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Potential health benefits and components of olive oil: An overview

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The nutritional regime of Mediterranean countries includes olive oil as one of the healthiest components. Olive oils are considered nutritious as it contain phenolic compounds whose health benefits are now widely established and documented. The anti-inflammatory actions of the phenolic compounds are demonstrated in various recent studies. The cumulative data reinforces the statement regarding the consumption of olive oil and its potential health benefits, specifically for preventing cardiovascular diseases, diabetes mellitus, and breast cancer. The phenolic compounds like oleuropein and hydroxytyrosol are abundantly present in extra virgin olive oil that is accountable for its distinctive taste and high stability. Both *in-vitro* and *in-vivo* studies demonstrate that the phenolics present in olive oil are powerful antioxidants. They also exhibit various other important biological activities that help promote the health of an individual to some extent.

Keywords: Olive oil, Hydroxytyrosol, Antioxidants, Oleuropein, Phenolics, Oleic acid.

INTRODUCTION

Natural products obtained from medicinal plants along with nutrition have always played a key role in the health and wellness systems of a considerable part of the entire population. Alternative forms of medicine (apart from allopathy) have been an important factor in the healthcare sector all over the world (Al-Attar et al. 2017). Despite the existence of contemporary hospitals and compliant health-care staff, local Arab people still use traditional therapeutic vegetations as an alternate for allopathic medication for the treatment of various chronic diseases and routine problems such as skin-related ailments, bone fracture, rheumatism, asthma, stomach problems, diabetes, constipation, ENT (ear, nose, and throat) problems, respiratory tract infections, colds, cough and fever, urological diseases, measles, hepatic and spleen diseases, typhoid, epilepsy, toothache, tuberculosis, anemia, hypertension, nervous system disorders, snake bites and scorpion stings along with many other tropical diseases including leishmaniasis, rift valley fever, malaria, and schistosomiasis (Al-Asmari et al. 2020). The olive tree is one of the most important trees in the world due to the economical and beneficial health effects of the oil obtained from its fruit. The olive tree originally comes from the Mediterranean basin due to climate requirements, and seventy five percent of the production of olive oil in the world is from this zone. Spain, Italy, Greece, Turkey, Morocco, and Syria being the main producers (Fraihat et al. 2017). The oil

extracted from olive (*Olea europaea*) contain high amount of lipids which is helpful in preventing the outcomes of primary and secondary cardiovascular disease, also, it improves insulin sensitivity and lipid profile. Along with this, olive oil controls the arterial blood pressure, increases the oxidative stability, and improves the inflammatory biomarkers (Estruch et al. 2006; Hohmann et al. 2016). In Mediterranean culture, it is used as a chief source of fat (Martínez-González et al. 2014). Olive oil contains a high amount of monounsaturated fatty acids (MUFAs), particularly oleic acid, and has a low concentration of saturated fatty acids (SFAs) (Huang and Sumpio, 2008). Fatty acids present in olive oil contain a high amount of oleic acid, about 55-83 %, followed by polyunsaturated fatty acids (4-20 %) and 8-14% SFAs. It also contains 1-2 % of some minor compounds like phenolics, triterpenes, tocopherols, some pigments, and sterols having eminent biological properties (Covas et al. 2015). During the last twenty years, agriculture of olive trees had been developed and spread in the northern part of Saudi Arabia. The Aljouf area exhibits a high potential for olive production due to climate and geographic factors, which made this area the main producer in Saudi Arabia (Alruqaie et al. 2013; Hemida et al. 2014).

OLIVE TREE

The species of the olive plant (*Olea europaea L.)* was one among the pioneers to get domesticated in ancient times. The most important materials produced by this crop are extra virgin olive oil and olive oil and there is a continuous increase in the demand of these two products as their health advantages are well-known by everyone (Ramírez-Tejero et al. 2020). More than 70% of olive trees around the world are cultivated in the European Union's (EU) Mediterranean countries (Greece, Italy, Portugal, and Spain) (Camarsa et al. 2010). The fruits, oils, and leaves of *Olea europaea* (olive tree) demonstrate the maximum antioxidant potential which is now been extensively established. The extracts obtained from the byproducts of the olive tree are used in the food and drug industry as they are loaded with various important phenolic compounds and antioxidants which help in preventing oxidative damage. It is widely known that the olive tree contains various antioxidant components such as hydroxytyrosol, oleuropein, tyrosol, and oleuropein aglycone (Jemai et al. 2008a, 2008b).

