalent cases of cancer, hypertension, diabetes, and cardiovascular disease.

[c]Mediterranean alcohol drinking pattern 8-item score proposed by Gea et al.[5]

d, day; g, grams; h, hour, min, minute; wk, week.

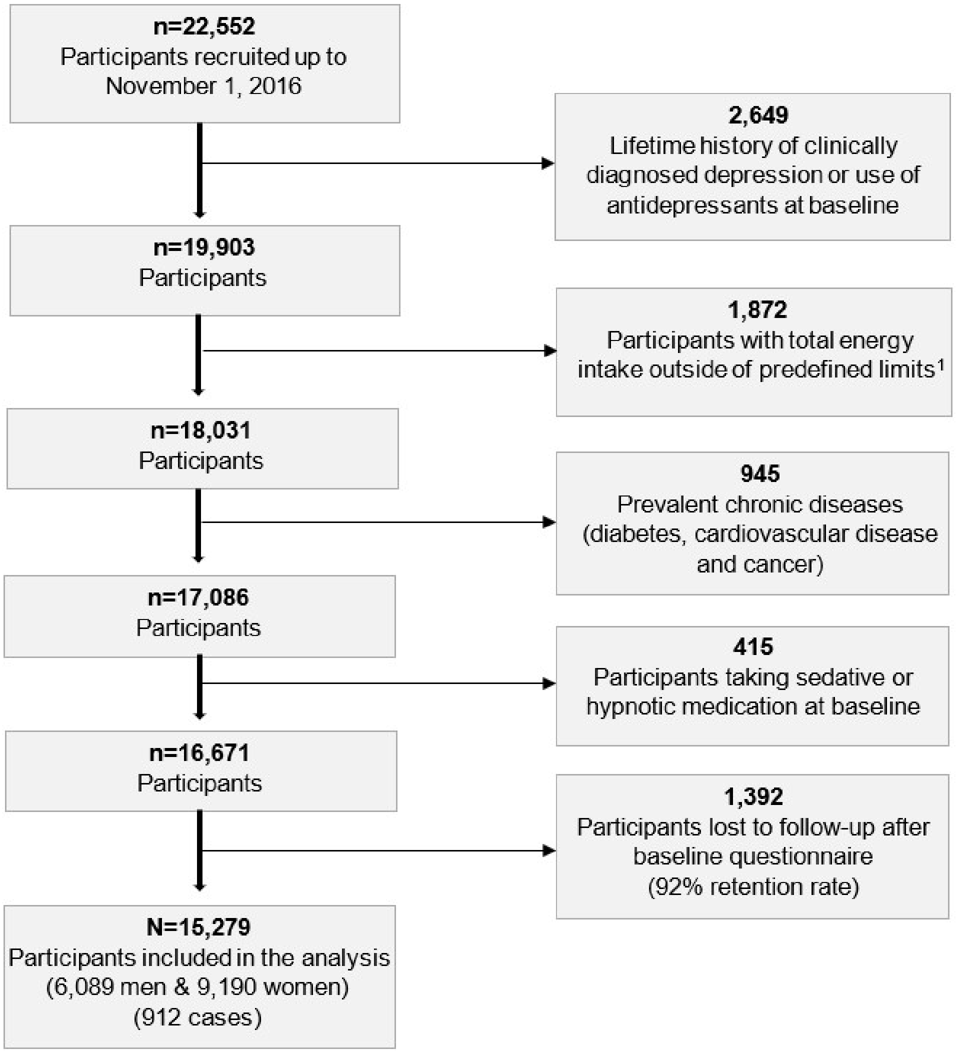
APPENDIX REFERENCES

1. Rico-Campà A, Martínez-González MA, Alvarez-Alvarez I, et al. Association between consumption of ultra-processed foods and all cause mortality: SUN prospective cohort study. *BMJ*. 2019;365:I1949. https://doi.org/10.1136/bmj.l1949.
2. Willett W. *Nutritional Epidemiology*. Third ed. New York, NY: Oxford University Press; 2012. https://doi.org/10.1093/acprof:oso/9780199754038.001.0001.
3. Trichopoulou A, Costacou T, Bamia C, Trichopoulos D. Adherence to a Mediterranean diet and survival in a Greek population. *N Engl J Med*. 2003;348(26):2599–2608. https://doi.org/10.1056/nejmoa025039.
4. Sotos-Prieto M, Moreno-Franco B, Ordovás JM, León M, Casasnovas JA, Peñalvo JL. Design and development of an instrument to measure overall lifestyle habits for epidemiological research: the Mediterranean Lifestyle (MEDLIFE) index. *Public Health Nutr*. 2015;18(6):959–967. https://doi.org/10.1017/s1368980014001360.
5. Gea A, Bes-Rastrollo M, Toledo E, et al. Mediterranean alcohol-drinking pattern and mortality in the SUN (Seguimiento Universidad de Navarra) Project: a prospective cohort study. *Br J Nutr*. 2014;111(10):1871–1880. https://doi.org/10.1017/s0007114513004376.

Supplementary Material 1.3

Hershey MS, Sanchez-Villegas A, Sotos-Prieto M, Fernández-Montero A, Pano O, Lahortiga F, Martínez-González MA, & Ruiz-Canela M. The Mediterranean Lifestyle and the Risk of Depression in Middle-Aged Adults. *The Journal of Nutrition*. 2021;131(2):604S-615S. doi:10.1093/jn/nxab333

Supplementary Figure 1. Flowchart of eligible participants in the SUN cohort 1999-2016.



[1]<800 kcal/day or >4000 kcal/day in men and <500 kcal/day or >3500 kcal/day in women[2]
[2]Willett W. *Nutritional Epidemiology*. third edit. New York, NY: Oxford University Press; 2012. doi:10.1093/acprof:oso/9780199754038.001.0001

Supplementary Table 1. Description of the Mediterranean Lifestyle (MEDLIFE) index modified for the SUN cohort.

Score items	Components (serving size)	Criteria for 1 point
Block 1: Mediterranean food consumption		
1. Sweets	Cookies, chocolate cookies, pastries, donuts, homemade baked goods, store-bought baked goods (50g), muffins (25-50g), tea biscuits (90g), chocolates (30g), churros (100g), turrón (35g)	≤ 2 servings/wk
2. Red meat	Beef, pork, lamb (100-150g)	< 2 servings/wk
3. Processed meat	Sausage, soft spicy sausage, bacon (50g), cured ham (60g), cooked ham (30g), hamburger (150g), liver (100-150g), organ meats (100-150g), pâté (25g)	≤ 1 serving/wk
4. Eggs	Eggs (1 unit)	2-4 servings/wk
5. Legumes	Lentils, beans, chickpeas, peas (60g uncooked)	≥ 2 servings/wk
6. White meat	Chicken/turkey with skin, chicken/turkey without skin, rabbit (100-150g)	2 servings/wk
7. Fish/seafood	White fish, fatty fish, codfish, salted or smoked fish, shrimp, octopus, calamari (100-150g), oysters and shellfish (6 units)	≥ 2 servings/wk
8. Potatoes	Baked or boiled potatoes (150g)	≤ 3 servings/wk
9. Low-fat dairy products	Skim milk (200cc), low-fat milk (200cc), low fat yogurt (125g), fresh soft cheese (50g)	2 servings/d
10. Nuts and olives	Almonds, peanuts, hazelnuts, walnuts (50g), olives (10 units)	1-2 servings/d
11. Sofrito	Olive oil, pepper, other vegetables (250g), tomato (150g)	>2/4 ingredients above the median
12. Fruit	Orange, banana, apple, pear, kiwi, mango, avocado, peach, apricot, nectarine (1 unit), clementine (2 units), strawberry (6 units), cherries, plums, figs, grapes (1 dessert plate), watermelon, melon (200-250g), dates and dried fruits (150g)	3-6 servings/d
13. Vegetables	Spinach, cauliflower, broccoli, lettuce, carrot, squash, green beans, eggplant, zucchini, cucumber, pepper, asparagus, gazpacho, garden salad, other vegetables (250g), tomato (150g) *(excludes potatoes)*	≥ 2 servings/d
14. Olive oil	Olive oil (13.5g)	≥ 3 servings/d
15. Cereals	White bread, whole-grain bread (3 slices), white rice, pasta (60g uncooked), pizza (200g), breakfast cereal (30g)	3-6 servings/d
Block 2: Mediterranean dietary habits		
16. Water	Tap water, bottled water (200 cc), coffee, decaffeinated coffee (50cc)	≥ 6 servings/d
17. Wine	Red/white wine (1 glass = 10g ethanol)	women:≤ 0.5 serving/d men:≤ 1 serving/d
18. Limit salt at meals	Do you limit salt at meals?	Yes
19. Preference for whole grains	Do you try to consume a lot of fiber? + fiber from grains	Yes, ≥ 6g/d fiber from grains
20. Snacks	Potato chips (150g)	< 1 serving/wk
21. Limit snacking in between meals	Do you tend to eat in between meals?	No
22. Limit sugar in beverages (including sugar-sweetened beverages)	Do you add sugar to some beverages? + soda + bottled juice (200cc)	No, ≤ 1 soda/wk, ≤ 1 bottled juice/wk
Block 3: Physical activity, rest, social habits, and conviviality		
23. Physical activity	Walking, jogging, running, climbing stairs, bicycling, stationary cycling, swimming, dance, aerobic exercise, martial arts, gymnastics, gardening, tennis, soccer, skiing, ice skating, team sports like basketball, other physical activities or sports	> 300 min/wk
24. Nap	Napping throughout the week	≤ 30 min/d
25. Hours of sleep	Sleeping throughout the week	6-8 h/d
26. Watching television	Watching TV/videos throughout the week	≤ 2 h/d
27. Socializing with friends	Socializing throughout the week	> 1 h/d
28. Collective sports	Playing soccer, tennis, squash, basketball and other team sports	≥ 1 h/wk

cc: cubic centimeter.

Supplementary Table 2. Age and sex-adjusted MEDLIFE characteristics according to MEDLIFE quartiles at baseline in the SUN cohort.

