aesthetic medicine

Review

Mediterranean Diet as a tool for achieving successful ageing

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Abstract

Targeted interventions to slow or postpone ageing and to favour an active life expectancy represent the new perspectives in ageing investigation. Some mechanisms that delay or prevent the onset of ageing disabilities and pathologies have been identified. In general, maintaining a healthy lifestyle seems to reduce many risk factors. In particular, eating habits represent the most concrete and low-cost method to address ageing. Of all dietary habits analysed, the Mediterranean diet has received much attention since it has consistently proven its beneficial influence on health and longevity. The Mediterranean diet is characterized by low glycaemic index and low animal protein intake, it is rich in nutraceuticals and functional foods thus reducing molecular pathways signalling, as nutrient sensitive pathways which affect the ageing process and lead to unhealthy ageing. This dietary model can be promoted as an "anti-ageing therapy" and can contribute to good health status and therefore to a better quality of life.

Keywords

Ageing, functional food, hormesis, longevity, mediterranean diet, nutraceuticals

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Introduction: healthy foods for healthy ageing

Healthy ageing as well as the occurrence of age-related disabilities and diseases result from several degrees of interaction between genetic, epigenetic and environmental factors[1]. To reach an active and independent end of life and to achieve a healthy life-span, the so called *health-span*, an integrative approach is needed[2]. With the aim of developing preventive and therapeutic measures, it is important to start managing daily habits. Nutrition is probably the most important one[3,4]. Our life, in different ways, turns around food. It can be a positive or negative relationship. It can be a reason for travelling or for working but no one can live without it.

Since ageing is an ineluctable process, the goal is to live longer but in good health rather than just delaying the end of life.

The Mediterranean Diet (MedDiet) is a cultural tradition that contributes to better health and quality of life in Mediterranean countries, where many longevous people exist. In fact, the Elderly Prospective Cohort Study identified a reduced overall mortality among old people who live in the *Mediterranean way*[5,6].

For many years, interventions to slow the rate of ageing and increase life-span have been tested by many scientists. They have tried and succeeded in modulating the nutrient-sensing pathways (NSPs) and in revealing new and interesting research suggestions in the field of anti-ageing medicine[2]. In both animal models and humans dietary intervention can decrease the chronic, generalized pro-inflammatory status, the inflammageing, positively modulating the ageing process[7-9]. The complex relationship between nutrition, the ageing process and healthy ageing is not completely understood. Specific dietary changes however, as recommended by NU-AGE project, such as the reduction or elimination of trans and saturated fats and an increased intake of omega-3 fatty acids, vitamins, minerals and antioxidants can help to minimize inflammation[10].

Similarly, appropriate intake of functional foods may confer health benefits, influencing the maintenance of immune homeostasis and contributing, directly, to the reduction of inflammation and metabolic disorders related to an inadequate diet[11,12].

In this review we evaluate the relationship between adherence to the MedDiet, rich in bioactive compounds, and healthy ageing, underlying its activating or inhibitory role in cellular pathways and the anti-ageing effects of its functional foods.

Ageing and longevity

Ageing is a complex, unavoidable phenomenon or trait than cannot be exhaustively defined.

In May 2012, a group of scientists and clinicians met in Athens to consider the relevance of ageing and longevity. The workshop led to the creation of a statement to highlight the importance of a common view related to these processes, since they represent phenotypes that are rapidly spreading worldwide.

The panel defined ageing as a process which makes people more vulnerable, leading to death but in a distinct moment of the chronological age (CA) as compared to the biological age (BA)[13].

The United Nations define "elderly" all individuals from the age of 60 years old[14]. Of course it can be considered only a necessary threshold in order to classify the population.

We are however interested in how long people live and remain engaged and physically active so the simplistic classification, based on the CA, is only theoretical and not very useful. CA refers to the date of birth, while BA is a personal feature that depends on how old a person appears to be. These two parameters often do not match. The BA is a dynamic concept, based on the biological health status of tissues, organs and systems[15].

By definition there are two ways to become old, i.e. with success (successful ageing) and without success (unsuccessful or pathological ageing). The ratio between CA and BA is greater than or equal to 1 in successful ageing and less than 1 in unsuccessful ageing.

Unsuccessfull ageing is manifested by people who develop one or more age-related diseases, such as neurodegenerative (Alzheimer's or Parkinson's) diseases, metabolic (metabolic syndrome and type 2 diabetes mellitus) and cardiovascular diseases (myocardial infarction and atherosclerosis) and cancer[13,16]. In fact, ageing is due to the loss of molecular fidelity that varies among different subjects, leading to both dangerous and non-dangerous features that can reduce the adaptation to the environment, involving DNA, cells, tissues and the whole system[17]. It results in compromised stress response, greater homeostatic imbalance, elevated risk of disease and, consequently, death[15,17].

Instead, successful ageing implies the avoidance (or the late onset) of diseases and disabilities with a preservation of cognitive and physical functions[13].

Centenarians represent the best model of successful ageing. They live longer than the rest of the population and without any relevant disease, thanks to a favourable genetic background and to a good response to adverse environmental conditions[13]. In 2012, Caruso et al. conducted a study in a small area of Sicily, the Sicani Mountains, where the number of centenarians is high.