OLIVE OIL

As olive oils are very helpful in the prevention of various diseases, it is being used as a key ingredient in the dietary habit of Mediterranean countries. The major nutritional components of olive oil are MUFAs, with oleic acid being the main fatty acid. It also contains some minor compounds acting as a potent antioxidant, for example, hydroxytyrosol (Marcelino et al. 2019). About 2000 years ago, olive oil was mostly used for religious and spiritual purposes as reported in religious scriptures such as the holy Koran, the Bible, and Homer's works (Belarbi et al. 2011). Various types of olive oil are present in the market but the 'extra virgin olive oil' is the most traded one throughout the world. The anti-inflammatory and antioxidant attributes of the olive oil, which helps in preventing various human ailments, became the reason for its ever-increasing popularity (Tuck et al. 2001; Tuck and Hayball, 2002; Covas, 2007).

Virgin Oive Oil

To obtain the extra virgin olive oil, only the preliminary steps i.e. washing, decantation, centrifugation, and filtration are pursued (no other treatment), followed by pressing the olives mechanically. This technique of obtaining the extra virgin olive oil preserves its minor components, thereby preventing the oxidative damage of the oil. Moreover, it also promotes health when ingested (Deiana et al. 2018). It is nutritionally abundant, has excellent sensory characteristics, decreased acidity index and the free fatty acid content is lower than 0.8% (Tarhan et al. 2017). The major portion of the lipid content of extra virgin olive oil is composed of oleic acid (55-83%), followed by about 4-20 % of linoleic and α-linolenic acids and a little amount of stearic and palmitic acids (saturated fatty acids). The lipid content of the olive depends on its ripening stage and the local growing conditions. Fruits harvested in cold regions have more MUFAs (Covas et al. 2015; Paz et al. 2016).

Olive Oil Extraction

Olive oil is extracted by crushing the olives and then segregating both the oil and the fruit pulp by providing an increased pressure condition. Apart from this, processes like extruding the olive oil, post-pressurizing, and re-pressing can be done both by using or not using the hot water. The oil extracted by this process generally demonstrates intense color, faint aroma, and an elevated level of free fatty acids (FFAs)

(Fernández et al. 1997; Gökçebag et al. 2013).

To extract olive oil from its fruit, different extraction technologies are applied which changes the chemical composition of the oil accordingly. The chemical extraction generates the oil that is fit for consumption only by going through the process of refining. The process of refining purifies the extracted olive oil by getting rid of residual solvents (if present) and other forms of impurities. The refined oil does not contain polyphenols, vitamins, phytosterols, and various other naturally occurring components having a low molecular weight (Kamm et al. 2001).

CHEMICAL COMPPOSITION OF OIIVE OIL

The yield of extra virgin olive oil is low, that is why it is costlier than any other kind of olive oil, however, the polyphenolic content is maximum in this type of oil (Kalogeropoulos and Tsimidou, 2014). As the extra virgin olive oil has a minimum content of FFAs, it imparts a subtle aroma, flavor, and pale color (Fernández et al. 1997; Fragaki et al. 2005; Lynch and Rozema, 2013).

The process of filtration removes extra polyphenols (having high polarity) along with water, which, on the other hand, is preserved in an unfiltered form of olive oil. The polyphenolic contents in olive oil change according to the number of technological processes involved. Olive oil contains about 98-99% triacylglycerols (TAG). Triacylglycerols (triglycerides) are a distinct assemblage of glycerol esters linked with different fatty acids. The olive oils predominantly have about 83% of oleic acid, a monounsaturated fatty acid. The remaining percentage is covered by palmitic acid, stearic acid, linoleic acid, and palmitoleic acid. A large number of amphiphilic or lipophylic microconstituents exist in virgin olive oil. Some of them are phytosterols, tocopherols, squalene, phenolic compounds, and derivatives of terpenic acids (Ramirez-Tortosa et al. 2006; Boskou, 2009).

Phenolic contents of olive oil are alcohols or phenolic acids, lignans, flavonoids, and oleuropein derivatives. Its average weight is between 50-100 mg per kg of olive oil which depends on many factors such as agronomic conditions, ripening stages of olives, extraction technology, and storage and packaging procedures (Tuck and Hayball, 2002; Bianco et al. 2002).

The fresh olive's pulp has around 2 to 3% of phenolic compounds existing as esters and glucosides. Nearly, 500 mg/L of polyphenolic compounds are present in virgin olive oils. Its quality and quantity depend upon milling of olives and other processing techniques. For this reason, virgin olive oils significantly have elevated levels of polyphenols as compared to refined or processed olive oils (Bianco et al. 2002; Naczk and Shahidi, 2004).

