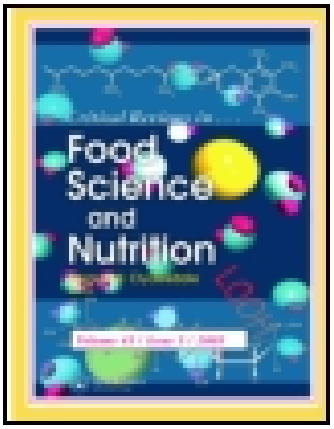

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Healthy virgin olive oil: a matter of bitterness

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Healthy virgin olive oil: a matter of bitterness

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Abstract

Virgin olive oil (VOO) is the pillar fat of Mediterranean diet. It is made from olive fruits and obtained by squeezing olives without any solvent extraction. Respect to the seed oils, an unique polar polyphenol-rich fraction gives to VOO a bitter and pungent taste.

The recent substantiation by European Food Safety Authority (EFSA) of a health claim for VOO polyphenols, may represent an efficient stimulus to get the maximum health benefit from one of the most valuable traditional product of Mediterranean countries educating consumers to the relationship between the VOO bitterness and its health effect.

Agronomical practices and new processing technology to avoid phenolic oxidation and hydrolysis and to enhance the aromatic components of the VOO have been developed and they can be used to modulate taste and flavour to diversify the products on the market. VOOs having high concentration of phenol compounds are bitter and pungent therefore many people do not consume them, thus loosing the health benefits related to their intake.

In this paper the chemist's and nutritionist's points of view have been considered to address possible strategies to overcome the existing gap between the quality perceived by consumer and that established by expert tasters. Educational campaigns emphasizing the bitter-health link for olive oils should be developed.

Keywords: secoiridoids, polyphenols, virgin olive oil, bitter taste, EFSA, Mediterranean diet.

Introduction

Several estimations of daily dietary polyphenol intake are present in the literature (Radtke et al., 1998; Saura-Calixto et al., 2007; Manach et al., 2004; Ovaskainen et al., 2008). According to those studies dietary polyphenol intake ranged from 222 mg/die of Radtke and co-workers (1998) to 2600-3000 mg/die of Saura-Calixto and co-workers (2007). This huge range was mainly due to the food included in the estimations and to the food preferences of the people living in the geographical area where the studies were performed. The low polyphenols intake reported by Radtke and co-workers (1998) was derived from the literature and did not take into account cereal products (Mattila et al., 2005). On the contrary, Saura-Calixto and co-workers (2007), included in their estimation cereal products, which are a very rich source of phenols bound to dietary fibre (for a review see Vitaglione et al., 2008). All in all there is a consensus that fruits, vegetables, cereals, coffee and tea are the major contribute to polyphenols intake worldwide (Saura-Calixto et al., 2007; Manach et al., 2004; Ovaskainen et al., 2008).

Looking at Mediterranean diet among potential polyphenols dietary contributors, also virgin olive oil (VOO) has to be included. Its daily consumption ranges, from 25 to 50 mL/die (Hardin-Fanning, 2008) and the average concentration of phenol compounds in commercial VOO ranges from 100 to 300 mg/kg (Montedoro et al., 1993; Visioli et al., 1995; Owen et al., 2000) despite some authors reported also concentrations > 600 mg/kg in VOO obtained from particular olive cultivars as Tonda Iblea (Sicily, Italy) or Picual (Andalucia, Spain) VOO (Galvano et al, 2007; de La Torre-Carbot, 2010). Therefore considering an average polyphenols intake of 1600 mg/die VOO may theoretical contribute for 0.3-0.6% of it; the contribution potentially being even higher than 1.8% in the case of VOO particularly rich of phenolics. Although this amount can be regarded as negligible, it is worth to notice that VOO may afford to the diet with a group of phenol compounds belonging to the secoiridoid family which is absent in other Mediterranean diet foods (**Table 1**).

EFSA recently approved an health claim stating that the dietary intake of VOO polyphenols is able to prevent LDL oxidation (EFSA journal, 2011). Therefore an increase of VOO polyphenols consumption without increasing the intake of fat would benefit the general population and should be recommended by health authorities. Unfortunately, polyphenols-rich VOOs are bitter and pungent so they dislike to most people who prefers sweet oil (such as refined olive oil or seed oils) thus missing this health opportunity.