	MEDLIFE adherence			
Characteristic	**Q1 (3-10 pts.)**	**Q2 (11-12 pts.)**	**Q3 (13-14 pts.)**	**Q4 (15-23 pts.)**
N	4,865	4,387	3,520	2,507
Sweets (serv/wk)	8.4 (6.8)	7.5 (6.4)	6.6 (5.9)	5.7 (5.8)
Red meat (serv/wk)	4.0 (2.0)	3.7 (2.1)	3.3 (2.1)	2.7 (2.2)
Processed meat (serv/wk)	6.5 (4.2)	6.3 (4.2)	6.3 (4.3)	6.0 (4.3)
Eggs (serv/wk)	2.8 (2.1)	2.7 (1.9)	2.7 (1.6)	2.7 (1.5)
Legumes (serv/wk)	2.3 (2.0)	2.7 (2.2)	2.8 (2.1)	3.1 (1.9)
White meat (serv/wk)	2.0 (1.6)	2.1 (1.7)	2.2 (1.6)	2.4 (1.7)
Fish (serv/wk)	4.2 (2.7)	4.9 (2.8)	5.4 (3.0)	6.1 (3.4)
Potatoes (serv/wk)	1.3 (1.3)	1.2 (1.3)	1.3 (1.3)	1.4 (1.3)
Dairy products (serv/d)	1.1 (1.3)	1.3 (1.3)	1.5 (1.5)	1.8 (1.6)
Nuts and olives (serv/d)	0.3 (0.3)	0.3 (0.3)	0.3 (0.4)	0.4 (0.4)
Sofrito (%)	4.1	9.7	19.0	34.1
Fruit intake (serv/d)	1.8 (1.5)	2.3 (1.9)	2.8 (2.0)	3.44 (2.2)
Vegetable intake (serv/d)	1.7 (1.0)	2.2 (1.3)	2.7 (1.5)	3.4 (1.8)
Olive oil (g/d)	15.8 (13.0)	17.8 (14.5)	20.0 (15.3)	22.9 (16.6)
Cereals (serv/d)	1.7 (1.1)	1.8 (1.2)	2.0 (1.3)	2.3 (1.4)
Water and coffee (serv/d)	5.1 (2.8)	5.9 (2.9)	6.4 (2.9)	7.0 (2.9)
Wine (g ethanol/d) (median (IQR))	0 (0-1.8)	0.6 (0-3.8)	0.6 (0-3.8)	1.2 (0-3.8)
Men (median (IQR))	0.6 (0-4.4)	1.3 (0-5.0)	1.4 (0.6-5.0)	1.8 (0.6-5.0)
Women (median (IQR))	0 (0-1.2)	0.6 (0-1.3)	0.6 (0-1.3)	0.6 (0-1.3)
Limit salt at meals (% yes)	29.1	43.3	54.1	67.1
Try to consume a lot of fiber (% yes)	43.5	57.2	68.5	81.1
Fiber intake from cereals (g/d)	3.3 (2.2)	3.7 (2.7)	4.2 (3.2)	5.3 (3.8)
Snacks (serv/wk)	1.6 (1.6)	1.2 (1.3)	1.0 (1.2)	0.8 (1.1)
Tend to snack in between meals (% no)	52.5	66.9	73.8	83.5
Add sugar to some beverages (% no)	61.6	72.0	78.3	84.1
Sugar-sweetened beverages (serv/d)	0.3 (0.5)	0.2 (0.3)	0.2 (0.3)	0.1 (0.3)
Bottled juice (serv/d)	0.1 (0.3)	0.1 (0.3)	0.1 (0.3)	0.1 (0.3)
Physical activity (min/wk)	253.6 (313.9)	393.1 (391.6)	494.7 (428.9)	628.2 (482.0)
Siesta/Napping (min/d)	24.7 (57.1)	21.0 (46.4)	18.6 (33.4)	19.3 (30.5)
Sleeping (h/d)	7.2 (1.0)	7.2 (0.8)	7.2 (0.7)	7.2 (0.7)
Watching TV (h/d)	1.8 (1.5)	1.6 (1.2)	1.5 (1.1)	1.5 (1.0)
Socializing (h/d)	1.3 (1.2)	1.4 (1.1)	1.5 (1.1)	1.6 (1.1)
Collective sports (h/wk)	0.3 (1.1)	0.6 (1.5)	0.8 (1.8)	1.09 (2.0)

[1]Characteristics are age and sex adjusted using the inverse probability weighting method.
[2]Continuous variables are expressed as mean and standard deviation, unless otherwise stated.
[3]IQR: interquartile range; percentile 25 to percentile 75, serv: servings, TV: television.

Supplementary Table 3. Frequency of adherence to individual items of the MEDLIFE index in the SUN cohort.

Score items	Criteria for 1 point	% with 1 point
Block 1: Mediterranean food consumption		
1. Sweets	≤ 2 servings/wk	18.5
2. Red meat	< 2 servings/wk	24.7
3. Processed meat	≤ 1 serving/wk	11.7
4. Eggs	2-4 servings/wk	61.1
5. Legumes	≥ 2 servings/wk	56.2
6. White meat	2 servings/wk	14.3
7. Fish/seafood	≥ 2 servings/wk	88.9
8. Potatoes	≤ 3 servings/wk	97.4
9. Low-fat dairy products	2 servings/d	10.2
10. Nuts and olives	1-2 servings/d	5.0
11. Sofrito	> 2/4 ingredients above the median	14.0
12. Fruit	3-6 servings/d	22.6
13. Vegetables	≥ 2 servings/d	51.6
14. Olive oil	≥ 3 servings/d	7.3
15. Cereals	3-6 servings/d	14.1
Block 2: Mediterranean dietary habits		
16. Water and coffee	≥ 6 servings/d	52.2
17. Wine	women: ≤ 0.5 serving/d (0.1-5g/d) men: ≤ 1 serving/d (0.1-10g/d)	49.5
18. Limit salt at meals	Yes	45.2
19. Preference for whole grain products	Try to consume a lot of fiber (yes), > 6g/d fiber from cereals	12.0
20. Snacks	< 1 serving/wk	49.0
21. Limit snacking between meals	Tend to snack between meals (no)	66.7
22. Limit sugar in beverages	Add sugar to some beverages (no), Sugar-sweetened beverages < 1/wk, Bottled juice < 1/wk	67.5
Block 3: Physical activity, rest, social habits, and conviviality		
23. Physical activity	> 300 min/wk	51.9
24. Siesta/nap	≤ 30 min/d	54.8
25. Hours of sleep	6-8 h/d	95.1
26. Watching television	≤ 2 h/d	85.0
27. Socializing with friends	> 1 h/d	38.5
28. Collective sports	≥ 1 h/wk	21.4

Supplementary Table 4. Sources of variability for MEDLIFE scores in the SUN cohort.

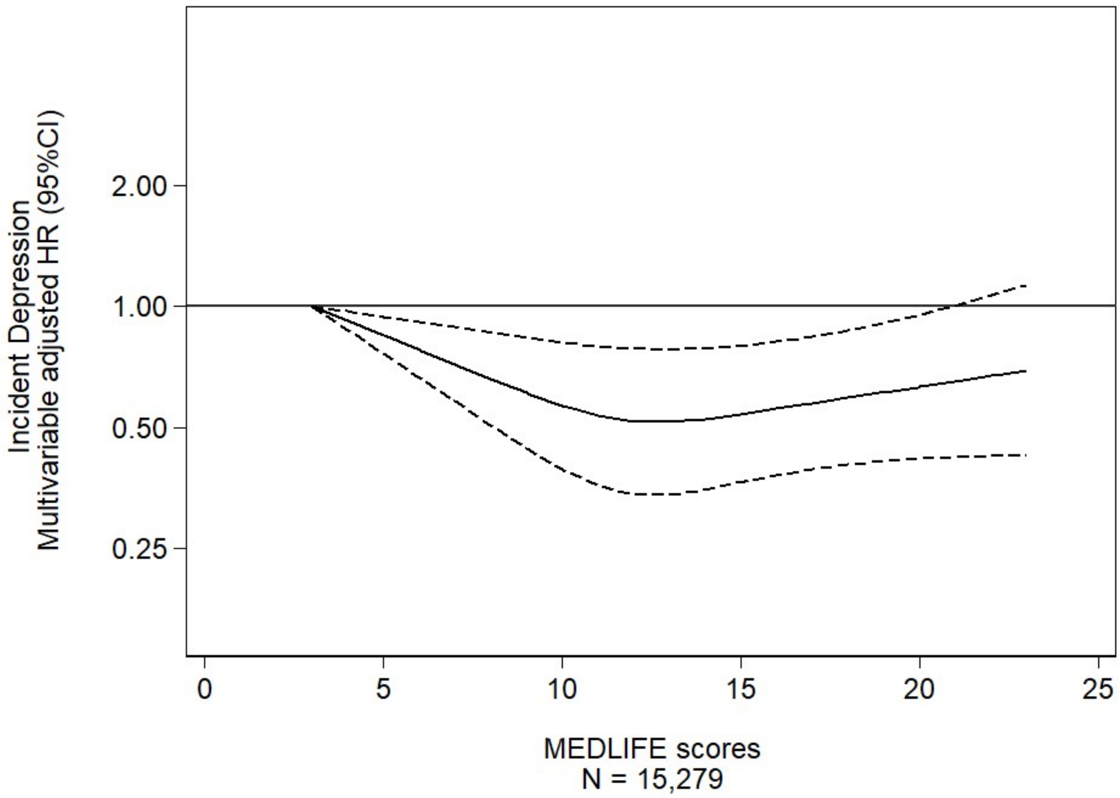
MEDLIFE item	Criteria for 1 point	Cumulative R^2	Change in R^2
Vegetables	≥ 2 servings/d	0.201	
Physical activity	> 300 min/wk	0.300	0.0995
Red meat	< 2 servings/wk	0.371	0.0707
Siesta/nap	≤ 30 min/d	0.423	0.0513
Fruit	3-6 servings/d	0.472	0.0493
Water and coffee	≥ 6 servings/d	0.520	0.0477
Limit salt at meals	Try to limit salt in meals (yes)	0.563	0.0438
Limit snacking between meals	Tend to snack between meals (no)	0.606	0.0432
Legumes	≥ 2 servings/wk	0.646	0.0391
Snacks	< 1 serving/wk	0.681	0.0353
Wine	women: ≤ 0.5 serving/d (0.1-5g/d) men: ≤ 1 serving/d (0.1-10g/d)	0.715	0.0340
Eggs	2-4 servings/wk	0.748	0.0328
Limit sugar in beverages	Add sugar to some beverages (no), Sugar-sweetened beverages < 1/wk, Bottled juice < 1/wk	0.777	0.0298
Preference for whole grain products	Try to consume a lot of fiber (yes), > 6 g/d fiber from cereals	0.807	0.0296
Socializing with friends	> 1 h/d	0.835	0.0280
Collective sports	≥ 1 h/wk	0.856	0.0209
Sweets	≤ 2 servings/wk	0.875	0.0194
Watching television	≤ 2 h/d	0.894	0.0184
Sofrito	> 2/4 ingredients above the median	0.911	0.0170
White meat	2 servings/wk	0.927	0.0166
Cereals	3-6 servings/d	0.942	0.0143
Low-fat dairy products	2 servings/d	0.955	0.0133
Fish/seafood	≥ 2 servings/wk	0.967	0.0121
Olive oil	≥ 3 servings/d	0.976	0.0089
Processed meat	≤ 1 serving/wk	0.984	0.0083
Nuts and olives	1-2 servings/d	0.991	0.0064
Hours of sleep	6-8 h/d	0.997	0.0060

[1]*Cumulative R^2 indicates the percentage of variability with the addition of each item to form the total MEDLIFE score.*
[2]*The change in R^2 indicates the variability corresponding to each item, indicating the most to least influential items for the difference in high and low scores.*
[3]*Potatoes are not featured because the stepwise-selection regressions and nested least-squares linear regressions require the exclusion of the least correlated item.*

Supplementary Table 5. Cox proportional hazard ratios (HR) and 95% confidence intervals (CI) for depression according to MEDLIFE quartiles.

	Categories of adherence to MEDLIFE				p for trend
	Q1 (3-10 pts)	Q2 (11-12 pts)	Q3 (13-14 pts)	Q4 (15-23 pts)	
N	4,865	4,387	3,520	2,507	
Incident cases	343	246	177	146	
Person-years	56491	50291	39082	26313	
Crude HR (95% CI)	1 Ref.	**0.81 (0.69-0.96)**	**0.75 (0.63-0.90)**	0.92 (0.76-1.12)	0.07
Age and sex adjusted HR (95% CI)	1 Ref.	**0.80 (0.68-0.94)**	**0.73 (0.61-0.87)**	0.85 (0.70-1.04)	**0.01**
Multivariable adjusted[1] HR (95% CI)	1 Ref.	**0.82 (0.69-0.96)**	**0.74 (0.61-0.89)**	0.89 (0.73-1.09)	**0.04**

[1]Age was the underlying time variable and the model was stratified by age group and year of recruitment.
[2]All models were adjusted for sex, BMI, total energy intake, special dieting, alcohol intake (not including wine), smoking status, cigarette pack-years, marital status, level of competitiveness, psychological tension, dependence, and hours working.
[3]N: observations, Q: quartile.



[1]Adjusted for age, year of recruitment, sex, BMI, total energy intake, special dieting, alcohol intake (not including wine), smoking status, cigarette pack-years, marital status, level of competitiveness, psychological tension, dependence, and hours working.