The phenols largely present in olive oils are glycides (for example, oleuropein), phenols and alcohols (e.g. hydroxytyrosol, tyrosol), and flavonoids (Boskou et al. 2006; Ramirez-Tortosa et al. 2006; Boskou, 2009). The distinct sensory attributes of virgin olive oil i.e. its bitter taste is because of the presence of phenolic compounds. As some of the micro-constituents present in olive oil can dissolve in water, therefore, its phenolic content mainly depends on the extraction procedure (Tuck and Hayball, 2002; Boskou et al. 2006; Visioli et al. 2006).

OIIVE OIL AND ITS MAJOR COMPONENTS

The compounds present in olive oil demonstrate significant biological activities that are dependent on the type of cultivar, geographical origin, and many other factors (Mansour et al. 2016).

The nutritional content of olive oil is composed of unsaponifiable fractions, corresponding to its total fatty acid which is majorly signified by MUFAs. About 2% of the total composition, some minor compounds are present in olive oil which is separated into soluble and unsaponifiable fractions. Two hundred minor compounds are identified until now including hydrocarbonates, tocopherols, phytosterols, pigments, and many other components (Covas et al. 2015; Deiana et al. 2018).

Fatty Acids

About 98-99% fatty acids are present in olive oils, majorly TAG, 55-83% oleic acid esters, 7.5-20% palmitic acid, 3.5-21% linoleic acid, and some other fatty acids like 0.5-5% stearic acid. Regarding stereospecificity, about 40% of the TAG present in olive oil is made up of triolein. On the other hand, two possibilities occur for less recurrent esterification. The first one is the presence of single palmitic acid at *sn*-3 position and two oleic acids at the *sn*-1 and *sn-2* positions. The second possibility is the presence of a single molecule of linoleic acid at the *sn*-2 position surrounded by two molecules of oleic acids (Karupaiah and Sundram, 2007).

Oleic Acid

A debate is constantly going on without reaching any firm conclusion about the role of

oleic acid to reduce the risk of heart diseases (Voelker, 2019). Certainly, substituting saturated fats with MUFA and PUFA reduces the risk of cardiovascular disease but it is difficult to determine its actual reason. It could be because of some biological activities associated with oleic acid or due to the dislocation of saturated fats. It is a known fact that oleic acid does not come under the category of essential fatty acids. The human body itself synthesizes it and no clinical symptoms or signs of its deficiency are reported until now. However, some countries such as the USA and UK (other than the Mediterranean nations) ingest oleic acids in the form of meat and poultry (Visioli et al. 2018). Therefore, the net nutritional intake of oleic acid does not show much difference between the olive oil consumers (people of Mediterranean countries) and subjects from other countries (Dougherty et al. 1987).

Certain ecological studies and clinical trials demonstrated the human evidence of the research depicting that the fatty acid composition of blood indicates the increased level of phospholipids or plasma in the range of 18:1 is associated with a higher risk of heart ailments. A prominent example was given by Würtz et al. (2015), who used the metabolomic studies to demonstrate that the increased concentration of serum MUFA is associated with a higher risk of cardiovascular disease. On the contrary, PUFA behaves in the exact opposite manner. Another similar study has been done by Marangoni et al. (2014) and Block et al. (2008). They reported higher concentrations of MUFAs in patients suffering from myocardial infarction when a comparison is made with control (healthy individuals). Their result also depicts the better prognosis of Ω-6 fatty acids. To summarize, it can be said that the advantages of MUFA (e.g. oleic acid) on cardiometabolics stand on inadequate data (Voelker, 2019). The major drawback in this type of research is based on the fact that the blood concentrations of about 18:1 depict a deprived state of ingestion as oleic acids can be produced *de novo*. Besides, some slow-gathering information is suggesting that the substantial difference in health depends upon the source of oleates, whether they originate from animals or plants (Zong et al. 2018). Although, some researchers (Gillingham et al. 2011) state that using oleic acid in place of saturated fats decreases the total cholesterol along with the low-density lipoprotein cholesterol (LDL-c). Also, using oleic acid by replacing carbohydrates decreases the level of triglycerides as well as LDL-c. Both of these effects will result in a reduced risk of cardiovascular disease.

Phenolic Compounds

Many recent studies suggest the positive health effect of phenolic compounds found in olive oils, mainly extra virgin olive oil (Crespo et al. 2018; Robles-Almazan et al. 2018). The soluble fraction of olive oil is chiefly made up of phenolic compounds such as phenolic alcohols (tyrosol and hydroxytyrosol), phenolic acids, secoiridoids (e.g. oleuropein), flavonoids, and hydroxytyrosol connected with dialdehydic type of 3,4-EDA (elenolic acid) (Rodriguez-Morato et al. 2016).