These areas showed low mortality rate for cancer and cardiovascular disease. The centenarians recruited tended to be physically active and to have a healthy diet, contributing to inflammation reduction[18,19].

On the other hand, physical inactivity and hypercaloric diet lead to an accumulation of visceral fat and infiltration of pro-inflammatory macrophages and T-cells in adipose tissue.

Moreover, adipose tissue releases adipokines and other pro-inflammatory cytokines, causing inflammation[20].

The centenarians' ability to reach the age of 100 in the Westernized countries and the reduction in overall mortality reflect the improvement of hygienic condition, the reduced exposure to infection and inflammation, the overall improvement of the quality of life, the attention to the diet and the advent of therapeutic and preventive medicine[15,21].

Mediterranean Diet

MedDiet is one of the most studied healthy dietary patterns. It represents more than an alimentary regimen. It can be considered a life-style that characterizes people, groups, regions and countries with similar but, at the same time, different ways to eat. As many studies demonstrated, MedDiet can positively influence several parameters, such as abdominal obesity, dyslipidaemia, elevated blood pressure and impaired glucose tolerance all being factors that predispose to onset of age-related diseases. Large intervention trials showed that MedDiet could prevent or delay age-related diseases with a great implication for health and social system[22-26]. Since 1995, this dietary pattern is represented by a pyramid updated over the years. The current one suggests a daily consumption of plant foods (fruit, vegetables, nuts, seeds, olives, herbs and spices), whole cereals, extra virgin olive oil (EVOO) as main source of fat, dairy products (principally cheese and yogurt, preferably low fat) and weekly consumption of poultry, fish, eggs (two to four) and legumes. Red and cured meats and sweets can be consumed in very low amount and red wine can be consumed responsibly during meals[27]. So, this diet consists with a low animal protein intake. It is hypocaloric, characterized by a low amount of saturated fats, cholesterol and sugars, and a high content of fibres[25]. Its beneficial effects could also be attributable to the presence of nutraceuticals and functional foods, other than only to the low glycaemic index (GI) and to the low animal protein and caloric intake[25]. Nutraceuticals are *Naturally derived bioactive compounds that are found in foods, dietary supplements and herbal products; they have health-promoting, disease-preventing or medicinal properties*[28,29]. They have antioxidant and anti-inflammatory effects that confer to MedDiet anti-ageing properties. In particular, fruit and vegetables are important sources of nutraceuticals such as polyphenols, as well as fish and red wine, rich in omega-3 fatty acids and resveratrol, respectively[9,30]. All of these belong to the big family of functional foods. Although a universal definition for them does not exist, the Functional Food Center defined them as *Natural or processed foods that contain known or unknown biologically-active compounds which, in defined, effective non-toxic amounts provide a clinically proven and documented health benefit for the prevention, management or treatment of chronic disease*[31].

Mediterranean diet as a strategy to modulate ageing

As previously stated, Mediterranean functional foods contain bioactive compounds with anti-inflammatory and anti-oxidant effects that can confer health benefits, contributing directly to the reduction of the inflammatory status and of metabolic disorders related to an unhealthy diet[9,25,29].

Thanks to the studies on the positive contribution of MedDiet to the extension of life-span in healthy condition, it can be considered a strategy to delay ageing. In fact, it is characterized by low GI and low animal protein intake and it is able to act directly on specific metabolic pathways, called NSPs[9,25].

The NSPs are signalling cascades activated by the level of nutrients, such as carbohydrates or protein (or aminoacids). The most representative are the insulin/insulin-like growth factor-1 (IGF-1) and the target of rapamicin (TOR) pathways. From yeast to human, they are genetically conserved although, of course, the molecular complexity varies among them[8,32].

Insulin and IGF-1 levels are, respectively, influenced by glucose and growth hormone (GH). During fasting, both GH and insulin decrease with a consequent reduction of IGF-1 circulating levels and a delay in the ageing process, as presented by several model organisms[33].

The insulin/IGF-1 signalling cascade starts from the binding of insulin or IGF-1 to the insulin or IGF-1 receptor (IGF-1R) that triggers, inside the cell, many molecular events, such as the activation of the phosphoinositide second messenger.

It leads to the activation of AKT that can stimulate the nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) signalling, involved in immune inflammatory mechanisms[34,35]. The translocation of NF-κB to the nucleus and its binding to the DNA triggers the transcription of a number of genes, including pro-inflammatory cytokines, chemokines, adhesion molecules, eicosanoids, growth factors, metallo-proteinases, nitric oxide, etc[34].

The modulation of this pathway, possibly due to low glycaemic and protein intake and, maybe, to some antioxidant and anti-inflammatory molecules, may lower insulin/IGF-1 signal and stimulate the action of different transcription factors (TFs)[36].