In this paper we focus on sensory markers of VOO polyphenols, i.e. bitterness and pungency. The chemist's and nutritionist's points of view will be explored in parallel to consumer's one, aiming at showing the role of VOO sensory markers in human health and to finally address possible short and long term strategies to overcome the gap actually existing between quality perceived by consumer and healthy VOO.

Chemistry of VOO phenolics

The major VOO polyphenols are secoiridoids, simple phenols, and lignans. The chemical structures of VOO polyphenol compounds are reported in **Figure 1**.

In olive secoiridoid structure, namely ligstroside or oleuropein glucoside, is made up of three blocks: phenolic acid that can be monohydroxylated (tyrosol) or dyhydroxylated (hydroxityrosol); elenolic acid (EA) and sugar moiety (mainly glucose). During ripening specific glycosidase hydrolyses the sugar moiety and the aglycons having intermediate polarity can be

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retained in the oil phase during the extraction process. However, secoiridoid aglycons are not stable and undergo to two principle degradation phenomena: i) opening of heterocyclic structure of elenolic moiety giving the dialdehydic compounds that are often the main form retrieved in the oil; ii) hydrolysis of the ester bond with formation of phenolic acid and elenolic acid. The hydrolysis spontaneously occurs during storage and it is favoured by the acidic pH (Covas et al., 2006).

All together tyrosol, hydroxytyrosol and their secoiridoid derivatives account for 60-90% of the total phenol compounds of a VOO (De la Torre et al., 2005). Anyway their final concentration in VOO can be modulated by olive variety and by several technological factors thus influencing both nutritional and sensory properties of VOO. All these issues have been discussed below.

Nutritional benefits of VOO phenolics

Many reviews focused on the nutritional properties of VOO phenolic compounds have been published in the last years (Cicerale et al., 2011; Cicerale et al., 2010; Corona et al., 2009; Covas 2007; Fito et al., 2007; Covas et al., 2006; Covas et al., 2009). *In vitro* and animal studies, conducted using some pure compounds typical of VOO (hydroxytirosol, oleocanthal, etc) or VOO polyphenol extracts, concurrently attributed to these compounds many health benefits. In many cases these studies did not allow the transposition of the results to humans as the concentrations used in the studies were too high to simulate human consumption through VOO, or the tested outcomes did not take into account their metabolism and biotransformation.

Randomized, crossover, controlled human studies in healthy subjects and in patients showed that the protective effects of VOO phenol compounds are clearly detected in humans under oxidative stress conditions i.e. in elderly people, in males subjected to a very strict antioxidant diet, and in patients with hyperlipidemia, peripheral vascular disease, or coronary heart disease. Unfortunately the compliance of the VOO polyphenols dietary intervention, i.e. biomarkers of intake of VOO phenols, were not measured. In fact, although there is a great scientific attention on *in vivo* properties of VOO, the issue of having a reliable biomarkers of intake, is still unresolved.

In the early VOO phenolic compounds bioavailability studies (Visioli et al., 2000; Miro-Casas et al., 2003a; Miro-Casas et al., 2003b; Miro-Casas et al., 2001) hydroxytyrosol was considered as biomarker of VOO intake despite it is also a well-known metabolite of dopamine and its systemic concentration is also influenced by other dietary components, such as alcohol. For this reason other authors do not consider it an appropriate biomarker of intake and function (de la Torre, 2008) and others suggested that tyrosol might be a better biomarker of VOO consumption for clinical studies (Miro-Casas et al., 2003). Recent evidence on human metabolism of oleuropein and ligstroside aglycon (summarized in **Figure 2**) suggested that it is possible to focus on other compounds that are specific of VOO and not inter-related with other metabolic pathways, such as elenoic acid and its metabolites. García-Villalba et al., (2010) showed that the hydrogenated metabolite of elenoic acid was the most abundant compound in human urine over 6h following consumption of 50 mL VOO; but the recovery within 6 hours from VOO consumption would limit its use as a short-term biomarker. Valls and co-workers (2010) showed that a higher concentration of hydroxy-flavanone type compound (2.90±0.04 µM vs 1.5±0.04 µM) and a catecholamine derivative (0.70±0.03 µM *vs* 0.56±0.03 µM) was in plasma of VOO consumers (50 mL/die, ~7 mg/die total phenols) than in non-consumers' one but these compounds were only indirectly related to the long-term regular VOO consumption.