When olive oil is extracted, its acidity rises above 0.8% which needs to get removed (refined). The process of refining considerably decreases the concentration of minor compounds present in olive oil. This, in turn, generates two commercial olive oil types, namely, olive oil and extra virgin olive oil (European Communities, 2002). The former generally lacks all the minor compounds, hence, the nutritional, as well as pharmaceutical characteristics of olive oil is specifically pertained by extra virgin olive oil.

Oleuropein

The olive plant contain oleuropein which is one of the most common biologically active compounds and serves as an antioxidant. It is made up of glucose, elenolic acid, and hydroxytyrosol (3, 4-dihydroxyphenyl ethanol) (Manna et al. 2002; Barbaro et al. 2014). Oleuropein consists of a carbohydrate group alongside the skeleton of oleosidic compounds which makes it a hydroxytyrosol ester (Figure 1).

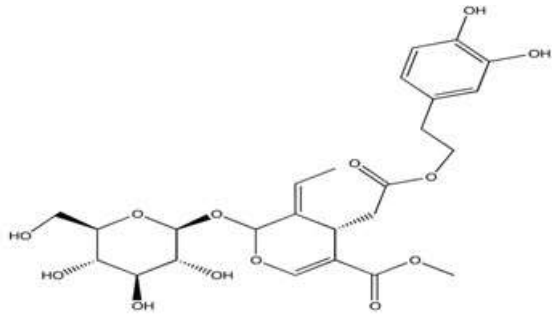


Figure 1: The chemical structure of oleuropein (Shamshoum et al. 2017).

Some studies suggest that the concentration of oleuropein can reach up to 140 mg g $^{-1}$ of dry weight in the fruits and 60-90 mg/g (6 to 9%) in the leaves of the olive plant (Servili et al. 1999). Various other researches also indicate the

concentration of oleuropein in olive leaves as 19% (w/w) (Omar, 2010).

OLIVE OIL MINOR CONSTITUENTS

Certain vitamins like α- and γ- tocopherol (about 200 mg per kg) together with β-carotene and various other components such as squalene, phytosterols, terpenic acids, pigments, flavonoids (e.g. quercetin, luteolin), and some phenolic compounds (polyphenols) are present in virgin olive oil. β-carotene, along with chlorophyll is accountable for imparting the oil its distinct color (Boskou, 2000).

Squalene

Olive oil is considered an important food source of squalene and contains a considerable amount of it, about 500-700 mg per kg. As the olive oil consumers are found to be protected from atherosclerosis, it is worth mentioning that squalene strongly demonstrates the inhibitory action on β-hydroxy-β-methylglutaryl-CoA reductase enzyme (HMG-CoA reductase). HMG-CoA reductase is an important enzyme that helps in the synthesis of cholesterol. The inhibitors of this enzyme are successfully utilized as a drug to decrease the cholesterol level in the blood. Also, they represent antiatherosclerotic and pleiotropic effects like inhibiting the proliferation of smooth muscle cells (Bellosta et al. 2000).

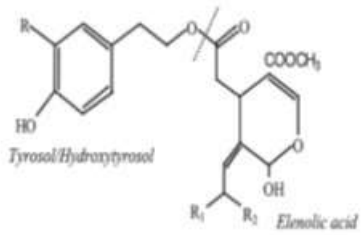
Olive Oil Phenols

The concentration of phenols present in olive oil fluctuates between 150-700 ppm. This fluctuation in the concentration depends upon many factors. The first factor is the chosen variety/cultivar of the olive plant, for example, olives obtained from the Coratina cultivar have maximum phenolic content. The second factor is the degree of maturation as the phenolic content in olive oil/olives generally decreases with the increase in its maturity. Other factors such as climatic conditions, production stages, and infestation with *Dacus oleae* (the olive fruit fly) also affect the number of phenolic compounds present in olives (Boskou, 2000).

In brief, the hand-picking of olives at the stage where the color of its skin changes from light green to dark brown, quick mill transportation, crushing and pressing it in a clean plant without any delay and at a temperature below 25-30 ºC gives a yield of premium olive oils that are rich in phenolic content (Boskou, 2000).

The Chemistry of Phenols of Olive Oil

The complex phenols in olive fruit are present in the form of glycosides but its oil contains the aglyconic structure, i.e. the increased fat-soluble molecular residues (Figure 2) (Visioli and Galli, 2002).



R	R_1	R_2	
OH	H	H	Oleuropein
H	H	H	Ligstroside
H	OH	OH	10-hydroxyligstroside
OH	OH	OH	10-hydroxyoleuropein

Figure 2: Structures of the most representative olive oil phenolics. Hydroxytyrosol derives from oleuropein by hydrolysis where indicated (Visioli and Galli, 2002).