Forkhead box O 3A (FOXO3A), extensively studied for its role in longevity, is one of these TFs, involved in the transcription activation of molecules that take part in cellular homeostasis. The gene encoding for this protein belongs to the FOXO family which has a typical DNA-binding forkhead box domain. It is one of the orthologue of DAF-16 in *Caenorhabditis elegans (C. elegans)*, a TF involved in stress resistance and longevity[37,38]. In addition, FOXO3A interacts with sirtuins (SIRTs), a family of histone deacetylase enzymes, identified as anti-ageing molecules in model organisms. SIRT1, one of the seven human sirtuin isoforms, SIRT1-SIRT7, deacetylates FOXO3A modulating its response to oxidative stress[39]. In mammals, downstream of IGF-1R, there is the mammalian TOR (mTOR) composed by mTOR complex 1 (mTORC1) and the mTOR complex 2. mTORC1 is activated by insulin and IGF-1, modulating growth, metabolism, and stress response through regulation of transcription, translation and autophagy, a cytoprotective process. Its inhibition by the protein

kinase AMP, a key sensor of the cellular energy state, activated by low levels of ATP, causes stress resistance and reduced age-related inflammatory status by the NF-κB pathway[40].
Decreased TOR activity extends longevity also in *C. elegans*[41,42] and *Drosophila melanogaster*[43]. In addition, its reduction in a mouse genetic model reduces the incidence of some age-related diseases, including cancer[44].
So, in humans, MedDiet, with its low intake of amino acids and low GI, might directly modulate the insulin/IGF-1 and the TOR pathways trough a down-regulation of the signals that lead to the activation of FOXO and, consequently, to the transcription of homeostatic genes that favour longevity thus decreasing inflammatory status and oxidative stress[25,45,46].

The Mediterranean hormetic effect

Many components of the MedDiet are known to have healthy properties. The abundance in mono and polyunsaturated fatty acids (MUFAs and PUFAs), fibres, vitamins, minerals and, more generally, nutraceuticals plays a key role in inflammation reduction and oxidative stress response, acting on important risk factors for age-related diseases[25,47,48].
Moreover, nutraceuticals stimulate a hormetic response[49,50]. In 1943, Chester Southam and John Ehrlich described[51], for the first time, the phenomenon of hormesis. The term comes from the Greek word *ὅρμησις* that means *rapid motion.* As it was then defined in 2002, it is a *biphasic dose response phenomenon, characterized by a low dose stimulation and a high dose inhibition*[52].
In biology and medicine, hormetic effect is an adaptive response of cells and organisms to moderate (usually intermittent in terms of intensity) stress, such as a low dose of chemical agent or environmental factor that induces an adaptive beneficial effect. On the contrary, the same stimulus is detrimental at higher doses[53].
In nutrition, hormetins are molecules that can activate the hormetic process; they are produced by plants as a protection against microorganisms, insects and other environmental agents[54]. Some examples of nutritional hormetins are phenolic acids, polyphenols, flavonoids, ferulic acid geranylgeranyl, rosmarinic acid, kinetin, zinc and the extracts of tea, dark chocolate, etc[55].
In human beings, as a reaction to damage, they likely activate stress response pathways, such as the NSPs, in cells as for example after exposure to mild heat stress[56]. The activation of NSPs results in increased production of homeostatic genes and therefore proteins such as heat-shock and others involved in the regulation of cellular energy metabolism or antioxidant enzymes[57].
For example, the isothiocyanates are active compounds obtained from the conversion of glucosinolates by myrosinase, a glucosidase possibly present in human microbiota contained in cruciferous vegetables (cauliflower, broccoli sprouts, cabbage, broccoli and similar green leafy vegetables) typical of the MedDiet. Inside the cell, they stimulate the activation of the nuclear factor erythroid 2-related factor 2 that, consequently, translocates to the nucleus, binding the antioxidant response element, encoding antioxidant enzymes[58].
Polyphenols, classified as phenolic acids, flavonoids, stilbenes and lignans, phytochemicals widely present in fruits and vegetables, including the Mediterranean ones, can regulate the TF NF-κB by reducing the expression of inflammatory cytokines and can activate SIRT1 that may also inhibit NF-κB, reducing the cellular stress response[59-61]. On the other hand, SIRT1 acts on FOXO3A which, as previously stated, modulates genes that encode antioxidant enzymes and other stress-response proteins[25,62].
MedDiet effects on gene expression therefore cause a successful response to environmental changes, allowing better use of nutritional resources and determining epigenetic modifications[25,63].
Phytochemicals, as well as other nutrients, can also influence miRNA expression, a class of small non-coding RNA (19-24 nucleotides) evolutionary conserved that, in turn, regulate post-translational gene expression, suppressing the translation or reducing the mRNA target stability[64]. Moreover, it has been demonstrated that polyphenols can modulate miRNA action on metabolic homeostasis and on chronic diseases but further studies are necessary to identify their targets[64-66].