In 2011, on the basis of "*a well conducted and powered study, and two smaller-scale studies, showing a dose-dependent and significant effect of olive oil polyphenol consumption (for three weeks) on appropriate markers of LDL peroxidation (oxLDL)*", and considering that "*these results were supported by one short-term and one acute study, and by supportive markers of LDL peroxidation (conjugated dienes, ex vivo resistance of LDL to oxidation) going in the same direction*", EFSA panel stated that "*a cause and effect relationship has been established between the consumption of olive oil polyphenols (standardized by the content of hydroxytyrosol and its derivatives) and protection of LDL particles from oxidative damage*" (EFSA Journal, 2011). The Panel also specified that the condition of use to have the health benefit are "*20 g of an olive oil with a polyphenol content of 200 mg/kg or a minimum amount of 2 mg per day of hydroxytyrosol* " and that "*the concentrations in some olive oils may be too low to allow the consumption of this amount of polyphenols in the context of a balanced diet*".

Beside the potential systemic benefits also the effects of VOO phenols on the gastro-intestinal (GI) tract are very interesting (for a review see Corona et al., 2009). Several *in vitro* studies showed the dose-dependent effect of VOO phenols as antimicrobial, antiinflammatory or chemopreventive substances on gastric or intestinal cells (Romero et al., 2007; Dell'Agli et al., 2010; Fini et al., 2008; Khanal et al., 2011; Sangiovanni et al., 2012). Nutritionists should actually recommend consumption of VOO rich of phenol compounds for their beneficial activities along the upper GI tract where their concentration is only influenced from the content of VOO consumed and there is no effect of bioavailability and biotransformation.

Factors affecting the concentration of phenol compounds in VOO

It is possible to modulate several agronomic and processing factors to produce VOO having a phenol concentration higher than 200 mg/kg in order to meet EFSA requirement for the health claim.

The main factors affecting the compositions of olive oil polyphenols are individually described as follow.

Cultivar and pedoclimatic environment: Olive polyphenols content depends both quantitatively and qualitatively from the olive variety. Different varieties, cultivated in the same environment and processed at a fixed ripening stage, produce VOO with different total polyphenols content. Due to this strict correlation with genotypes and environment, VOO phenol compounds are important markers of typical oils according to the origin area (Dabbou et al., 2011; El Riachy et al., 2011; Rotondi et al., 2010; Vekiari and Koutsaftakis, 2002; Temime et al., 2006; Tura et al., 2007; Tura et al., 2008).

Agronomic practices (irrigation and rainfall): Phenol composition of olive oil seems influenced by the irrigation management of the origin olive plants. The effect of amount and time of water applied to the plants on phenol composition of VOO is still controversy. Some studies report that the olive plants grown on soils with an intense and long water deficit have an early ripening and give polyphenol-rich VOO. On the contrary other authors report no effect, or even an increased phenol content, in VOO obtained by olives from irrigated plants (particularly during the first stages of fruit ripening) compared to rain fed ones (Ahmed et al., 2009; Bucelli et al., 2011;

Gomez-Rico et al., 2007; Motilva et al., 1999; Motilva et al., 2000; Patumi et al., 2002; Stefanoudaki et al., 2009; Tovar et al., 2001).

Fly attack (Bactrocera oleae): A decreased phenol content of VOO from olives attacked by *Bactrocera oleae* has been reported. The severity of the effect depends from the olive variety, the time of the attack in relation to the olive grown, and from the intensity of the attack. In general, VOO obtained from olives with nymphal state or with the exit holes of the pest can register a decrease of phenolic compounds compared to VOO obtained by intact fruits (Evangelisti et al., 1994; Gomez-Caravaca et al., 2008; Montedoro et al., 1985; Pereira et al., 2004; Tamendjari et al., 2004; Tamendjari et al., 2009).