Supplementary Figure 2. Restricted cubic spline represents multivariable adjusted hazard ratio (HR) and 95% confidence intervals (CI) for each 1-point increment of MEDLIFE and risk of incident depression.

Supplementary Table 6. Sensitivity analyses for the association between MEDLIFE quartiles and the risk of incident depression.

			Multivariable adjusted HR (95% CI)			
Sensitivity Analysis	N	cases	Q1 (3-10 pts)	Q2+Q3 (11 pts-14 pts)	Q4 (15-23 pts)	P for trend
1 extra point for non-smokers	15,279	912	1 Ref.	0.82 (0.70-0.95)	0.90 (0.76-1.08)	0.14
1 extra point for nondrinkers in addition to light wine consumers	15,279	912	1 Ref.	0.82 (0.71-0.95)	0.92 (0.76-1.12)	0.18
Including participants taking sedative or hypnotic medication at baseline	15,659	954	1 Ref.	0.78 (0.68-0.90)	0.89 (0.73-1.09)	0.04
Outcome defined as having both a reported diagnosis and treatment of depression	15,279	314	1 Ref.	0.80 (0.63-1.02)	0.75 (0.52-1.07)	0.06
Exclusions						
Missing ≥30 FFQ items	14,841	876	1 Ref.	0.78 (0.67-0.90)	0.86 (0.70-1.05)	0.02
Diagnosed depression within first 2 years of follow-up	15,051	684	1 Ref.	0.76 (0.64-0.90)	0.88 (0.69-1.11)	0.05
Substitutions						
Trichopoulou's Mediterranean diet score (0-8 pts)[1] instead of Block 1	15,279	912	1 Ref.	0.86 (0.74-0.99)	0.89 (0.72-1.09)	0.11
High adherence (7-10 pts) to MADP[2] instead of wine	15,279	912	1 Ref.	0.81 (0.70-0.94)	0.90 (0.75-1.08)	0.10
Physical activity (>150 min/wk)	15,279	912	1 Ref.	0.78 (0.68-0.91)	0.86 (0.70-1.05)	0.03
Siesta/nap on weekends (yes)	15,279	912	1 Ref.	0.81 (0.70-0.94)	0.99 (0.81-1.22)	0.32
Hours of sleep (6-8 h/d weekdays)	15,279	912	1 Ref.	0.84 (0.72-0.98)	0.86 (0.72-1.03)	0.06
Watching television (<1 h/d weekdays)	15,279	912	1 Ref.	0.87 (0.75-1.00)	0.91 (0.76-1.09)	0.17
Socializing with friends (≥2 h/d weekends)	15,279	912	1 Ref.	0.82 (0.71-0.95)	0.90 (0.74-1.10)	0.14
Collective sports (≥2 h/wk)	15,279	912	1 Ref.	0.76 (0.66-0.88)	0.90 (0.74-1.11)	0.04
Original criteria used by Sotos-Prieto[3] for block 3: physical activity, rest, social habits, and conviviality	15,279	912	1 Ref.	0.88 (0.75-1.02)	0.96 (0.80-1.14)	0.45

[1]Trichopoulou A, Costacou T, Bamia C, Trichopoulos D. Adherence to a Mediterranean Diet and Survival in a Greek Population. *N Engl J Med*. 2003;348(26):2599-2608. doi:10.1056/NEJMoa025039

[2]Mediterranean alcohol drinking pattern 8-item score proposed by Gea et. al.[4]

[3]Sotos-Prieto M, Moreno-Franco B, Ordovás JM, León M, Casasnovas JA, Peñalvo JL. Design and development of an instrument to measure overall lifestyle habits for epidemiological research: the Mediterranean Lifestyle (MEDLIFE) index. *Public Health Nutr*. 2015;18(6):959-967. doi:10.1017/S1368980014001360

[4]Morales G, Martínez-González MA, Barbería-Latasa M, Bes-Rastrollo M, Gea A. Mediterranean diet, alcohol-drinking pattern and their combined effect on all-cause mortality: the Seguimiento Universidad de Navarra (SUN) cohort. *Eur J Nutr*. 2021;60(3):1489-1498. doi:10.1007/s00394-020-02342-w

[5]FFQ: food frequency questionnaire, N: observations, Q: quartile.

2.A. Invitation to enroll in the "Feeding America's Bravest" trial

[Fishers Fire Department Letterhead]

Mediterranean Diet Nutrition Intervention: **Feeding America's Bravest**

Dear Fishers Firefighter,

Researchers across the United States are working to decrease firefighters' risks of heart disease and cancer. One area of health and wellness that scientists are actively studying is **nutrition**. The Fishers Fire Department is delighted to partner with Dr. Kales, a Professor at Harvard and Dr. Steven Moffatt of Public Safety Medical on a Department of Homeland Security-funded research study involving nutrition, entitled "**Feeding America's Bravest**".

This study will be conducted at the Fishers Fire Department over a one-year period. Using a method similar to a coin toss, the researchers will divide all Fishers fire stations into two groups. Fire stations in Group 1 will receive a Mediterranean diet intervention over the first six months of 2018. Group 2 will have no changes the first six months. In the second half of 2018, Group 1 will try to continue the Mediterranean way of eating on its own, and Group 2 will receive the Mediterranean diet intervention for six months. Therefore, all participants will have the chance to benefit from the study. During the intervention, firefighters and their families will have access to discounts for healthy food purchases and educational programs and materials to healthy eating.

Possible benefits of participating include improving your health and sense of well-being. Mediterranean diets are proven to decrease the risk of heart disease, cancer, and many other types of chronic disease and are associated with living longer and better. Based on the study's ability to help firefighters, it has been endorsed by the International Association of Fire Fighters and the National Fallen Firefighter Foundation.

The researchers have done previous national surveys of firefighters. In these surveys, firefighters most often chose the Mediterranean diet as the most popular way to eat healthier.

Over the next several weeks, the researchers will be meeting with each Fishers fire station to speak with our members about the study and offer them the opportunity to participate. You will receive more information from the researchers and have the opportunity to review the study's consent (permission) form (attached for your reference) and ask any questions in private.

We want to assure Fishers members that participation in the study is completely voluntary. Participation or non-participation has no bearing on your employment at the Fishers Fire Department or on the occupational health care that you receive from Public Safety Medical.

If you have questions about this research study, you can contact Steven M. Moffatt, MD (317-972-1180) or Stefanos N. Kales, MD (617-665-1580, skales@hsph.harvard.edu).

Sincerely,

[Chief Orusa signature] [Union signature]

HARVARD UNIVERSITY | **Dietary Assessment** | DA80 Printed in U.S.A. | **Page 1**

Please use #2 pencil only.

ID: ______ — __

0 1 2 3 4 5 6 7 8 9

1. Do you currently take multi-vitamins? (Please report other individual vitamins in the next section.)

○ No — a) How many do you take per week? ○ 2 or less ○ 3–5 ○ 6–9 ○ 10 or more

○ Yes ▶ b) What specific brand (or equivalency) do you usually take?

○ Centrum Silver ○ Centrum ○ Other ➡ e.g., AARP Alphabet II Formula 643 Multivitamins and Minerals

○ Theragran M ○ One-A-Day Essential

Not counting multi-vitamins, do you take any of the following preparations?

Preparation				Dose per day:					
a) Vitamin A	○ No	○ Yes, seasonal only ○ Yes, most months	If Yes,	Dose per day:	○ Less than 10,000 IU	○ 10,000 to 15,000 IU	○ 16,000 to 22,000 IU	○ 23,000 IU or more	○ Don't know
b) Potassium	○ No	○ Yes	If Yes,	Dose per day:	○ Less than 2.5 mEq (100 mg)	○ 3 to 10 mEq	○ 11 to 20 mEq	○ 21 mEq or more	○ Don't know
c) Vitamin C	○ No	○ Yes, seasonal only ○ Yes, most months	If Yes,	Dose per day:	○ Less than 400 mg	○ 400 to 700 mg	○ 750 to 1250 mg	○ 1300 mg or more	○ Don't know
d) Vitamin B_6	○ No	○ Yes	If Yes,	Dose per day:	○ Less than 50 mg	○ 50 to 99 mg	○ 100 to 149 mg	○ 150 mg or more	○ Don't know
e) Vitamin E	○ No	○ Yes	If Yes,	Dose per day:	○ Less than 100 IU	○ 100 to 250 IU	○ 300 to 500 IU	○ 600 IU or more	○ Don't know
				Type:	○ Natural	○ Regular (dl)	○ Unknown		
f) Calcium (Include Calcium in Tums, etc.)	○ No	○ Yes	If Yes,	Dose per day (elemental calcium):	○ Less than 600 mg	○ 600 to 900 mg	○ 901 to 1500 mg	○ 1501 mg or more	○ Don't know
g) Selenium	○ No	○ Yes	If Yes,	Dose per day:	○ Less than 80 mcg	○ 80 to 130 mcg	○ 140 to 250 mcg	○ 260 mcg or more	○ Don't know
h) Vitamin D (In calcium supplement or separately)	○ No	○ Yes, seasonal only ○ Yes, most months	If Yes,	Dose per day:	○ Less than 300 IU	○ 300 to 500 IU	○ 600 to 900 IU	○ 1000 IU or more	○ Don't know
i) Zinc	○ No	○ Yes	If Yes,	Dose per day:	○ Less than 25 mg	○ 25 to 74 mg	○ 75 to 100 mg	○ 101 mg or more	○ Don't know

2. Are there other supplements that you take on a regular basis?

○ Metamucil/Citrucel ○ Flax Seed ○ Niacin ○ Choline ○ DHEA

○ Cod Liver Oil ○ Beta-carotene ○ Chromium ○ Folic Acid ○ Iron ○ Other (Please specify)

○ Vitamin B_{12} ○ Magnesium ○ Lecithin ○ B-Complex

○ Flax Seed Oil ○ Fish oil ○ Coenzyme Q_{10} ○ Lycopene

3. How many teaspoons of sugar do you add to your beverages or food each day? ➡ ____ tsp.

4. What brand and type of cold breakfast cereal do you usually eat? ➡ Specify cereal brand & type (e.g., Kellogg's Raisin Bran)

○ Don't eat cold breakfast cereal.

5. What form of margarine or spread do you usually use (exclude pure butter)? ➡ What specific brand & type of margarine (e.g., Shedd's Country Crock plus calcium and vitamins)

○ None **Form?** ○ Stick ○ Tub ○ Spray ○ Squeeze (liquid)

Type? ○ Reg ○ Light ○ Nonfat

6. For each food listed, fill in the circle indicating how often on average you have used the amount specified during the past year.

Ch rb cf sw gn t k w

V S M

AVERAGE USE LAST YEAR

DAIRY FOODS		Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
Milk (8 oz. glass)	Skim milk	○	○	W	○	○	D	○	○	○
	1 or 2 % milk	○	○	W	○	○	D	○	○	○
	Whole milk	○	○	W	○	○	D	○	○	○
	Soy milk	○	○	W	○	○	D	○	○	○
Cream, e.g., coffee, whipped or sour cream (1 Tbs)		○	○	W	○	○	D	○	○	○
Non-dairy coffee whitener (1 Tbs)		○	○	W	○	○	D	○	○	○
Frozen yogurt, sherbet or low-fat ice cream (1 cup)		○	○	W	○	○	D	○	○	○
Regular ice cream (1 cup)		○	○	W	○	○	D	○	○	○
Yogurt (1 cup)	Low-carb, artificially sweetened or plain	○	○	W	○	○	D	○	○	○
	Sweetened–with fruit or other flavoring	○	○	W	○	○	D	○	○	○
Spreads added to food or bread; exclude use in cooking	Margarine	○	○	W	○	○	D	○	○	○
	Pure Butter	○	○	W	○	○	D	○	○	○
Cottage or ricotta cheese (1/2 cup)		○	○	W	○	○	D	○	○	○
Cream cheese (1 oz.)		○	○	W	○	○	D	○	○	○
Other cheese, e.g., American, cheddar, etc., plain or as part of a dish (1 slice or 1 oz. serving)		○	○	W	○	○	D	○	○	○

➡ **What type of cheese do you usually eat?** ○ Soy ○ Regular ○ Low fat or Lite ○ Nonfat ○ None

6. ***(continued)*** **For each food listed, fill in the circle indicating how often on average you have used the amount specified during the past year.**

Please try to average your seasonal use of foods over the entire year. For example, if a food such as cantaloupe is eaten 4 times a week during the approximate 3 months that it is in season, then the average use would be once per week.