Mediterranean functional food: the example of EVOO and table green olives

As previously stated, typical Mediterranean foods contain phytochemicals, defined as chemical compounds produced by plants. They are often characterized by phenolic groups, good source of natural antioxidants that may affect health with their anti-inflammatories and anti-cancer properties[25].
EVOO, the main and common food in the Mediterranean basin, is the principal source of fats in the MedDiet regimen. It has many nutraceutical properties thanks to the complex mixture of bioactive compounds[67].
Several studies have shown that it decreases pro-inflammatory environment induced by oxidized low-density lipoproteins (LDLs) both in experimental models and in humans, reducing the levels of C reactive protein, a powerful marker of inflammation, and the levels of pro-inflammatory cytokine interleukin-6 (IL-6)[68-70].
With its high content in MUFAs and polyphenols, EVOO might exert beneficial effects on the development and progression of age-related diseases. In fact, its non-saponifiable portion (1-2%) contains about 230 bioactive molecules, such as carotenoids, mainly lycopene, sterols, and the phenolic compounds oleoeuropein, oleocanthal, hydroxytyrosol and tyrosol, with antioxidant and anti-inflammatory properties[67,70]. Oleic acid is the main MUFA and is claimed to increase the resistance of LDL to oxidation. It has also been suggested that MUFAs can decrease all-cause (11%) and cardiovascular mortality (12%), cardiovascular events (9%) as well as stroke (17%)[68].
Moreover, in models, oleic acid suppresses cytotoxic function of natural killer cells with a consequent anti-inflammatory effect, although it is simply a possible lower response to microorganism infection[71].
In general, unsaturated fatty acids improve endothelial function, decreasing intercellular adhesion molecule-1 production by endothelial cells and reducing leukocyte adhesion[72]. In fact both in vitro and in vivo studies demonstrated the endothelium-protective properties of

these molecules[68,70,73-76].
Some trials showed that polyphenols intake has been associated with low mortality rates caused by coronary heart disease[77,78].
These compounds are able to bind LDLs, increasing resistance to oxidation and acting as radical scavengers[79]. Moreover, it was observed that the consumption of polyphenol-rich olive oil could decrease blood pressure (BP) and improve endothelial function in young women with high-normal BP[75].
Generally, polyphenols inhibit NF-κB pathway that leads to the expression of pro-inflammatory genes[80-82]. In particular hydroxytyrosol and oleocanthal have ibuprofen-like activity, inhibiting the cyclooxygenases 1 and 2, responsible for prostaglandin production[70,80].
As reported in a recent review, also green table olives are an extremely rich source of polyphenols, especially oleuropein and hydroxytyrosol, comprising 1-3 % of fresh pulp weight.
Despite the high levels of hydroxytyrosol in both table olives and EVOO, in humans its bioavailability was proven only in oil[83]. The amount of polyphenols in olives, as well as in oil, is strongly influenced by the variety and the geographical origin. Greek *Koroneiki* have a very high content, while polyphenol content of the Spanish *Arbequina* is low and that of Sicilian *Nocellara* is medium-high[67], with a conceivable anti-inflammatory and anti-oxidant effect.
In 2013, a new compound was extracted from the wastewaters obtained during olive oil production from *Nocellara del Belice* olives. This compound, known as nocellaralactone, is also present in the leaves and appears to be structurally similar to monoterpenoids, secondary metabolites found in higher plants, with a significant *in vitro* anti-inflammatory activity[84]. Moreover, a pilot study demonstrated that daily consumption of table green olives Nocellara del Belice is likely linked to a decrease in IL-6 and malondialdehyde (MDA) levels[85].
MDA is the main product of PUFAs peroxidation and is an important index of oxidative stress[86]. Noteworthy, this study highlighted a reduction of fat mass with an increase of muscle mass in subjects recruited for nutritional intervention. The possible explanation could be linked to the capacity of conjugated linoleic acid (CLA) to reduce body fat levels, strictly linked to production of adipokines (pro-inflammatory cytokines)[87].
CLA is present both in EVOO and table olives and can also be produced during their digestion.
In experimental models, acting as signalling mediator, CLA inhibits lipogenesis, increases fat oxidation and reduces adipocytes size[88,89].
In fact, such levels significantly decreased at the end of the dietary intervention.

Conclusion

The extraordinary increase of elderly population in developed countries underscores the importance of studies on ageing and longevity and the need for a prompt spread of knowledge about these topics, in order to satisfactorily decrease medical, economic and social problems associated with old age.
In fact, European public health policy focuses its attention on the achievement of a healthy lifespan which represents an important challenge[16,90].
According to positive biology, an effective approach is needed to understand the causes of positive phenotypes to try and explain the biological mechanisms of healthy ageing, rather than making age-related diseases the central focus of research[91]. Much evidence shows that dietary restriction[2,8] as well as allelic variations in gene encoding proteins that take part in NSPs can increase lifespan[62,92-99]. This phenomenon is evolutionary conserved but the achievement of dietary restriction is not easy in human beings because it implies a considerable caloric restriction. A close daily adherence to MedDiet, including a healthy lifestyle, seems to be one of the more realistic ways to apply dietary restriction. Moreover, the possibility of managing this pattern, based on the combined use of functional foods, should permit to create a new therapeutic strategy. It would be based not only on a specific bioactive molecule or on a specific food but also on an integrated approach. Starting from local dietary habits, it can be extended to a *nutra-functional diet* applicable worldwide.
Oxidative stress and low grade inflammation play a part in the pathogenesis of age-related diseases and consequently in the ageing process. Thus, it is very relevant to study in greater depth the mechanism of action of nutraceuticals contained in functional foods that may constitute a natural remedy with health benefits through the reduction of cellular and tissue damage with the final aim to prevent or fight age-related diseases[100].