Ripening: Polyphenol concentration in olive increase with ripening until the “Cherry” stage. After that, a sharp reduction of polyphenols until the “Black maturation” phase, that is a rapid decline of polyphenols due to increased activity of hydrolytic enzymes, occurs. VOO polyphenols depend from the ripening stage of olives at processing (Anastasopoulos et al., 2011; Baccouri et al., 2007; Beltran et al., 2005; Conde et al., 2008; Gambacorta et al., 2010; Salvador et al., 2001; Skevin et al., 2003; Youssef et al., 2010).

Milling/Crushing: During crushing, the main glycosides (oleuropein, demethyloleuropein, ligstroside) present in the fruit are hydrolysed by action of endogenous β-glucosidases leading to the formation of secoiridoid aglycons. Mechanical (hammer or disk) crushers are more effective in the extraction of phenolic compounds than traditional granite millstones (Angerosa et al., 1995; Caponio et al., 2003; Servili et al., 2004; Vichi et al., 2011).

Stones removal: Oil extraction after removal of stones significantly increase VOO phenolic concentration in the oil. In fact, stones have a high concentration of β-glucosidases and their release during crushing promote phenol compounds hydrolysis (Servili et al., 2007; Lavelli and Bondesan, 2005).

Malaxation: The effect of temperature and time of malaxation on qualitative and quantitative composition of VOO polyphenols is contrasting. Several authors found a negative relationship between malaxation temperature and total phenols concentration whereas others found more hydrophylic polyphenols in VOO when the temperature was increased. Anyway, the application of new techniques able to control oxidative reactions, such as the use of a saturated nitrogen athmosphere during malaxation may be useful (Amirante et al., 2006; Angerosa et al., 2001; Artajo et al., 2007; El Riachy et al., 2011; Gomez-Rico et al., 2007; Kalua et al., 2006; Parenti et al., 2008; Ranalli et al., 2001; Servili et al., 2003; Servili et al., 2004; Servili et al., 2008).

Extraction: The quantity of water added to the paste before oil separation markedly affects VOO phenol composition. Water addition causes the elimination of hydrophilic phenol compounds in the vegetation waste waters. The modern equipments such as the two- and three-phases centrifugation systems, characterized by short processing time and no or low water addition, respectively, guarantee a higher retention of polyphenols in the VOO than the traditional pressure and old three-phase systems (De Stefano et al., 1999; Di Giovacchino et al., 1994; Di Giovacchino et al., 2002; El Riachy et al., 2011; Servili et al., 2003; Servili et al., 2004).

Filtration: The enzyme activity in the cloudy phase which is a characteristic feature of fresh VOO leads to a decrease in the phenolic composition of the VOO, mainly of secoiridoids aglycons. The filtration, removing the residual water and solids in suspension, blocks this enzymatic degradation (Lozano Sanchez et al., 2010).

Storage: During storage VOO polyphenols undergo qualitative and quantitative modifications due to hydrolytic decomposition and oxidation reaction. The total phenols content decreases and, consequently, the intensity of the typical bitter taste and pungent note of fresh VOO is markedly reduced. Temperature, light, packaging and, at minor extent, the storage time, are the main factors affecting the phenol composition of aged oils (Cinquanta et al., 1997; Sinesio et al., 2005; Sacchi et al., 2008).

Thus, summarizing, the main phenols in fresh made VOO are aglycones from secoiridoids and lignans, with only low amount of not bitter phenyl alcohols. These latter compounds, if present in the olives, partitioned preferentially into the water phase during the milling and centrifugation process being especially found in less acceptable unfiltered cloudy oils or they may form in VOO during storage from hydrolysis of secoiridoids and together with other unwanted oxidation products. Using centrifugal decanters not involving abundant process water may be useful to obtain higher amount of tyrosol and hydroxytyrosol in VOO, however, this technology also preserve losses of oleuropein aglycons. Thus, finding an equilibrium between secoiridoid aglycones and phenyl alcohol in VOO that is towards the latter is not a technologically easy way to run to obtain polyphenol rich VOO.

All in all these facts sustain the consideration that having natural (avoiding external enrichment) polyphenol rich VOO, inevitably means having secoiridoid aglycones rich VOO and, as consequence, bitter VOO.

Sensory markers of VOO phenol compounds

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The chemist's point of view

The content and the composition of polyphenols in VOO are strictly related to the sensory attributes of "bitterness" and "pungency" (also defined as "throat irritating" sensation).