FRUITS	Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
Raisins (1 oz. or small pack) or grapes (1/2 cup)			W			D			
Prunes or dried plums (6 prunes or 1/4 cup)			W			D			
Prune juice (small glass)			W			D			
Bananas (1)			W			D			
Cantaloupe (1/4 melon)			W			D			
Avocado (1/2 fruit or 1/2 cup)			W			D			
Fresh apples or pears (1)			W			D			
Apple juice or cider (small glass)			W			D			
Oranges (1)			W			D			
Orange juice (small glass) – Calcium fortified			W			D			
Orange juice (small glass) – Regular (not calcium fortified)			W			D			
Grapefruit (1/2) or grapefruit juice (small glass)			W			D			
Other fruit juices (small glass)			W			D			
Strawberries, fresh, frozen or canned (1/2 cup)			W			D			
Blueberries, fresh, frozen or canned (1/2 cup)			W			D			
Peaches or plums (1 fresh or 1/2 cup canned)			W			D			
Apricots (1 fresh, 1/2 cup canned or 5 dried)			W			D			

VEGETABLES	Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
Tomatoes (2 slices)			W			D			
Tomato or V-8 juice (small glass)			W			D			
Tomato sauce (1/2 cup) e.g., spaghetti sauce			W			D			
Salsa, picante or taco sauce (1/4 cup)			W			D			
String beans (1/2 cup)			W			D			
Beans or lentils, baked, dried or soup (1/2 cup)			W			D			
Tofu, soy burger, soybeans, miso or other soy protein			W			D			
Peas or lima beans (1/2 cup fresh, frozen, canned)			W			D			
Broccoli (1/2 cup)			W			D			
Cauliflower (1/2 cup)			W			D			
Cabbage or coleslaw (1/2 cup)			W			D			
Brussels sprouts (1/2 cup)			W			D			
Carrots, raw (1/2 carrot or 2–4 sticks)			W			D			
Carrots, cooked (1/2 cup) or carrot juice (2–3 oz.)			W			D			
Corn (1 ear or 1/2 cup frozen or canned)			W			D			
Mixed or stir-fry vegetables (1/2 cup), veg. soup (1 cup)			W			D			
Yams or sweet potatoes (1/2 cup)			W			D			
Dark orange (winter) squash (1/2 cup)			W			D			
Eggplant, zucchini or other summer squash (1/2 cup)			W			D			
Kale, mustard greens or chard (1/2 cup)			W			D			
Spinach, cooked (1/2 cup)			W			D			
Spinach, raw as in salad (1 cup)			W			D			
Iceberg or head lettuce (1 serving)			W			D			
Romaine or leaf lettuce (1 serving)			W			D			
Celery (2–3 sticks)			W			D			
Peppers: green, yellow or red (3 slices)			W			D			
Onions as a garnish or in salad (1 slice)			W			D			
Onions as a cooked vegetable, rings or soup (1/2 cup)			W			D			

EGGS, MEAT, ETC.	Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
Eggs (1) – Omega-3 fortified including yolk			W			D			
Eggs (1) – Regular eggs including yolk			W			D			
Beef or pork hot dogs (1)			W			D			
Chicken or turkey hot dogs or sausage (1)			W			D			
Chicken/turkey sandwich or frozen dinner			W			D			
Other chicken or turkey, with skin (3 oz.)			W			D			
Other chicken or turkey, without skin (3 oz.)- including ground			W			D			
Bacon (2 slices)			W			D			

6. ***(continued)*** **For each food listed, fill in the circle indicating how often on average you have used the amount specified during the past year.**

EGGS, MEAT, ETC.		Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
Salami, bologna, or other processed meat sandwiches										
Other processed meats, e.g., sausage, kielbasa, etc. (2 oz. or 2 small links)										
Hamburger (1 patty)	Lean or extra lean									
	Regular									
Beef, pork, or lamb as a sandwich or mixed dish, e.g., stew, casserole, lasagna, frozen dinners, etc.										
Pork as a main dish, e.g., ham or chops (4–6 oz.)										
Beef or lamb as a main dish, e.g., steak, roast (4–6 oz.)										
Canned tuna fish (3–4 oz.)										
Breaded fish cakes, pieces, or fish sticks (1 serving, store bought)										
Shrimp, lobster, scallops as a main dish										
Dark meat fish, e.g., tuna steak, mackerel, salmon, sardines, bluefish, swordfish (3–5 oz.)										
Other fish, e.g., cod, haddock, halibut (3–5 oz.)										

BREADS, CEREALS, STARCHES		Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
Cold breakfast cereal (1 serving)										
Cooked oatmeal/cooked oat bran (1 cup)										
Other cooked breakfast cereal (1 cup)										
Bread (1 slice)	White bread, including pita									
	Rye/Pumpernickel									
	Whole wheat, oatmeal, other whole grain									
Crackers, regular or lowfat e.g., Triscuits, Ritz (6)										
Bagels, English muffins, or rolls (1)										
Muffins or biscuits (1)										
Pancakes or waffles (2 small pieces)										
Brown rice (1 cup)										
White rice (1 cup)										
Pasta, e.g., spaghetti, noodles, couscous, etc. (1 cup)										
Tortillas (2)										
French Fries (6 oz. or 1 serving)										
Potatoes, baked, boiled (1) or mashed (1 cup)										
Potato chips or corn/tortilla chips (small bag or 1 oz.)										
Pizza (2 slices)										

		BEVERAGES	Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
CARBONATED BEVERAGES Consider the serving size as 1 glass, bottle or can for these carbonated beverages.	Low-Calorie (sugar-free) types	Low-calorie beverage with caffeine, e.g., Diet Coke, Diet Mt. Dew									
		Other low-cal bev. without caffeine, e.g., Diet 7-Up									
	Regular types (not sugar-free)	Carbonated beverage with caffeine & sugar, e.g., Coke, Pepsi, Mt. Dew, Dr. Pepper									
		Other carbonated beverage with sugar, e.g., 7-Up, Root Beer, Ginger Ale, Caffeine-Free Coke									
OTHER BEVERAGES		Other sugared beverages: Punch, lemonade, sports drinks, or sugared ice tea (1 glass, bottle, can)									
		Beer, regular (1 glass, bottle, can)									
		Light Beer, e.g., Bud Light (1 glass, bottle, can)									
		Red wine (5 oz. glass)									
		White wine (5 oz. glass)									
		Liquor, e.g., vodka, gin, etc. (1 drink or shot)									
		Water: bottled, sparkling, or tap (8 oz. cup)									
		Herbal tea or decaffeinated tea (8 oz. cup)									
		Tea with caffeine (8 oz. cup), including green tea									
		Decaffeinated coffee (8 oz. cup)									
		Coffee with caffeine (8 oz. cup)									
		Dairy coffee drink (hot/cold) e.g., Cappuccino (16 oz.)									

6. ***(continued)*** **For each food listed, fill in the circle indicating how often on average you have used the amount specified during the past year.**

SWEETS, BAKED GOODS, MISCELLANEOUS		Never, or less than once per month	1–3 per month	1 per week	2–4 per week	5–6 per week	1 per day	2–3 per day	4–5 per day	6+ per day
Milk chocolate (bar or pack), e.g., Hershey's, M&M's				W			D			
Dark chocolate, e.g., Hershey's Dark or Dove Dark				W			D			
Candy bars, e.g., Snickers, Milky Way, Reeses				W			D			
Candy without chocolate (1 oz.)				W			D			
Cookies (1)	Fat free or reduced fat			W			D			
	Other			W			D			
Brownies (1)				W			D			
Doughnuts (1)				W			D			
Cake	Fat free or reduced fat			W			D			
	Other			W			D			
Pie, homemade or ready made (slice)				W			D			
Jams, jellies, preserves, syrup, or honey (1 Tbs)				W			D			
Peanut butter (1 Tbs)				W			D			
Popcorn (3 cups)	Fat free or light			W			D			
	Regular			W			D			
Sweet roll, coffee cake or other pastry (serving)	Fat free or reduced fat			W			D			
	Other			W			D			
Breakfast bars, e.g., Nutrigrain, granola, Kashi (1)				W			D			
Energy bars, e.g., Clif, Luna, Glucerna, Powerbar (1)				W			D			
Low carb bars, e.g., Atkins, Zone, South Beach (1)				W			D			
Pretzels (1 small bag or serving)				W			D			
Peanuts (small packet or 1 oz.)				W			D			
Walnuts (1 oz.)				W			D			
Other nuts (small packet or 1 oz.)				W			D			
Oat bran, added to food (1 Tbs)				W			D			
Other bran (wheat, etc.), added to food (1 Tbs)				W			D			
Chowder or cream soup (1 cup)				W			D			
Ketchup or red chili sauce (1 Tbs)				W			D			
Splenda (1 packet)				W			D			
Other artificial sweetener (1 packet)				W			D			
Olive oil added to food or bread (1 Tbs)				W			D			
Low-fat or fat-free mayonnaise (1 Tbs)				W			D			
Regular mayonnaise (1 Tbs)				W			D			
Salad dressing (1-2 Tbs)				W			D			

Type of salad dressing: ◯ Nonfat ◯ Low-fat ◯ Olive oil ◯ Other vegetable oil

7. Liver: (beef, calf or pork 4 oz.) ◯ Never ◯ Less than 1/mo ◯ 1/mo ◯ 2–3/mo ◯ 1/week or more

Liver: (chicken or turkey 1 oz.) ◯ Never ◯ Less than 1/mo ◯ 1/mo ◯ 2–3/mo ◯ 1/week or more

8. How often do you eat fried or sautéed food at home? (Exclude "Pam"-type spray)
◯ Less than once a week ◯ 1–3 times per week ◯ 4–6 times per week ◯ Daily

9. What kind of fat is usually used for frying and sautéing at home? (Exclude "Pam"-type spray)
◯ Real butter ◯ Margarine ◯ Olive oil ◯ Vegetable oil ◯ Veg. shortening ◯ Lard ◯ N/A

10. What kind of fat is usually used for baking at home?
◯ Real butter ◯ Margarine ◯ Olive oil ◯ Vegetable oil ◯ Veg. shortening ◯ Lard ◯ N/A

11. What type of cooking oil is usually used at home? (e.g., Mazola Corn Oil) Specify brand and type →

12. How often do you eat deep fried chicken, fish, shrimp, clams or onion rings away from home?
◯ Less than once a week ◯ 1–3 times per week ◯ 4–6 times per week ◯ Daily

13. How often do you eat toasted breads, bagel or English muffin (e.g., slice or 1 half bagel)?
◯ Less than once a week ◯ 1–3 times per week ◯ 4–6 times per week ◯ Daily ◯ 2+ times/day

14. Are there any other important foods that you usually eat at least once per week?

Include for example: Applesauce, mushrooms, bulgur, radish, horseradish, Eggbeaters, dates, figs, rhubarb, mango, mixed dried fruit, papaya, wheat germ, custard, venison, hot peppers, pickles, olives, SlimFast, Ensure (regular or plus), Glucerna Shake.

(Do not include dry spices and do not list something that has been listed in the previous sections.)