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REFERENCES

1. Montesanto A, Dato S, Bellizzi D, Rose G, Passarino G. Epidemiological, genetic and epigenetic aspects of the research on healthy ageing and longevity. *Immun Ageing.* 2012; 9(1):6.
2. Longo VD, Antebi A, Bartke A, et al. Interventions to Slow Aging in Humans: Are We Ready? *Aging Cell.* 2015; 14(4):497-510.
3. Dato S, Bellizzi D, Rose G, Passarino G. The impact of nutrients on the aging rate: A complex interaction of demographic, environmental and genetic factors. *Mech Ageing Dev.* 2016; 154:49-61.
4. Cannella C, Savina C, Donini LM. Nutrition, longevity and behavior. *Arch Gerontol Geriatr.* 2009; 49 Suppl 1:19-27.
5. Trichopoulou A. Traditional Mediterranean diet and longevity in the elderly: a review. *Public Health Nutr.* 2004; 7(7):943-947.
6. Bürkle A, Caselli G, Franceschi C, et al. Pathophysiology of ageing, longevity and age related diseases. *Immun Ageing.* 2007; 4:4.
7. Franceschi C, Capri M, Monti D, et al. Inflammaging and anti - inflammaging: a systemic perspective on aging and longevity emerged from studies in humans. *Mech Ageing Dev.* 2007; 128(1):92-105.
8. Fontana L, Partridge L, Longo VD. Extending healthy life span--from yeast to humans. *Science.* 2010; 328(5976):321-6.
9. Aiello A, Accardi G, Candore G, et al. Nutrigerontology: a key for achieving successful ageing and longevity. *Immun Ageing.* 2016; 13:17.
10. http://www.nu-age.eu/
11. Santoro A, Pini E, Scurti M, et al. Combating inflammaging through a Mediterranean whole diet approach: the NU-AGE project's conceptual framework and design. *Mech Ageing Dev.* 2014; 136-137:3-13.
12. Jones PJ, Varady KA. Are functional foods redefining nutritional requirements? *Appl Physiol Nutr Metab.* 2008; 33(1):118-23.
13. Avery P, Barzilai N, Benetos A, et al. Ageing, longevity, exceptional longevity and related genetic and non genetics markers: panel statement. *Curr Vasc Pharmacol.* 2014; 12(5):659-61.
14. http://www.un.org/en/development/desa/population/theme/ageing/WPA2015.shtml
15. Caruso C, Candore G. L'invecchiamento e la medicina di genere. In: Caruso C, Candore G, eds. La malattia: dagli sciamani alla medicina di precisione. *Medical Books (Palermo).* 2016; 127-150.
16. Kolovou G, Barzilai N, Caruso C, et al. The challenges in moving from ageing to successful longevity. *Curr Vasc Pharmacol.* 2014; 12(5):662-73.
17. Troen BR. The biology of aging. *Mt Sinai J Med.* 2003; 70(1):3-22.
18. Vasto S, Rizzo C, Caruso C. Centenarians and diet: what they eat in the Western part of Sicily. *Immun Ageing.* 2012; 9(1):10.
19. Vasto S, Scapagnini G, Rizzo C, Monastero R, Marchese A, Caruso C. Mediterranean diet and longevity in Sicily: survey in a Sicani Mountains population. *Rejuvenation Res.* 2012; 15(2):184-8.
20. Balistreri CR, Caruso C, Candore G The role of adipose tissue and adipokines in obesity-related inflammatory diseases. *Mediators Inflamm.* 2010; 2010:802078.
21. Kirkwood TB. Time of our lives. What controls the length of life? *EMBO Rep.* 2005; 6 Spec No:S4-8.
22. Estruch R, Martínez-González MA, Corella D, et al. Effects of a Mediterranean-style diet on cardiovascular risk factors: a randomized trial. *Ann Intern Med.* 2006; 145(1):1-11.
23. Fitó M, Guxens M, Corella D, et al. Effect of a traditional Mediterranean diet on lipoprotein oxidation: a randomized controlled trial. *Arch Intern Med.* 2007; 167(11):1195-203.
24. Casas R, Sacanella E, Estruch R. The immune protective effect of the Mediterranean diet against chronic low-grade inflammatory diseases. *Endocr Metab Immune Disord Drug Targets.* 2014; 14(4):245-54.