HEALTH BENEFITS OF OLIVE OIL CONSUMPTION

As olive oil increases high-density lipoprotein (HDL-c) concentration and decreases the LDL-c concentration, consuming it as a whole, its isolated compounds (or synergism) can impart primary and secondary protection in contradiction of developing cardiovascular disease. Besides, it affects the inflammatory biomarkers which are regarded as the pro-inflammatory components inside the human body such as tumor necrosis factor and interleukin-6. These compounds are also found helpful in increasing the health of the intestine as they can stimulate and enhance the balance of a wide range of useful gut microbiota (Marcelino et al. 2019). These advantages are accredited due to its nutrient content which is mainly represented by MUFA, having oleic acid (C18:1) concentration of about 55-83%. This is followed by PUFA, such as linoleic acid (C18:2) and linolenic acid (C18:3) making up to 4-20% of the total nutritional content. Some minor but nutritionally significant compounds are also present like phenols, hydroxytyrosol, and oleuropein (Covas et al. 2015; Paz et al. 2016; Giner et al. 2016; Foscolou et al. 2018).

There is a lower risk of getting inflammatory diseases because olive oils are a poor source of

FFAs. Fatty acids can cause cell apoptosis which is a major factor associated with inflammation and creating insulin resistance in the body (Roncero-Ramos et al. 2018). Consuming extra virgin olive oil can also help in controlling blood pressure both in hypertensive and normotensive individuals which are connected with the presence of high amount of MUFA, particularly, oleic acid. MUFA can also help in reducing many other cardiac-related risk factors (Martín-Peláez et al. 2017).

Oleic acid also protects the intestinal lining/mucosa by reducing the production of hydrochloric acid, thereby preventing the formation of ulcers (Piroddi et al. 2017).

Some minor components are also present in olive oils such as vitamin E which improves the stability and enhances the sensory as well as nutritional characteristics along with acting as an antioxidant (Covas et al. 2015; Hohmann et al. 2016; Martín-Peláez et al. 2017; Venturini et al. 2015).

Hydroxytyrosol is an antioxidant present in extra virgin olive oil which has gained remarkable attention in late literature (Foscolou et al. 2018). Hydroxytyrosol displays anti-teratogenic and anti-inflammatory actions, improves the blood lipid profile, reduce the inflammatory cell activation as well as oxidative damage. Furthermore, it also helps in expressing peroxisome proliferators-activated receptors gamma- and alpha- which reduces the size of adipocyte (Foscolou et al. 2018; Castro-Barquero et al. 2018).

Cardiovascular Diseases and Olive Oil

The intake of extra virgin olive oil acts as a primary preventive measure for those individuals who are not suffering from cardiovascular diseases (Hohmann et al. 2016; Shen et al. 2015). Furthermore, its consumption can also be beneficial as a secondary preventive measure for diseased persons as it has the potential to reduce the synthesis of LDL-c and increase the production of HDL-c, which in turn triggers the reverse transfer of cholesterol, thereby, slowing the process of developing illness and reducing the threat of upcoming events that can cause heart ailments (Hohmann et al. 2016; Shen et al. 2015; Avci et al. 2018).

The phenolic compounds, present in extra virgin olive oil act as protective agents against the oxidation of low-density lipoprotein cholesterol. The oleic acid has the potential to minimize these kinds of oxidation, hence adjusting the flow of cholesterol and decreasing the level of circulatory low-density lipoprotein cholesterol (Covas et al. 2015; Hernáez et al. 2017). This study was done to evaluate the effects of Mediterranean dietary intake containing olive oil as the main ingredient. The study was conducted on 68 individuals for about one year. The researchers observed an increased resistance in the oxidation of low-density lipoprotein. Also, a reduction in the alteration process occurring due to oxidative damage was observed along with the increment in the particle size of low-density lipoprotein (Hernáez et al. 2017).

The phenolic compounds present in extra virgin olive oil works as an antioxidant. It prevents and reduces the occurrence of cardiovascular diseases by inhibiting lipid peroxidation triggered by heavy metals or free radicals, hence reducing the oxidation of high-density lipoprotein and terminating the action of low-density lipoprotein. These phenols restrain the reactions of superoxide and disrupt the propagation phase of oxidation chain reactions (Berrougui et al. 2015; Katsarou et al. 2015; Katsarou et al. 2016). The consumption of extra virgin olive oil also regulates hypertension. This was observed in individuals taking olive oil for 12 weeks. The systolic and diastolic pressure was normalized in these individuals when a comparison was made with individuals taking butter (62.5% saturated fatty acids) in their diet (Hernáez et al. 2017).