Other foods that you usually eat at least once per week	Servings per week
(a)	
(b)	
(c)	

2.C. Table of modified criteria for 1 point in the "Feeding America´s Bravest" trial from the original MEDLIFE index designed and validated by Sotos-Prieto et. al.

Item	Description of components derived from baseline FFQ in the FAB trial (serving size)	Criteria for 1 point in *Feeding America's Bravest*	Original criteria for 1 point
1. Sweets	milk chocolate, dark chocolate, candy bars, candy (1 oz.), cookies, brownies, doughnuts, cake, pie, muffins, biscuits (1 unit) pancakes, waffles (2 small units)	≤ 2 servings/wk	≤ 2 servings/wk
2. Red meat	Beef, pork, lamb as main dish, mixed dish, or sandwich (4-6 oz.)	< 2 servings/wk	< 2 servings/wk
3. Processed meats	Hamburger, hotdog (1 unit), salami, bologna, other processed meat (2 oz.), chicken or turkey hotdogs or sandwich, sausage, frozen dinner (1 unit), bacon (2 slices), beef liver (4 oz.), chicken liver (1 oz.)	≤ 1 serving/wk	≤ 1 serving/wk
4. Eggs	Regular eggs including yolk and omega-3 fortified including yolk	2-4 units/wk	2-4 servings/wk
5. Legumes	Beans or lentils, baked, dried, or soup, peas, lima beans (1/2 cup)	≥ 2 servings/wk	≥ 2 servings/wk
6. White meat	Chicken or turkey with or without skin (3 oz.)	2 servings/wk	2 servings/wk
7. Fish	Dark meat fish (tuna steak, mackerel, salmon, sardines, bluefish, swordfish), other fish (3-5 oz.), canned tuna (3-4 oz.), shrimp, lobster, scallops as a main dish	≥ 2 servings/wk	≥ 2 servings/wk
8. Potatoes	potatoes, baked, boil (1 unit) or mashed (1 cup) and French fries (6 oz. or 1 serving)	≤ 3 servings/wk	≤ 3 servings/wk
9. Dairy products	Skim, 1 or 2%, whole, soy milk (8 oz.), cream (1 Tbs), frozen yogurt, sherbet, ice cream, plain or sweetened yogurt (1 cup), cottage or ricotta cheese (1/2 cup), margarine, butter, cream cheese, other cheese (1 oz./1 slice)	2 servings/d	2 servings/d
10. Nuts*	Nuts (e.g. walnuts, almonds, hazelnuts, pistachio, peanuts)	1-2 serving/d	1-2 serving/d
11. Fruit	Raisins (1 oz), grapes (1/2 cup), prunes (6 units), apple, orange, grapefruit, prune, and other fruit juices (small glass), bananas (1 unit), cantaloupe (1/4 melon), grapefruit, avocado (1/2 fruit or cup), apples, pears, oranges, peaches, plums, apricots (1 unit), strawberries, blueberries fresh, frozen, or canned (1/2 cup)	3-6 servings/d	3-6 servings/d
12. Vegetables	Tomatoes (2 slices), tomato or carrot juice (small glass), broccoli, string beans, cauliflower, cabbage, Brussel sprouts, raw or cooked carrots, corn, mixed vegetables, yams, sweet potatoes, squash, eggplants, zucchini, kale, cooked spinach, cooked onions (1/2 cup), spinach (1 cup), head or leaf lettuce (1 serving), celery (2-3 sticks), peppers (3 slices), raw onion (1 slice)	≥ 2 servings/d	≥ 2 servings/d
13. Olive oil	Olive oil added to food or bread (1 Tbs.) Main oil usually used for frying and sautéing at home?	≥ 1 servings/d + Olive oil	≥ 3 servings/d
14. Cereals	Cold breakfast cereal (1 serving), oatmeal (1 cup), other cooked cereals (1 cup) white bread (1 slice), rye bread (1 slice), whole grain bread (1 slice), English muffin, bagel, rolls (1 unit), brown rice (1 cup), white rice (1 cup), pasta, noodles, couscous (1cup), tortillas (2), pizza (2 slices)	3-6 servings/d	3-6 servings/d

15. Water, coffee, and tea	Which of the following non-alcoholic beverages do you most frequently drink at home? Which of the following non-alcoholic beverages do you most frequently drink at the firehouse? Water (bottled, sparkling, or tap), herbal tea, decaffeinated tea, tea with caffeine, decaffeinated coffee, coffee with caffeine (8 oz.)	Water, coffee, and tea are most frequent non-alcoholic drinks at home and at the firehouse + ≥ 6 cups/d of water, coffee, or tea	6-8 servings/d water or ≥ 3 servings/wk tea
16. Wine	When you drink alcoholic beverages, what type do you drink?	Red or white wine are usual alcoholic beverage	1-2 servings/d
17. Limit salt	Sodium (mg)	< 2.3 g/d	yes
18. Preference for whole grain products	Is whole grain the main type of bread or starch that you eat?	Yes	yes/fiber >25g/d
19. Snacks	Regular popcorn (3 cups), fat free popcorn (3 cups), potato chips (small bag or 1 oz), crackers (6), pretzels (1 small bag or serving), breakfast, energy, and low carb bars (1 unit)	≤ 2 serving/wk	≤ 2 serving/wk
20. Limit sugar in beverages (sugary beverages)	Carbonated beverages caffeinated or caffeine-free with sugar and other sugared beverages: punch, iced tea, lemonade, sports drinks (1 glass, bottle, or can)	≤ 1 serving/wk	yes
21. Local, seasonal, or organic products	Do you usually consume local/seasonal or organic products?	Yes	---
22. Physical activity >150 min/wk	Statement that best describes your physical activity in the past month	Run over 10 miles/wk or spend over 3 hrs/wk in comparable PA	yes
23. Siesta/nap	How many times do you take a nap per week?	Regular naps (≥ 3/wk)	yes
24. Hours of sleep	Total hours of actual sleep in a typical 24-hour period	6-8 hours/d	6-8h/d
25. Watching TV	During the past weeks, what was your average total time per week at each of the following activities?	≤ 4 hours/wk	< 1h/d
26. Time spent eating	Time you usually spend eating each meal at home (in minutes). Time you usually spend eating each meal at the firehouse (in minutes).	≥ 20 minutes	---

__ indicates modified criteria. d: day, FAB: Feeding America's Bravest, g: grams, h: hours, mg: milligrams, min: minutes, oz: ounces, pts: points, TV: television, wk: week
Eliminated items: Herbs, spices, and garnish; Snacking in between meals; Socializing with friends; Collective sports.
Added items: Local, seasonal, or organic products; Time spent eating
*Olive consumption was unavailable to consider the combined intake with nuts in item 10 (originally nuts and olives).

2.D. Baseline characteristics of the participants by group assignment.

Characteristic	Control Group	Mediterranean Diet Nutrition Intervention	*p* *
N	244	241	-
Female (%)	4.10	6.64	0.214
Age (yrs)	46.28 (7.74)	45.18 (8.55)	0.128
mMDS (pts)	21.17 (11.10)	19.64 (11.52)	0.636
Total energy intake (kcal/d)	2481.19 (1035.87)	2573.01 (1347.14)	0.400
Protein intake (g/d)	107.29 (44.83)	113.93 (78.73)	0.254
Carbohydrate intake (g/d)	265.91 (115.52)	281.13 (152.32)	0.215
Whole grains (g/d)	37.07 (31.43)	37.79 (31.41)	0.801
Total fiber intake (g/d)	24.36 (11.76)	24.89 (16.26)	0.683
Added sugar (g/d)	64.47 (45.15)	75.35 (57.84)	0.021
Fat intake (g/d)	102.06 (47.99)	104.90 (55.95)	0.548
Saturated fat (g/d)	33.73 (16.74)	34.91 (18.80)	0.466
Polyunsaturated fat (g/d)	21.60 (10.48)	22.10 (12.65)	0.634
Monounsaturated fat (g/d)	38.50 (19.27)	39.36 (21.57)	0.644
Alcohol (g/d)	13.94 (21.41)	11.08 (15.95)	0.097
Nondrinkers (%)	17.62	16.60	0.764
Smoking status (%)			0.869
never	33.20	35.27	
current	50.82	48.55	
former	15.98	16.18	
Education (%)			0.240
Technical school/some college/associates degree	72.95	68.46	
Bachelor's degree	25.82	30.29	
Postgraduate degree	1.23	0.41	
Civil status (%)			0.014
married	90.57	82.99	
single	9.43	17.01	
Vitamin use (%)	18.03	28.63	0.006
Supplement use (%)	14.75	20.33	0.106
Sitting (hrs/wk)	20.37 (12.93)	19.84 (12.10)	0.643
Physical activity (%)			0.061
no regular exercise	6.56	12.03	
regular modest exercise	44.67	37.34	
regular heavy exercise	48.77	50.62 |	
TV, computer, and driving (hrs/wk)	6.53 (3.72)	6.45 (4.16)	0.824
Sleep (hrs/d)	6.51 (0.70)	6.4 (0.78)	0.197

Values are means (SD) except for qualitative variables, expressed as n (%).
[*] *P* value for comparisons between groups by chi square test for categorical variables and one-way ANOVA for continuous variables.

2.D. (continued) Baseline levels of anthropometric measurements and cardiometabolic parameters by group assignment.

Characteristic	Control Group (n=241)	Mediterranean Diet Nutrition Intervention (n=244)	*p* *
BMI (kg/m^2)	30.13 (34.36)	29.93 (4.34)	0.593
Weight (kg)	216.73 (38.54)	211.08 (36.22)	0.097
Waist circumference (cm)	101.12 (12.26)	98.29 (12.36)	0.012
Body fat (%)	27.62 (6.27)	28.72 (6.58)	0.061
Triglycerides (mg/dl)	126.08 (73.18)	132.67 (140.74)	0.517
Total cholesterol (mg/dl)	195.03 (37.94)	198.73 (36.45)	0.274
HDL cholesterol (mg/dl)	48.19 (11.43)	48.95 (11.03)	0.458
LDL cholesterol (mg/dl)	122.01 (32.88)	123.11 (22.97)	0.720
HDL:LDL	4.24 (1.17)	4.26 (1.41)	0.806
Glucose (mg/dl)	99.01 (18.85)	99.87 (19.59)	0.624
Systolic BP (mmHg)	125.27 (10.10)	125.75 (11.94)	0.631
Diastolic BP(mmHg)	78.37 (5.60)	79.76 (7.57)	0.022

Values are means (SD) except for qualitative variables, expressed as n (%).
BP: Blood Pressure; BMI: body mass index, LDL: low-density lipoprotein; HDL: high-density lipoprotein
[*]*P* value for comparisons between groups by chi square test for categorical variables and one-way ANOVA for continuous variables.

Supplementary Material 2.1

Hershey MS, Sotos-Prieto M, Ruiz-Canela M, Martínez-González MA, Cassidy A, Moffatt S, & Kales SN. Anthocyanin Intake and Physical Activity: Associations with the Lipid Profile of a US Working Population. *Molecules*. 2020;25(19):4398. doi:10.3390/molecules25194398

Table S1: Independent associations of anthocyanin intake (SD) and physical activity level with lipid profile measures (mg/dL).