25. Vasto S, Buscemi S, Barera A, Di Carlo M, Accardi G, Caruso C. Mediterranean diet and healthy ageing: a Sicilian perspective. *Gerontology.* 2014; 60(6):508-18.
26. Kastorini CM, Milionis HJ, Esposito K, Giugliano D, Goudevenos JA, Panagiotakos DB. The effect of Mediterranean diet on metabolic syndrome and its components: a meta-analysis of 50 studies and 534,906 individuals. *J Am Coll Cardiol.* 2011; 57(11):1299-313.
27. Willett WC, Sacks F, Trichopoulou A, et al. Mediterranean diet pyramid: a cultural model for healthy eating. *Am J Clin Nutr.* 1995; 61(6 Suppl):1402S-1406S.
28. http://www.fimdefelice.org/archives/arc.researchact.html
29. Gupta S, Chauhan D, Mehla K, Sood P, Nair A. An overview of nutraceuticals: current scenario. *J Basic Clin Pharm.* 2010; 1(2):55-62.
30. Vasto S, Barera A, Rizzo C, Di Carlo M, Caruso C, Panotopoulos G. Mediterranean diet and longevity: an example of nutraceuticals? *Curr Vasc Pharmacol.* 2014; 12(5):735-8.
31. Martirosyan DM, Singh J. A new definition of functional food by FFC: What makes a new definition unique? *Funct Food Health Dis.* 2015; 5(6):209-223.
32. Efeyan A, Comb WC, Sabatini DM. Nutrient-sensing mechanisms and pathways. *Nature.* 2015; 517(7534):302-10.
33. Anisimov VN, Bartke A. The key role of growth hormone - insulin - IGF-1 signaling in aging and cancer. *Crit Rev Oncol Hematol.* 2013; 87(3):201-223.
34. Gilmore TD, Wolenski FS. NF-κB: where did it come from and why? *Immunol Rev.* 2012; 246(1):14-35.
35. Newton K, Dixit VM. Signaling in innate immunity and inflammation. *Cold Spring Harb Perspect Biol.* 2012; 4(3). pii: a006049.
36. Rincon M, Muzumdar R, Atzmon G, Barzilai N. The paradox of the insulin/IGF-1 signaling pathway in longevity. *Mech Ageing Dev.* 2004; 125(6):397-403.
37. Gems D, McElwee JJ. Ageing: Microarraying mortality. *Nature.* 2003; 424(6946):259-61.
38. Kenyon C. The plasticity of aging: insights from long-lived mutants. *Cell.* 2005; 120(4):449-60.
39. Brunet A. The multiple roles of FOXO transcription factors. *Med Sci (Paris).* 2004; 20(10):856-9.
40. Martin DE, Hall MN. The expanding TOR signaling network. *Curr Opin Cell Biol.* 2005; 17(2):158-66.
41. Hertweck M, Göbel C, Baumeister R. C. elegans SGK-1 is the critical component in the Akt/PKB kinase complex to control stress response and life span. *Dev Cell.* 2004; 6(4):577-88.
42. Vellai T, Takacs-Vellai K, Zhang Y, Kovacs AL, Orosz L, Müller F. Genetics: influence of TOR kinase on lifespan in C. elegans. *Nature.* 2003; 426(6967):620.
43. Kapahi P, Zid BM, Harper T, Koslover D, Sapin V, Benzer S. Regulation of lifespan in Drosophila by modulation of genes in the TOR signaling pathway. *Curr Biol.* 2004; 14(10):885-90.
44. Hsieh CC, Papaconstantinou J. Akt/PKB and p38 MAPK signaling, translational initiation and longevity in Snell dwarf mouse livers. *Mech Ageing Dev.* 2004; 125(10-11):785-98.
45. Levine ME, Suarez JA, Brandhorst S, et al. Low protein intake is associated with a major reduction in IGF-1, cancer, and overall mortality in the 65 and younger but not older population. *Cell Metab.* 2014; 19(3):407-17.
46. Kennedy BK, Lamming DW. The Mechanistic Target of Rapamycin: The Grand ConducTOR of Metabolism and Aging. *Cell Metab.* 2016; 23(6):990-1003.
47. Sofi F, Abbate R, Gensini GF, Casini A. Accruing evidence on benefits of adherence to the Mediterranean diet on health: an updated systematic review and meta-analysis. *Am J Clin Nutr.* 2010; 92(5):1189-96.