Gut Microbiota

There is an influential role of the environment, genetics, and dietary factors in the proliferation of intestinal microbiota. Extra virgin olive oil promotes intestinal health by supporting the richer biodiversity of probiotics/gut microbiota (Hidalgo et al. 2018).

Neuropsychiatric Disorders

In an animal experimentation study, the researchers fed the mice with extra virgin olive oil and compared it with the control (mice fed on a diet without olive oil). They observed that the mice fed on olive oil have lesser neuropathology for Alzheimer's disease as compared to the control (Qosa et al. 2015). These effects of olive oil along with the Mediterranean diet is not only specific to certain brain outcomes such as cerebrovascular diseases (for example, stroke) and cognition but it is also related with a decreased depression risk, as stated in various individual and meta-analysis studies (Psaltopoulou et al. 2013).

Chemoprevention

Olive oil intake has been linked with a lower

incidence of breast cancer (Calahorra et al. 2020).

Various early epidemiological studies have described that adhering to the Mediterranean diet can reduce the risk of developing breast cancer. Particularly, it can prevent the onset of postmenopausal breast cancer (Cottet et al. 2009; van den Brandt and Schulpen, 2017).

Many *in vitro* researches have suggested that the minor components abundantly present in extra virgin olive oil are more potent in imparting beneficial effects rather than the major compounds. Olive oil demonstrates multiple chemopreventives including inhibition of tumor progression, abnormal cell proliferation, and amplified apoptosis (Casaburi et al. 2013).

Type 2 Diabetes

There is a substantial amount of evidence that explains the association of high dietary intake of olive oil with improved risk factors of cardiometabolics and type 2 diabetes, particularly because it contains a high amount of monounsaturated fatty acid and polyphenols. A significant reduction in the risk of developing type 2 diabetes has been observed in individuals taking a Mediterranean diet together with extra virgin olive oil when compared with a controlled diet (without olive oil) (Salas-Salvado et al. 2014).

Antineoplastic Properties of Phenols in Olive Oil

Several studies have described the anticancerous properties present in leaf extracts of the olive tree through animal experimentation. The leaf extracts result in the death of the cancerous cells, triggering early apoptosis followed by necrosis (Mijatovic et al. 2011; Bhatia et al. 2009).

Synergism is observed when polyphenols present in extracts of olive leaves are combined with other antineoplastic agents (Mijatovic et al. 2011). The anticancerous potential of polyphenols present in olive oil such as hydroxytyrosol and oleuropein was confirmed by many studies. These compounds inhibit the proliferation of cancerous cells and induce cancer-cell death as seen in cases of neuroblastoma, osteosarcoma, and breast cancer.

Health Benefits of Hydroxytyrosol

Hydroxytyrosol is a major polyphenolic compound present in extra virgin olive oil. It shows anti-teratogenic and anti-inflammatory properties; also, it improves the lipid profile, activates the inflammatory cells, and reduces the oxidative stress (Foscolou et al. 2018).

The positive health effects of polyphenols are attributed to its ability to act as a free radical scavenger for reactive nitrogen/oxygen species. It also activates antioxidant systems present inside the human body. The studies conducted on alloxan-induced type 2 diabetic rats persuasively confirmed the free radical scavenging actions of hydroxytyrosol (Jemai et al. 2009).