Lipid profile:	Anthocyanin intake (SD) β (95%CI)	p-value	Physical activity level* β (95%CI)	p-value
Triglycerides				
Age, sex, and energy adjusted model (95% CI)	-9.13 (-17.76 to -0.50)	0.04	-8.14 (-12.48 to -3.79)	<0.001
Multivariable adjusted model 1 (95% CI) [a]	-7.69 (-16.16 to 0.79)	0.08	-5.75 (-10.23 to -1.28)	0.01
Multivariable adjusted model 2 (95% CI) [b]	-9.06 (-18.10 to -0.03)	0.05	-5.70 (-10.59 to -0.81)	0.02
Multivariable adjusted model 3 (95% CI) [c/d]	-7.73 (-16.83 to 1.37)	0.10	-5.54 (-10.40 to -0.67)	0.03
Total cholesterol				
Age, sex, and energy adjusted model (95% CI)	-2.19 (-6.80 to 2.42)	0.35	-2.69 (-5.03 to -0.34)	0.03
Multivariable adjusted model 1 (95% CI) [a]	-2.91 (-7.61 to 1.80)	0.23	-3.15 (-5.63 to -0.67)	0.01
Multivariable adjusted model 2 (95% CI) [b]	-2.16 (-7.13 to 2.80)	0.39	-2.06 (-4.75 to 0.62)	0.13
Multivariable adjusted model 3 (95% CI) [c/d]	-1.36 (-6.38 to 3.67)	0.60	-2.06 (-4.75 to 0.64)	0.13
HDL cholesterol				
Age, sex, and energy adjusted model (95% CI)	1.43 (0.09 to 2.76)	0.04	0.99 (0.31 to 1.67)	0.01
Multivariable adjusted model 1 (95% CI) [a]	1.23 (-0.05 to 2.52)	0.06	0.59 (-0.09 to 1.28)	0.09
Multivariable adjusted model 2 (95% CI) [b]	1.15 (-0.22 to 2.52)	0.10	0.59 (-0.15 to 1.34)	0.12
Multivariable adjusted model 3 (95% CI) [c/d]	1.23 (-0.16 to 2.62)	0.08	0.56 (-0.18 to 1.30)	0.14
LDL cholesterol				
Age, sex, and energy adjusted model (95% CI)	-1.75 (-5.83 to 2.33)	0.40	-2.34 (-4.42 to -0.27)	0.03
Multivariable adjusted model 1 (95% CI) [a]	-2.47 (-6.65 to 1.70)	0.24	-2.94 (-5.13 to -0.75)	0.01
Multivariable adjusted model 2 (95% CI) [b]	-1.56 (-5.97 to 2.85)	0.49	-1.99 (-4.38 to 0.39)	0.10
Multivariable adjusted model 3 (95% CI) [c/d]	-1.05 (-5.53 to 3.43)	0.65	-1.99 (-4.37 to 0.40)	0.10
LDL:HDL				
Age, sex, and energy adjusted model (95% CI)	-0.11 (-0.21 to -0.01)	0.04	-0.11 (-0.16 to -0.06)	<0.001
Multivariable adjusted model 1 (95% CI) [a]	-0.12 (-0.22 to -0.01)	0.03	-0.10 (-0.16 to -0.05)	<0.001
Multivariable adjusted model 2 (95% CI) [b]	-0.10 (-0.21 to 0.01)	0.07	-0.09 (-0.15 to -0.03)	0.004
Multivariable adjusted model 3 (95% CI) [b/c]	-0.09 (-0.20 to 0.02)	0.11	-0.09 (-0.14 to -0.03)	0.005
TG:HDL				
Age, sex, and energy adjusted model (95% CI)	-0.29 (-0.55 to -0.02)	0.04	-0.25 (-0.39 to -0.12)	<0.001
Multivariable adjusted model 1 (95% CI) [a]	-0.24 (-0.50 to 0.02)	0.08	-0.18 (-0.32 to -0.04)	0.01
Multivariable adjusted model 2 (95% CI) [b]	-0.29 (-0.57 to -0.01)	0.05	-0.19 (-0.34 to -0.04)	0.02
Multivariable adjusted model 3 (95% CI) [c/d]	-0.25 (-0.53 to 0.03)	0.08	-0.18 (-0.33 to -0.03)	0.02
Total cholesterol:HDL				
Age, sex, and energy adjusted model (95% CI)	-0.17 (-0.30 to -0.04)	0.01	-0.15 (-0.21 to -0.08)	<0.001
Multivariable adjusted model 1 (95% CI) [a]	-0.17 (-0.30 to -0.04)	0.01	-0.12 (-0.19 to -0.06)	<0.001
Multivariable adjusted model 2 (95% CI) [b]	-0.16 (-0.29 to -0.02)	0.02	-0.11 (-0.18 to -0.03)	0.004
Multivariable adjusted model 3 (95% CI) [c/d]	-0.14 (-0.27 to -0.01)	0.04	-0.10 (-0.18 to -0.03)	0.005

*Physical activity was assessed using a scale of 0-7 representing levels of physical activity ranging from none to running >10 miles/wk or spending >3 hrs/wk in comparable physical activity.

[a]Adjusted for age, sex, BMI, smoking status, education level, marital status, prevalent hypertension, dyslipidemia, and type 2 diabetes.

[b]Adjusted for age, sex, BMI, total energy intake, mMDS, smoking status, education level, marital status, multivitamin use, supplement use, sleep, prevalent hypertension, dyslipidemia, and type 2 diabetes.

[c]Adjusted for age, sex, BMI, total energy intake, mMDS, smoking status, education level, marital status, multivitamin use, supplement use, sleep, activity level, sitting, sedentary behavior, prevalent hypertension, dyslipidemia, and type 2 diabetes.

[d]Adjusted for age, sex, BMI, total energy intake, mMDS, smoking status, education level, marital status, multivitamin use, supplement use, sleep, anthocyanin intake, prevalent hypertension, dyslipidemia, and type 2 diabetes.

TG: triglycerides, HDL: high density lipoprotein cholesterol, LDL: low density lipoprotein cholesterol

Boldface indicates statistical significance (p<0.05)

Table S2. Self-Reported Physical Activity (SRPA) questionnaire.

Physical Activity in the Past Month. Below circle ONE of the values (0 to 7) which best represents your general ACTIVITY LEVEL for the PREVIOUS MONTH.

0-1: I did not participate regularly in programmed recreation, sport, or heavy physical activity.	
0 -	Avoid walking or exertion (as an example, always use elevator, drive whenever possible instead of walking, biking or rollerblading).
1 -	Walk for pleasure, routinely use stairs, occasionally exercise sufficiently to cause heavy breathing or perspiration.
2-3: I participated regularly in recreation or work requiring modest physical activity, such as golf, horseback riding, calisthenics, gymnastics, table tennis, bowling, weightlifting, yard work.	
2 -	10 to 60 minutes per week.
3 -	Over one hour per week.
4-7: I participated regularly in heavy physical exercise such as running or jogging, swimming, cycling, rowing, skipping rope, running in place or engaging in vigorous aerobic activity type exercise such as tennis, basketball or handball.	
4 -	Run less than 1 mile per week or spend less than 30 minutes per week in comparable physical activity.
5 -	Run 1 to 5 miles per week or spend 30 to 60 minutes per week in comparable physical activity.
6 -	Run 5 to 10 miles per week or spend 1 to 3 hours per week in comparable physical activity.
7 -	Run over 10 miles per week or spend over 3 hours per week in comparable physical activity.

Table S3. Description of the mMDS score developed using Feeding America's Bravest diet and lifestyle questionnaire.
The possible responses indicated in the top row are matched with their corresponding points in the row directly below.
Calculation of the modified Mediterranean diet score (mMDS):
mMDS1+(mMDS2+mMDS3)+mMDS4+mMDS5+mMDS6+mMDS9+mMDS12+mMDS15+mMDS16+(mMDS7+mMDS10+mMDS13)*(1-fh) +(mMDS8+mMDS11+mMDS14)*fh
[1] Weighted by the proportion of meals at home relative to the total number of meals per week (breakfast + lunch + dinner)
[2] Weighted by the proportion of meals at the firehouse (or on work time) relative to the total number of meals per week (breakfast + lunch + dinner)

mMDS items	Components	Score range	Points for each answer								
1. Fast-food or Take-out food	How many times per week do you eat the following?	0-4	never	≤ 1	2-3	3-4	5-6	every day	8-10	≥ 11	missing
			4 pts	3 pts	2 pts	2 pts	1 pts	1 pts	0 pts	0 pts	0 pts
2. Fruits	How many servings of each of the following do you consume per day?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7			missing
			0 pts	1 pts	2 pts	3 pts	4 pts	4 pts			0 pts
3. Vegetables (not including potatoes)	How many servings of each of the following do you consume per day?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7			missing
			0 pts	1 pts	2 pts	3 pts	4 pts	4 pts			0 pts
4. Legumes (e.g. beans, chickpeas, lentils)	How many servings of each of the following foods do you eat per week?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7			missing
			0 pts	0 pts	1 pts	2 pts	3 pts	4 pts			0 pts
5. Nuts (e.g. walnuts, almonds, hazelnuts, pistachio, peanuts)	How many servings of each of the following foods do you eat per week?	0-4	0	≤ 1	2-3	3-4	5-6	≥ 7			missing
			0 pts	0 pts	1 pts	2 pts	3 pts	4 pts			0 pts
6. Sweet Desserts (cake, cookies, pie, ice cream, etc.)	How many times per week do you eat the following?	0-4	never	≤ 1	2-3	3-4	5-6	≥ 7			missing
			4 pts	4 pts	3 pts	2 pts	1 pts	0 pts			0 pts
7. Primary cooking oil/fat use at home[1]	Which oil or fat do you use most often for cooking and serving food at home?	0-5	Butter	Lard or other animal fat	Margarine	Corn or vegetable oil	Benechol or Smart Balance	Olive oil	EVOO	other	missing
			0	0 pts	1 pts	2 pts	3 pts	4 pts	5 pts	0 pts	0 pts
8. Primary cooking oil/fat use at work[2]	Which oil or fat do you use most often for cooking and serving food at the firehouse?	0-5	Butter	Lard or other animal fat	Margarine	Corn or vegetable oil	Benechol or Smart Balance	Olive oil	EVOO	other	missing
			0 pts	0 pts	1 pts	2 pts	3 pts	4 pts	5 pts	0 pts	0 pts
9. Fried foods (French fries, fried chicken, chicken nuggets, etc.)	How many times per week do you eat the following?	0-4	never	≤ 1	2-3	3-4	5-6	every day	8-10	≥ 11	missing
			4 pts	3 pts	2 pts	1 pts	0 pts	0 pts	0 pts	0 pts	0

10. Breads/starches consumed at home[1]	Which bread or starch do you most frequently eat at home?	0-4	I do not eat bread or starch 3 pts	White bread, filled pasta, white rice, or potatoes 0 pts	Durum wheat bread or dry pasta 3 pts	French bread or Italian bread or multigrain or other crusty bread 2 pts	Whole wheat bread or brown rice or whole wheat pasta 4 pts	missing 0 pts								
11. Breads/starches consumed at work[2]	Which bread or starch do you most frequently eat at the firehouse?	0-4	I do not eat bread or starch 3 pts	White bread, filled pasta, white rice, or potatoes 0 pts	Durum wheat bread or dry pasta 3 pts	French bread or Italian bread or multigrain or other crusty bread 2 pts	Whole wheat bread or brown rice or whole wheat pasta 4 pts	missing 0 pts								
12. Baked, broiled, grilled, or blackened (NOT fried) ocean fish (salmon, tuna, cod, haddock, etc.)	How many times per week do you eat the following?	0-4	never 0 pts	≤ 1 1 pts	2-3 2 pts	3-4 3 pts	5-6 4 pts	every day 4 pts	8-10 4 pts	≥ 11 4 pts	missing 0 pts					
13. Non-alcoholic beverages at home[1]	Which of the following non-alcoholic beverages do you most frequently drink at home?	0-4	Cola/ soda 0 pts	Diet cola/soda 1 pts	Fruit drink or punch 1 pts	Milk 1 pts	Tea/ coffee 2 pts	Juice 2 pts	Water 4 pts	Other 0 pts	missing 0 pts					
14. Non-alcoholic beverages at work[2]	Which of the following non-alcoholic beverages do you most frequently drink at the firehouse?	0-4	Cola/ soda 0 pts	Diet cola/soda 1 pts	Fruit drink or punch 1 pts	Milk 1 pts	Tea/ coffee 2 pts	Juice 2 pts	Water 4 pts	Other 0 pts	missing 0 pts					
15. Quantity of alcoholic beverages	How many alcoholic beverages (beer, wine, hard liquor, etc.) do you drink over a typical week?	0-4	I do not drink 0 pts	0 0 pts	1-2 2 pts	3-4 2 pts	5-6 4 pts	7-8 4 pts	9-10 4 pts	11-12 4 pts	13-14 4 pts	15-16 4 pts	17-18 4 pts	19-20 4 pts	≥ 21 1 pts	missing 0 pts
16. Wine consumption	When you drink alcoholic beverages, what type do you drink?	0-2	White wine 2 pts	Red wine 2 pts	Beer 0 pts	Hard liquors 0 pts	Don't drink 0 pts									

Table S4: Sensitivity analysis for anthocyanin intake (SD) and physical activity level on lipid profile measures (mg/dL).