48. Benzie IF, Choi SW. Antioxidants in food: content, measurement, significance, action, cautions, caveats, and research needs. *Adv Food Nutr Res.* 2014; 71:1-53.

49. Rattan SI. Hormesis in aging. *Ageing Res Rev.* 2008; 7(1):63-78.

50. Mattson MP. Hormesis defined. *Ageing Res Rev.* 2008; 7(1):1-7.

51. Southam CM, Ehrlich J. Effects of extracts of western red-cedar heartwood on certain wood-decaying fungi in culture. *Phytopathology.* 1943; 33:517-524.

52. Calabrese EJ. Hormesis: changing view of the dose-response, a personal account of the history and current status. *Mutat Res.* 2002; 511(3):181-189.

53. Calabrese EJ, Bachmann KA, Bailer AJ, et al. Biological stress response terminology: Integrating the concepts of adaptive response and preconditioning stress within a hormetic dose-response framework. *Toxicol Appl Pharmacol.* 2007; 222(1):122-128.

54. Rattan SI. Rationale and methods of discovering hormetins as drugs for healthy ageing. *Expert Opin Drug Discov.* 2012; 7(5):439-48.

55. Rattan SI. Biogerontology: from here to where? The Lord Cohen Medal Lecture-2011. *Biogerontology.* 2012; 13(1):83-91.

56. Gems D, Partridge L. Stress-response hormesis and aging: “that which does not kill us makes us stronger”. *Cell Metab.* 2008; 7(3):200-3.

57. Mattson MP, Cheng A. Neurohormetic phytochemicals: Low-dose toxins that induce adaptive neuronal stress responses. *Trends Neurosci.* 2006; 29(11):632-9.

58. Lee JS, Surh YJ. Nrf2 as a novel molecular target for chemoprevention. *Cancer Lett.* 2005; 224(2):171-84.

59. Mann GE, Rowlands DJ, Li FY, de Winter P, Siow RC. Activation of endothelial nitric oxide synthase by dietary isoflavones: role of NO in Nrf2-mediated antioxidant gene expression. *Cardiovasc Res.* 2007; 75(2):261-74.

60. Scapagnini G, Vasto S, Abraham NG, Caruso C, Zella D, Fabio G. Modulation of Nrf2/ARE pathway by food polyphenols: a nutritional neuroprotective strategy for cognitive and neurodegenerative disorders. *Mol Neurobiol.* 2011; 44(2):192-201.

61. Vauzour D. Dietary polyphenols as modulators of brain functions: biological actions and molecular mechanisms underpinning their beneficial effects. *Oxid Med Cell Longev.* 2012; 2012:914273.

62. Di Bona D, Accardi G, Virruso C, Candore G, Caruso C. Association between genetic variations in the insulin/insulin-like growth factor (Igf-1) signaling pathway and longevity: a systematic review and meta-analysis. *Curr Vasc Pharmacol.* 2014; 12(5):674-81.

63. Corella D, Ordovás JM. How does the Mediterranean diet promote cardiovascular health? Current progress toward molecular mechanisms: gene-diet interactions at the genomic, transcriptomic, and epigenomic levels provide novel insights into new mechanisms. *Bioessays.* 2014; 36(5):526-37.

64. Shukla S, Meeran SM, Katiyar SK. Epigenetic regulation by selected dietary phytochemicals in cancer chemoprevention. *Cancer Lett.* 2014; 355(1):9-17.

65. Srivastava SK, Arora S, Averett C, Singh S, Singh AP. Modulation of microRNAs by phytochemicals in cancer: underlying mechanisms and translational significance. *Biomed Res Int.* 2015; 2015:848710.

66. García-Segura L, Pérez-Andrade M, Miranda-Ríos J. The emerging role of MicroRNAs in the regulation of gene expression by nutrients. *J Nutrigenet Nutrigenomics.* 2013; 6(1):16-31.

67. Aiello A, Guccione GD, Accardi G, Caruso C. What olive oil for healthy ageing? *Maturitas.* 2015; 80(2):117-8.

68. Schwingshackl L, Christoph M, Hoffmann G. Effects of Olive Oil on Markers of Inflammation and Endothelial Function-A Systematic Review and Meta-Analysis. *Nutrients.* 2015; 7(9):7651-75.

69. Caruso C, Candore G. Alimentazione e malattia. In: Caruso C, Candore G, eds. La malattia: dagli sciamani alla medicina di precisione. *Medical Books (Palermo).* 2016; 152-175.

70. Virruso C, Accardi G, Colonna-Romano G, Candore G, Vasto S, Caruso C. Nutraceutical properties of extra-virgin olive oil: a natural remedy for age-related disease? *Rejuvenation Res.* 2014; 17(2):217-20.

71. Yaqoob P, Newsholme EA, Calder PC. Inhibition of natural killer cell activity by dietary lipids. *Immunol Lett.* 1994; 41(2-3):241-7.

72. Bemelmans WJ, Lefrandt JD, Feskens EJ, et al. Change in saturated fat intake is associated with progression of carotid and femoral intima-media thickness, and with levels of soluble intercellular adhesion molecule-1. *Atherosclerosis.* 2002; 163(1):113-20.

73. Frankel EN. Nutritional and biological properties of extra virgin olive oil. *J Agric Food Chem.* 2011; 59(3):785-92.

74. Martín MA, Ramos S, Granado-Serrano AB, et al. Hydroxytyrosol induces antioxidant/detoxificant enzymes and Nrf2 translocation via extracellular regulated kinases and phosphatidylinositol-3-kinase/protein kinase B pathways in HepG2 cells. *Mol Nutr Food Res.* 2010; 54(7):956-66.

75. Moreno-Luna R, Muñoz-Hernandez R, Miranda ML, et al. Olive oil polyphenols decrease blood pressure and improve endothelial function in young women with mild hypertension. *Am J Hypertens.* 2012; 25(12):1299-304.

76. Accardi G, Aiello A, Gambino CM, Virruso C, Caruso C, Candore G. Mediterranean nutraceutical foods: Strategy to improve vascular ageing. *Mech Ageing Dev.* 2016; 159:63-70.

77. Hertog MG, Kromhout D, Aravanis C, et al. Flavonoid intake and long-term risk of coronary heart disease and cancer in the seven countries study. *Arch Intern Med.* 1995; 155(4):381-6.