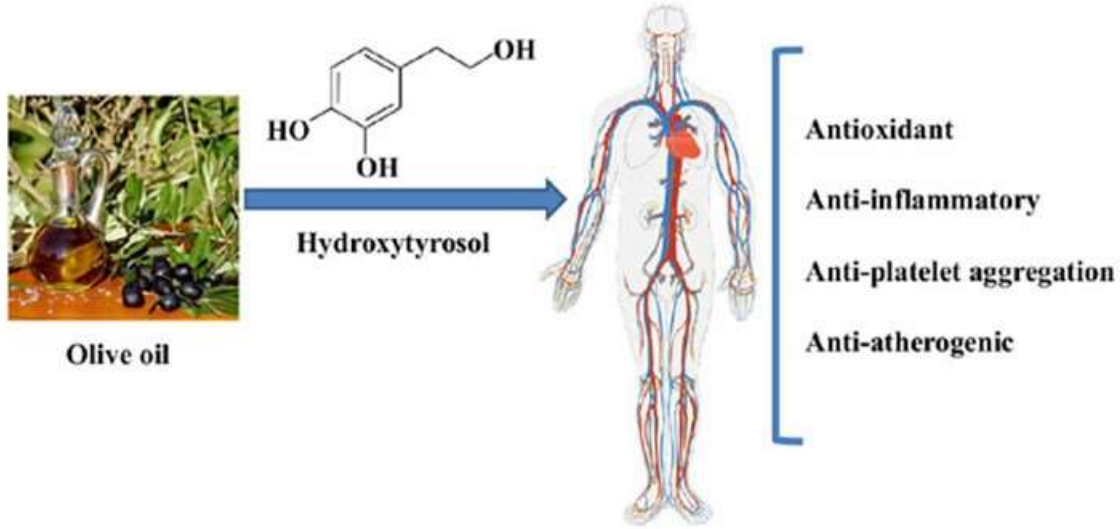


Figure 3: Hydroxytyrosol exerts protective effects against cardiovascular diseases (Tejada et al., 2017)

Hydroxytyrosol can stimulate the biosynthesis of mitochondria by upregulating PGC-1α which was decreased during the induction of type 2 diabetes as depicted by the studies conducted on the cell line of 3T3-L1 adipocyte cells. The adipocyte cells have fewer hydroxytyrosols which in turn elevates the mitochondrial respiratory chain expression, together with ATP synthase. Hydroxytyrosol guards the mitochondria opposing the reduction of mitochondrial DNA synthesis and modifies the activity of critical transcription factors like transcription factor A and nuclear respiratory factor 1. These exceptional properties demonstrated by hydroxytyrosol helps in reducing the risk factors associated with the induction of diabetes mellitus (Hao et al. 2010). Hydroxytyrosols present in olive oil exhibit anti-inflammatory, antioxidant, anti-atherogenic, and anti-platelet aggregating properties both in animal models and *in vitro* studies. Though, its potential therapeutic role in humans still requires further clinical trials (Figure 3) (Tejada et al. 2017).

Health Benefits of Oleuropein

Oleuropein is another compound present in olive leaves and immature olive fruits (14% of dry weight) which exhibits antioxidant properties. It is also found helpful in improving the anti-inflammatory properties in many inflammatory models (Soler-Rivas et al. 2000). Apart from that, it also represents anti-proliferative and tumor-suppressing properties by inducing apoptosis of the cancerous cells (Hohmann et al. 2016; Giner et al. 2016; Scoditti et al. 2015).

In an animal experimentation study, a reduction of 64% and 16% was observed in subjects with colorectal cancer when ingested with 50 and 100 mg/kg oleuropein respectively. A reduction in the inflammation of the colon was observed at the higher dose, thereby providing protection for epithelium cells and suppressing the development of new tumor cells (Giner et al. 2016).

After hydrolysis, the derivative forms of oleuropein such as glycoside and aglycon are more commonly observed in olive oil as a very small quantity of oleuropein is present in it (Soler-Rivas et al. 2000). The oleuropein can modulate the expression of various genes and can modify a variety of signaling proteins that are the key factors for cell proliferation and apoptosis which help provide the above-mentioned effects of oleuropein (Shamshoum et al. 2017). It acts against many bacteria, fungi, molds, viruses, and even parasites. Also, it prevents the aggregation of blood platelets (Benavente-Garcıa et al. 2000).

Oleuropein is an important element of a formula that has been patented and used as an inhibitor for endothelial cell proliferation. Ingesting oleuropein orally can lower down the growth of new blood vessels thus, acting as a powerful anti-angiogenic agent (Hamdi et al. 2002).

Phenolic composites such as oleuropein and protocatechuic acid present in extra virgin olive oil can inhibit LDL-c oxidation which is mediated by macrophages (Masella et al., 2004). Extracts obtained from the leaves and fruits of the olive tree are loaded with oleuropein which protects the β-cell line of the pancreas (insulin producer) against the harmful consequence of cytokines (Cumaoglu et al. 2011).

These findings indicate that oleuropein improves postprandial glycaemic profile *via* hamperi ng Nox2-derived oxidative stress.

HUMAN TRIALS

So far, only two trials of hydroxytyrosol have been done on human beings. These studies are carried out by Lopez-Huertas and coworkers (Gonzalez-Santiago et al. 2010; Lopez-Huertas and Fonolla, 2017). In the first study, the absorption of hydroxytyrosol was confirmed in humans and its temporary linkage with low-density lipoprotein was recorded. This study confirmed the data of Bonanome et al. (2000) who used extra virgin olive oil instead of pure hydroxytyrosol. In the second study (non-placebo), the authors administered 5 mg per day of hydroxytyrosol to subjects having minor hyperlipidemia for about eight weeks. This was done to explore its effect on the markers of blood lipid profiles, cardiovascular diseases, inflammatory biomarkers, electrolyte balance, and kidney/liver functions. They did not report any significant difference except for the plasma concentration of vitamin C which gets doubled after four and eight weeks when a comparison was made with the baseline levels. Through the raised levels of endogenous vitamin C, the researchers proposed an antioxidative function for hydroxytyrosol which was physiologically significant (Afshin et al. 2019). From the above-mentioned discussion, it can be said that the actual involvement of increased antioxidant levels on human pathophysiology is outlying from being revealed.