Sensitivity Analysis		Anthocyanin intake (SD)		Physical activity level*	
Exclusions	**n**	**β (95%CI)**	**p-value**	**β (95%CI)**	**p-value**
Chronic diseases[†a]	199				
Triglycerides		-9.09 (-19.24 to 1.06)	0.08	-5.89 (-11.64 to -0.14)	0.05
Total cholesterol		-2.29 (-7.67 to 3.12)	0.41	-0.55 (-3.62 to 2.52)	0.72
HDL cholesterol		0.92 (-0.59 to 2.44)	0.23	0.85 (-0.01 to 1.70)	0.05
LDL cholesterol		-1.44 (-6.14 to 3.27)	0.55	-0.83 (-3.50 to 1.84)	0.54
LDL:HDL		-0.09 (-0.21 to 0.03)	0.16	-0.08 (-0.15 to -0.01)	0.03
TG:HDL		-0.27 (-0.58 to 0.04)	0.09	-0.21 (-0.39 to -0.04)	0.02
Total cholesterol:HDL		-0.14 (-0.29 to 0.01)	0.07	-0.10 (-0.18 to -0.01)	0.03
Women[a]	236				
Triglycerides		-9.32 (-18.81 to 0.16)	0.05	-5.69 (-10.76 to -0.63)	0.03
Total cholesterol		-2.10 (-7.25 to 3.04)	0.42	-1.97 (4.72 to 0.77)	0.16
HDL cholesterol		0.72 (-0.67 to 2.12)	0.31	0.64 (-0.11 to 1.38)	0.09
LDL cholesterol		-1.01 (-5.58 to 3.55)	0.66	-1.97 (-4.40 to 0.46)	0.11
LDL:HDL		-0.08 (-0.20 to 0.03)	0.16	-0.09 (-0.15 to -0.03)	0.004
TG:HDL		-0.29 (-0.58 to 0.01)	0.06	-0.19 (-0.35 to -0.03)	0.02
Total cholesterol:HDL		-0.14 (-0.28 to 0.00)	0.05	-0.11 (-0.18 to -0.03)	0.004
Supplement use (proteins, glutamine, amino acids, etc.)[a]	170				
Triglycerides		-8.72 (-19.39 to 1.96)	0.11	-4.79 (-10.38 to 0.80)	0.09
Total cholesterol		-4.22 (-10.53 to 2.08)	0.19	-3.56 (-6.84 to -0.29)	0.03
HDL cholesterol		0.38 (-1.36 to 2.11)	0.69	0.23 (-0.68 to 1.14)	0.61
LDL cholesterol		-2.77 (-8.44 to 2.91)	0.34	-2.83 (-5.78 to 0.12)	0.06
LDL:HDL		-0.09 (-0.22 to 0.05)	0.21	-0.07 (-0.14 to -0.00)	0.04
TG:HDL		-0.21 (-0.51 to 0.10)	0.18	-0.10 (-0.26 to 0.06)	0.21
Total cholesterol:HDL		-0.13 (-0.29 to 0.03)	0.12	-0.09 (-0.18 to -0.01)	0.03

*Physical activity was assessed using a scale of 0-7 representing levels of physical activity ranging from none to running >10 miles/wk or spending >3 hrs/wk in comparable physical activity.

†Chronic diseases was defined as reporting a previous diagnosis or current treatment for hypertension, dyslipidemia, or diabetes, respectively.

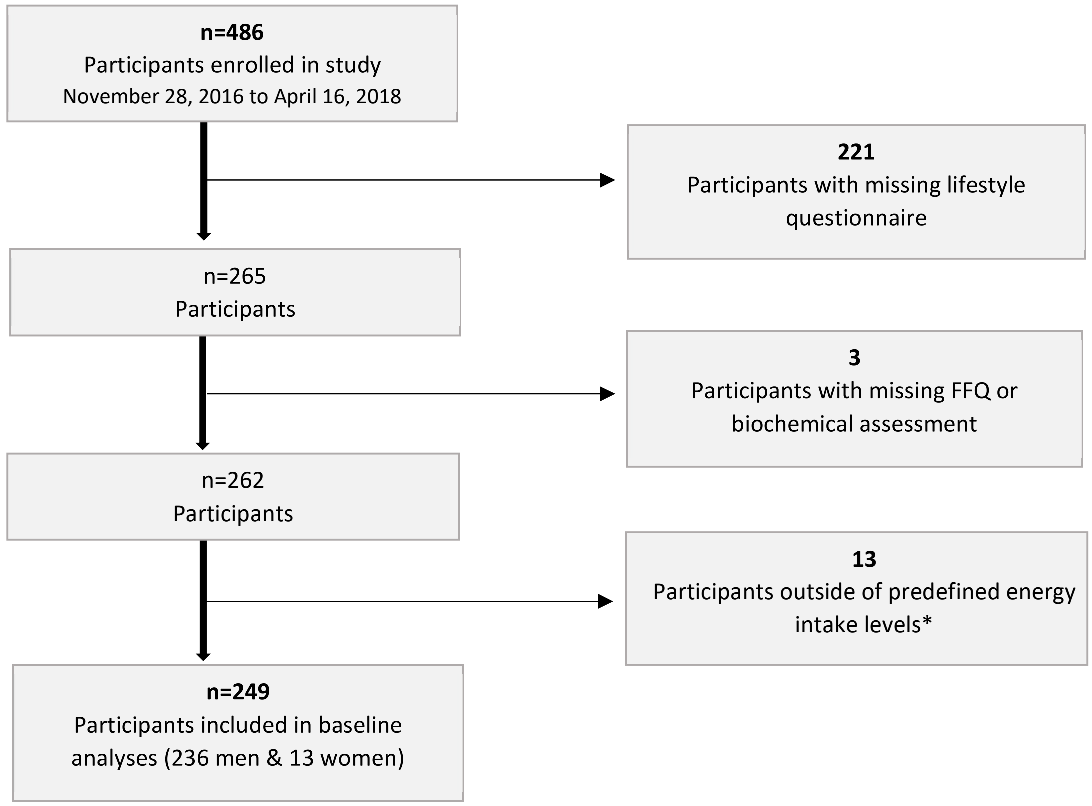
[a]Adjusted for age, sex, BMI, total energy intake, mMDS, smoking status, education level, marital status, multivitamin use, supplement use, sleep, anthocyanin intake, prevalent hypertension, dyslipidemia, and type 2 diabetes

TG: triglycerides, HDL: high density lipoprotein cholesterol, LDL: low density lipoprotein cholesterol

Boldface indicates statistical significance (p<0.05)

Supplementary Material 2.2

Hershey MS, Sotos-Prieto M, Ruiz-Canela M, Christophi CA, Moffatt S, Martínez-González MA, & Kales SN. The Mediterranean lifestyle (MEDLIFE) index and metabolic syndrome in a non-Mediterranean working population. *Clinical Nutrition.* 2021;40(5):2494-2503. doi:10.1016/j.clnu.2021.03.026



Supplementary Figure 1. Flowchart of eligible participants in the *Feeding America's Bravest* trial, 2016-2019.
*800-5000kcal/d men, 500-3500kcal/d women

Supplementary Table 1. Description of the Mediterranean Lifestyle (MEDLIFE) index modified for *Feeding America's Bravest*.

Item	Components (serving size)	Criteria for 1 point
Block 1: Mediterranean food consumption		
1. Sweets	milk chocolate, dark chocolate, candy bars, candy (1 oz.), cookies, brownies, doughnuts, cake, pie, muffins, biscuits (1 unit) pancakes, waffles (2 small units)	≤ 2 servings/wk
2. Red meat	Beef, pork, lamb as main dish, mixed dish, or sandwich (4-6 oz.)	< 2 servings/wk
3. Processed meats	Hamburger, hotdog (1 unit), salami, bologna, other processed meat (2 oz.), chicken or turkey hotdogs or sandwich, sausage, frozen dinner (1 unit), bacon (2 slices), beef liver (4 oz.), chicken liver (1 oz.)	≤ 1 serving/wk
4. Eggs	Regular eggs including yolk and omega-3 fortified including yolk	2-4 servings/wk
5. Legumes	Beans or lentils, baked, dried, or soup, peas, lima beans (1/2 cup)	≥ 2 servings/wk
6. White meat	Chicken or turkey with or without skin (3 oz.)	2 servings/wk
7. Fish	Dark meat fish (tuna steak, mackerel, salmon, sardines, bluefish, swordfish), other fish (3-5 oz.), canned tuna (3-4 oz.), shrimp, lobster, scallops as a main dish	≥ 2 servings/wk
8. Potatoes	potatoes, baked, boil (1 unit) or mashed (1 cup) and french fries (6 oz. or 1 serving)	≤ 3 servings/wk
9. Dairy products	Skim, 1 or 2%, whole, soy milk (8 oz.), cream (1 Tbs), frozen yogurt, sherbet, ice cream, plain or sweetened yogurt (1 cup), cottage or ricotta cheese (1/2 cup), margarine, butter, cream cheese, other cheese (1 oz./1 slice)	2 servings/d
10. Nuts	Nuts (e.g. walnuts, almonds, hazelnuts, pistachio, peanuts)	1-2 serving/d
11. Fruit	Raisins (1 oz), grapes (1/2 cup), prunes (6 units), apple, orange, grapefruit, prune, and other fruit juices (small glass), bananas (1 unit), cantaloupe (1/4 melon), grapefruit, avocado (1/2 fruit or cup), apples, pears, oranges, peaches, plums, apricots (1 unit), strawberries, blueberries fresh, frozen, or canned (1/2 cup)	3-6 servings/d
12. Vegetables	Tomatoes (2 slices), tomato or carrot juice (small glass), broccoli, string beans, cauliflower, cabbage, Brussel sprouts, raw or cooked carrots, corn, mixed vegetables, yams, sweet potatoes, squash, eggplants, zucchini, kale, cooked spinach, cooked onions (1/2 cup), spinach (1 cup), head or leaf lettuce (1 serving), celery (2-3 sticks), peppers (3 slices), raw onion (1 slice)	≥ 2 servings/d
13. Olive oil	Olive oil added to food or bread (1 Tbs.) Main oil usually used for frying and sautéing at home?	≥ 1 servings/d + Olive oil
14. Cereals	Cold breakfast cereal (1 serving), oatmeal (1 cup), other cooked cereals (1 cup) white bread (1 slice), rye bread (1 slice), whole grain bread (1 slice), English muffin, bagel, rolls (1 unit), brown rice (1 cup), white rice (1 cup), pasta, noodles, couscous (1 cup), tortillas (2 units), pizza (2 slices)	3-6 servings/d
Block 2: Mediterranean dietary habits		
15. Water, coffee, and tea	Which of the following non-alcoholic beverages do you most frequently drink at home? Which of the following non-alcoholic beverages do you most frequently drink at the firehouse? Water (bottled, sparkling, or tap), herbal tea, tea with caffeine, decaffeinated tea, coffee with caffeine, decaffeinated coffee (8 oz.)	Water, coffee, and tea are most frequent non-alcoholic drinks at home and firehouse + ≥ 6 cups/d of water, coffee, or tea
16. Wine	When you drink alcoholic beverages, what type do you drink?	Red or white wine are usual alcoholic beverage
17. Limit salt	Sodium (mg)	< 2.3 g/d
18. Preference for whole grain products	Is whole grain the main type of bread or starch that you eat?	Yes
19. Snacks	Regular popcorn (3 cups), fat free popcorn (3 cups), potato chips (small bag or 1 oz), crackers (6), pretzels (1 small bag or serving), breakfast, energy, and low carb bars (1 unit)	≤ 2 serving/wk
20. Limit sugar in beverages (sugary beverages)	Carbonated beverages caffeinated or caffeine-free with sugar and other sugared beverages: punch, iced tea, lemonade, sports drinks (1 glass, bottle, or can)	≤ 1 serving/wk
21. Local, seasonal, or organic products	Do you usually consume local/seasonal or organic products?	Yes
Block 3: Physical activity, rest, social habits, and conviviality		
22. Physical activity	Statement that best describes your physical activity in the past month	Run >10 miles/wk or spend >3 hrs/wk in comparable physical activity
23. Siesta/nap	How many times do you take a nap per week?	≥ 3 naps/wk
24. Hours of sleep	Total hours of actual sleep in a typical 24-hour period	6-8 hrs/d
25. Watching TV	During the past weeks, what was your average total time per week at each of the following activities?	≤ 4 hrs/wk
26. Time spent eating	Time you usually spend eating each meal at home (in minutes). Time you usually spend eating each meal at the firehouse (in minutes).	≥ 20 min