78. Horn P, Amabile N, Angeli FS, et al. Dietary flavanol intervention lowers the levels of endothelial microparticles in coronary artery disease patients. *Br J Nutr.* 2014; 111(7):1245-52.

79. Gimeno E, de la Torre-Carbot K, Lamuela-Raventós RM, et al. Changes in the phenolic content of low density lipoprotein after olive oil consumption in men. A randomized crossover controlled trial. B*r J Nutr.* 2007; 98:1243-50.

80. Beauchamp GK, Keast RS, Morel D, et al. Phytochemistry: ibuprofen-like activity in extra-virgin olive oil. *Nature.* 2005; 437(7055):45-6.

81. Romier B, Van De Walle J, During A, Larondelle Y, Schneider YJ. Modulation of signalling nuclear factor-kappaB activation pathway by polyphenols in human intestinal Caco-2 cells. *Br J Nutr.* 2008; 100(3):542-51.

82. Zhang X, Cao J, Zhong L. Hydroxytyrosol inhibits pro-inflammatory cytokines, iNOS, and COX-2 expression in human monocytic cells. *Naunyn Schmiedebergs Arch Pharmacol.* 2009; 379(6):581-6.

83. Charoenprasert S, Mitchell A. Factors influencing phenolic compounds in table olives (Olea europaea). *J Agric Food Chem.* 2012; 60(29):7081-7095.

84. Serrilli AM, Frasca G, Rizza L, Bonina FP, Bianco A. Nocellaralactone, a new monoterpenoid with anti-inflammatory activity, from Olea europaea L., cultivar Nocellara del Belice. *Nat Prod Res.* 2013; 27(24):2311-9.

85. Accardi G, Aiello A, Gargano V, et al. Nutraceutical effects of table green olives: a pilot study with Nocellara del Belice olives. *Immun Ageing.* 2016; 13:11.

86. Czerska M, Mikołajewska K, Zieliński M, Gromadzińska J, Wąsowicz W. Today’s oxidative stress markers. *Med Pr.* 2015; 66(3):393-405.

87. Lehnen TE, da Silva MR, Camacho A, Marcadenti A, Lehnen AM. A review on effects of conjugated linoleic fatty acid (CLA) upon body composition and energetic metabolism. *J Int Soc Sports Nutr.* 2015; 12:36.

88. Fazzari M, Trostchansky A, Schopfer FJ, et al. Olives and olive oil are sources of electrophilic fatty acid nitroalkenes. *PLoS One.* 2014; 14(9):e84884.

89. Wang Y, Jones PJ. Dietary conjugated linoleic acid and body composition. *Am J Clin Nutr.* 2004; 6 Suppl 79:1153S-8S.

90. www.europarl.europa.eu/atyourservice/en/displayFtu.html?ftuId=FTU_5.5.3.html

91. Caruso C, Passarino G, Puca A, Scapagnini G. "Positive biology": the centenarian lesson. *Immun Ageing.* 2012; 9(1):5.

92. Bonafè M, Barbieri M, Marchegiani F, et al. Polymorphic variants of insulin-like growth factor I (IGF-I) receptor and phosphoinositide 3-kinase genes affect IGF-I plasma levels and human longevity: cues for an evolutionarily conserved mechanism of life span control. *J Clin Endocrinol Metab.* 2003; 88(7):3299-304.

93. Suh Y, Atzmon G, Cho MO, et al. Functionally significant insulin-like growth factor I receptor mutations in centenarians. *Proc Natl Acad Sci U S A.* 2008; 105(9):3438-42.

94. Willcox BJ, Donlon TA, He Q, et al. FOXO3A genotype is strongly associated with human longevity. *Proc Natl Acad Sci U S A.* 2008; 105(37):13987-92.

95. Albani D, Batelli S, Polito L, et al. A polymorphic variant of the insulin-like growth factor 1 (IGF-1) receptor correlates with male longevity in the Italian population: a genetic study and evaluation of circulating IGF-1 from the "Treviso Longeva (TRELONG)" study. *BMC Geriatr.* 2009; 9:19.

96. Anselmi CV, Malovini A, Roncarati R, et al. Association of the FOXO3A locus with extreme longevity in a southern Italian centenarian study. *Rejuvenation Res.* 2009; 12(2):95-104.

97. Flachsbart F, Caliebe A, Kleindorp R, et al. Association of FOXO3A variation with human longevity confirmed in German centenarians. *Proc Natl Acad Sci U S A.* 2009; 106(8):2700-5.

98. Soerensen M, Dato S, Christensen K, et al. Replication of an association of variation in the FOXO3A gene with human longevity using both case-control and longitudinal data. *Aging Cell.* 2010; 9(6):1010-7.

99. Ziv E, Hu D. Genetic variation in insulin/IGF-1 signaling pathways and longevity. *Ageing Res Rev.* 2011; 10(2):201-4.

100. Aiello A, Accardi G, Candore G, et al. Nutrient sensing pathways as therapeutic targets for healthy ageing. *Expert Opin Ther Targets.* 2017; 21(4):371-380.