Some other human trials have been done on wastewaters collected from olive mills. The results include elevated plasma concentration of glutathione (Visioli et al. 2009), decreased production of thromboxane B2 (Leger et al. 2005),

psoriasis amelioration (Acosta et al. 2016), and reduced pain and inflammation in women after breast cancer surgery (Martinez et al. 2019). A number of patents are available for different methodologies used for concentrating and purifying the polyphenols present in wastewaters obtained from olive mills such as reverse osmosis and ion-exchange chromatography (Visioli and Bernardini, 2011). The commercial application of polyphenols present in olive oil ranges from functional foods/nutraceuticals, cosmetology to animal feed.

ANIMAL STUDIES

After the preliminary research by Schaffer et al. (2007), some suggestions emerge out demonstrating the neuroprotective nature of phenolic compounds present in olive oil, for example, a reduction in nervous damage was observed after brain ischemia (De La Cruz et al. 2015). Accumulation of hydroxytyrosol was also observed in the brain of rats fed with olive oil containing hydroxytyrosol (López de las Hazas et al. 2015) but a decreased quantity of brain-induced neurotrophic factors are found in the hippocampus which is a harmful trait however not confirmed yet (Carito et al. 2014; De Nicoló et al. 2013). Extra virgin olive oil also enhances the learning capabilities and memory of the mice probably by modulating the antioxidant system (Farr et al. 2012; Pitozzi et al. 2012).

Another evolving research area is the study of musculoskeletal disorders like loss of bone tissues (after menopause or induced by drugs) and arthrosis/arthritis. Lots of evidence is collecting every day indicating that phenolic compounds present in olive can find a firm place alongside the contemporary medical applications (Hagiwara et al. 2011; Keiler et al. 2014).

The positive effects of MUFA have been found on the lipid profile of blood when replaced with the same quantity of SFA, as reported in some earlier scientific studies (Gillingham et al. 2011). Conversely, a recent report on the meta-analysis of nine cohort analyses suggests no significant connection between the ingestion of MUFA and cardiovascular disease risk factors (Chowdhury et al. 2014). Various studies demonstrated the comparison of olive oil with other oils such as sunflower and flaxseed oil that shows the better results of olive oil in oxidizing LDL-c, concentrating lipoprotein, and reducing its size (Aguilera et al. 2004; Harper et al. 2006). Meta-analysis and systematic review of various studies demonstrated the positive effect of olive oil in improving the release of HDL-c when compared with other seed oils Ghobadi et al. (2019), compared the olive oil with other plant oils. The results indicated a more significant increase in the level of high-density lipoprotein in the case of olive oil (1.37 mg/dl: 95% CI: 0.4, 2.36). Consuming olive oil also reduced the total cholesterol level (6.27 mg/dl, 95% CI: 2.8, 10.6), triglycerides (4.31 mg/dl, 95% CI: 0.5, 8.12) and LDL-c (4.2 mg/dl, 95% CI: 1.4, 7.01) significantly more than any other seed oils. No significant difference was observed on apolipoprotein A1 and B olive oil decreases the total serum cholesterol, triglycerides, and LDL-c and increases the production of HDL-c to a greater extent when compared with other oils (Ghobadi et al. 2019).

CONCLUSION

Consuming the Mediterranean diet is said to improve the overall quality of life. It has been indicated both in human studies and animal trials that ingesting extra virgin olive oil promotes health by reducing the risk of cardiovascular diseases, improving inflammatory responses, enhancing the growth and proliferation of healthy gut microbes, etc. These advantages are due to the excellent nutritional content present in extra virgin olive oil such as elevated levels of MUFAs (especially oleic acid) and some minor components like phenolics (hydroxytyrosol and oleuropein).

In spite of having so many health benefits, the mechanisms involved in these progressions are unknown, particularly the individual action of a compound and the possible synergism with other compounds along with its health effects. Thus, in order to fully understand the possible mechanism of action of these important compounds in human/animal metabolism, further *in vitro* and *in vivo* studies are still needed to access the new findings.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

AUTHOR CONTRIBUTIONS

The authors were contributed equally and have been involved in the writing of the manuscript at draft, any revision stages, and have read and approved the final version.

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