Abbreviations: min: minutes, h: hours, d: day, wk: week, oz.: ounces, g: grams, Tbsp: tablespoons

Supplementary Table 2. Adjusted MEDLIFE characteristics according to MEDLIFE tertiles at baseline in *Feeding America's Bravest*.

Characteristic	MEDLIFE adherence T1 (2-7 pts.)	T2 (8-10 pts.)	T3 (11-17 pts.)	p-value
N	90	99	60	
Sweets (serv/wk)	11.17 (12.51)	7.58 (7.29)	5.07 (5.07)	**<0.001**
Red meat (serv/wk)	4.08 (2.51)	3.56 (2.52)	2.54 (2.12)	**<0.001**
Processed meat (serv/wk)	11.07 (7.91)	9.71 (8.86)	7.36 (6.06)	**0.006**
Eggs (serv/wk)	4.29 (4.64)	3.59 (3.45)	5.51 (7.76)	0.15
Legumes (serv/wk)	1.23 (1.29)	1.71 (1.65)	2.19 (1.94)	**0.003**
White meat (serv/wk)	3.18 (2.83)	3.06 (2.60)	3.07 (2.19)	0.95
Fish (serv/wk)	1.19 (1.06)	2.13 (1.92)	2.66 (1.80)	**<0.001**
Potatoes (serv/wk)	3.69 (2.22)	2.70 (2.03)	1.69 (1.62)	**<0.001**
Dairy products (serv/d)	3.60 (2.09)	2.71 (1.92)	2.39 (1.52)	**<0.001**
Nuts (serv/d)	0.34 (0.48)	0.51 (0.89)	0.64 (0.57)	**0.004**
Fruits (serv/d)	1.46 (1.05)	1.97 (1.37)	2.47 (1.45)	**<0.001**
Vegetables (serv/d)	2.36 (1.62)	3.10 (2.00)	3.94 (2.02)	**<0.001**
Olive oil (serv/d)	0.45 (0.68)	0.34 (0.34)	0.58 (0.69)	**0.03**
Olive oil is main cooking oil (%)	49.04	63.46	85.47	**<0.001**
Cereals (serv/d)	2.11 (1.29)	1.98 (1.26)	1.57 (1.00)	**0.01**
Water, coffee, and tea are most frequent drinks at home (%)	57.98	66.89	89.47	**<0.001**
Water, coffee, and tea are most frequent drinks at firehouse (%)	71.54	77.43	94.45	**0.004**
Water, coffee, and tea (serv/d)	4.09 (2.64)	4.63 (3.12)	7.05 (3.43)	**<0.001**
Wine drinkers (%)	5.50	9.98	16.37	0.10
Sodium <2.3g/d (%)	26.30	48.96	67.99	**<0.001**
Whole grain is main type of bread/starch consumed (%)	42.83	74.51	83.87	**<0.001**
Snacks (serv/wk)	8.09 (7.08)	7.16 (6.47)	5.35 (5.34)	**0.03**
Sugary beverages (serv/wk)	5.10 (6.65)	2.38 (3.44)	1.04 (2.79)	**<0.001**
Usually consume local, seasonal, or organic products (%)	16.52	35.62	75.56	**<0.001**
Run >10 miles/wk or spend >3 hrs/wk in comparable PA (%)	2.14	13.88	24.32	**<0.001**
Siesta/nap ≥ 3/wk (%)	13.10	27.75	28.95	**0.03**
Sleep 6-8hrs/d (%)	76.38	88.70	90.92	**0.03**
Watching TV ≤ 4hrs/wk (%)	52.97	65.28	77.00	**0.01**
Time spent eating at home (min/meal)	19.24 (10.77)	18.05 (8.29)	21.90 (10.22)	0.06
Time spent eating at firehouse (min/meal)	19.89 (12.59)	21.19 (8.73)	20.63 (9.33)	0.73

Characteristics are age and sex adjusted using the inverse probability weighting method. Continuous variables are expressed as mean and standard deviation and categorical variables as a percentage
d: day, g: grams, hrs: hours, min: minute, PA: physical activity, serv: serving, TV: television, wk: week

Supplementary Table 3. Frequency of adherence to individual items of the MEDLIFE index in *Feeding America's Bravest*, 2016-2019.

MEDLIFE items	Criteria for 1 point	% with 1 point
	Block 1: Mediterranean food consumption	
1. Sweets	≤ 2 servings/wk	18.5
2. Red meat	< 2 servings/wk	25.7
3. Processed meat	≤ 1 serving/wk	1.6
4. Eggs	2-4 servings/wk	37.8
5. Legumes	≥ 2 servings/wk	30.1
6. White meat	2 servings/wk	24.5
7. Fish/seafood	≥ 2 servings/wk	45.4
8. Potatoes	≤ 3 servings/wk	61.5
9. Dairy products	2 servings/d	32.5
10. Nuts	1-2 servings/d	12.1
11. Fruit	3-6 servings/d	18.9
12. Vegetables	≥ 2 servings/d	65.5
13. Olive oil	≥ 1 servings/d + olive oil	12.9
14. Cereals	3-6 servings/d	14.1
	Block 2: Mediterranean dietary habits	
15. Water, coffee, and tea	Water, coffee, and tea are most frequent non-alcoholic drinks at home and at the firehouse + ≥ 6 servings/d	28.9
16. Wine	Red or white wine are usual alcoholic beverage	10.4
17. Limit salt at meals	sodium < 2.3 g/d	45.0
18. Preference for whole grain products	Is whole grain the main type of bread or starch that you eat? Yes	65.5
19. Snacks	≤ 2 serving/wk	20.1
20. Limit sugar in beverages	sugary and sweetened beverages ≤ 1 serving/wk	51.0
21. Local, seasonal, or organic products	Do you usually consume local/seasonal or organic products? Yes	38.6
	Block 3: Physical activity, rest, social habits, and conviviality	
22. Physical activity	Run over 10 miles/wk or exercise > 3 hrs/wk	12.5
23. Siesta/nap	≥ 3 naps/wk	22.9
24. Hours of sleep	6-8 h/d	85.1
25. Watching television	≤ 4 hours/wk	63.1
26. Time spent eating	Time you usually spend eating each meal at home and work ≥ 20 minutes	32.9

Supplementary Table 4. Odds ratios (OR) and 95% confidence intervals (CI) according to MEDLIFE tertiles on metabolic syndrome and its components.

		Categories of adherence to MEDLIFE			
	n	**T1 (2-7 pts.)**	**T2 (8-10 pts.)**	**T3 (11-17 pts.)**	**p for trend**
Metabolic Syndrome:	44				
Crude model (95% CI)		1 Ref.	0.68 (0.33 to 1.39)	**0.37 (0.14 to 0.97)**	**0.04**
Multivariable adjusted model (95% CI)[a*]		1 Ref.	0.75 (0.35 to 1.61)	0.36 (0.12 to 1.04)	0.06
Multivariable adjusted model (95% CI)[b*]		1 Ref.	0.70 (0.32 to 1.54)	**0.29 (0.10 to 0.90)**	**0.04**
- **Abdominal obesity:**	95				
Crude model (95% CI)		1 Ref.	0.85 (0.48 to 1.51)	**0.42 (0.20 to 0.85)**	**0.02**
Multivariable adjusted model (95% CI)[a*]		1 Ref.	1.00 (0.53 to 1.90)	**0.44 (0.20 to 0.97)**	0.06
Multivariable adjusted model (95% CI)[b*]		1 Ref.	0.94 (0.48 to 1.82)	0.42 (0.18 to 0.99)	0.07
- **Hyperglycemia**	92				
Crude model (95% CI)		1 Ref.	0.85 (0.48 to 1.53)	0.61 (0.31 to 1.23)	0.18
Multivariable adjusted model (95% CI)[a]		1 Ref.	0.82 (0.43 to 1.57)	0.61 (0.28 to 1.31)	0.21
Multivariable adjusted model (95% CI)[b*]		1 Ref.	0.72 (0.37 to 1.41)	0.48 (0.21 to 1.09)	0.08
- **Hypertension:**	18				
Crude model (95% CI)		1 Ref.	0.43 (0.14 to 1.30)	0.42 (0.11 to 1.60)	0.13
Multivariable adjusted model (95% CI)[a]		1 Ref.	0.47 (0.15 to 1.48)	0.47 (0.11 to 1.93)	0.21
Multivariable adjusted model (95% CI)[b*]		1 Ref.	0.43 (0.13 to 1.46)	0.37 (0.08 to 1.69)	0.15
- **Hypertriglyceridemia:**	66				
Crude model (95% CI)		1 Ref.	**0.43 (0.23 to 0.81)**	**0.23 (0.10 to 0.54)**	**<0.001**
Multivariable adjusted model (95% CI)[a]		1 Ref.	**0.43 (0.22 to 0.86)**	**0.24 (0.10 to 0.62)**	**0.001**
Multivariable adjusted model (95% CI)[b*]		1 Ref.	**0.42 (0.21 to 0.86)**	**0.24 (0.09 to 0.63)**	**0.002**
- **Low HDL cholesterol:**	61				
Crude model (95% CI)		1 Ref.	0.93 (0.49 to 1.76)	0.52 (0.23 to 1.18)	0.14
Multivariable adjusted model (95% CI)[a]		1 Ref.	0.90 (0.45 to 1.79)	0.55 (0.23 to 1.32)	0.20
Multivariable adjusted model (95% CI)[b*]		1 Ref.	0.88 (0.43 to 1.81)	0.54 (0.22 to 1.36)	0.21

[a]Adjusted for age, sex, BMI, total daily energy intake, smoking status, and education level
[b]Adjusted for age, sex, BMI, total daily energy intake, alcohol intake (not including wine), smoking status, education level, civil status, multivitamin use, supplement use, sleep medication, prevalent hypertension, dyslipidemia, and T2DM
*adjusted for all covariables in the model with the exclusion of BMI, T2DM, hypertension, or dyslipidemia, respectively
Boldface indicates statistical significance (p<0.05)