A good correlation between the intensity of these attributes perceived by a panel of expert tasters and the overall content of phenol compounds has been reported by many studies (Gutierrez-Rosales et al.,1992, Gutierrez et al., 2000; Gutierrez-Rosales et al., 2003; Tsimidou, 1998; Skevin et al., 2003; Inarejos-Garcia et al., 2009; Fogliano and Sacchi, 2006; Bendini et al., 2007; Cerretani et al., 2008). According to recent studies, particularly some complex phenols contribute more than others to the taste perception of bitterness and pungency. However, the precise identification of compounds responsible for each perception cannot be predicted by the molecular structure of the compound as it depends on the molecular size, functional group, sugar position, decrease of hydrophilicity and stereochemistry of the molecule. Anymore a precise identification is complicated by the interaction of phenol compounds with other compounds that simultaneously stimulate similar perceptions such as certain C5 and C6 volatile compounds (e.g. (E)-2-hexenal, hexanol) (Morales et al., 1996; Caporale et al., 2004; Kalua et al., 2008).

According to the current knowledge about chemical composition of VOO and sensory perception it is clear that:

- A good correlation between bitterness and the aldehydic form of oleuropein aglycone exists as evidenced by linear regression coefficients (~0.6) found by several authors (Mateos et al., 2004; Siliani et al., 2006; Sinesio et al., 2005; Garcia et al.,2001);
- The intensity of sensory perception of pungency is mainly related to the content of the deacetoxy form of p-HPEA-EDA, the correlation concentration-pungency perception

showing a linear regression coefficient of ~0.5 (Andrewes et al., 2003, Beauchamp et al., 2005; Fogliano and Sacchi, 2006; Smith et al., 2005);

- The green odour note of the fruity can have a positive significant effect on the perception of bitterness: the presence of cut grass odour enhancing it (Caporale et al., 2004);
- The composition of the lipid matrix can play a key role in the perception of bitterness in VOO. In fact, it has been shown that pungency and bitterness were more pronounced in monounsaturated matrices than in polyunsaturated ones (Garcia-Mesa et al., 2008).

The physiologist's point of view

The most abundant polyphenols in VOO are bitter and pungent. The perception of bitterness is due to activation of about twenty-five 7-transmembrane receptors (the T2Rs) and trigeminal nerve endings, located within the oral, sensitive to chemical stimuli (Bachmanov and Beauchamp, 2007). Recent evidence indicated a genetic variation between populations in sensitivity to bitter taste with a frequency of non-tasters that can vary from as low as 7% to 40% (Nabhan, 2004). Surprisingly the so-called taste receptors were identified, not only in the mouth but also in the gut, the pancreas and the brain thus suggesting that they may play a role in nutrient recognition and regulation (Margolskee et al., 2007; Sclafani 2007; Behrens and Meyerhof, 2011).

On the other hand liking for taste stimuli is generally strongly influenced by inborn (innate) factors (Bartoshuk and Beauchamp, 1994). Sweet foods are innately preferred by all animals, presumably since sweetness reflects the presence of caloric sugars in plants. On the contrary bitter taste signals the presence of potentially toxic compounds and hence bitter substances are

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generally disliked (Glendinning, 1994). From an evolutionary perspective a sensitive perception of bitter taste was strictly linked to survival during Palaeolithic: the hunter gathered men able to perceive bitterness were also able to avoid dangerous foods. On the other hand, also several compounds with antimicrobial activity have a bitter taste (as many spices), and for populations living in regions where malaria was endemic the insensitivity to this taste allowed survival. Thus, looking at the human history, the different grade of bitter perceptions seems strictly co-evolved with society habits and environment (Krebs, 2009).

Generally infants or children have more preferences than adults and these preferences can be modified by experience (for review, see Cowart et al., 2004). Interestingly prenatal experiences with food flavours transmitted from the mother's diet to amniotic fluid lead to greater acceptance and enjoyment of these foods during weaning. Anyway, flavour learning continues after birth mainly regulated by lactating modality (breast feeding or infant formula). In particular, it has been demonstrated that there is a sensitive learning period in the first months of life during which unpalatable flavours (to those not familiar with them) can become palatable (Bechaump and Mennella, 2011). Thus both individual experience and social environment highly influence food liking and, as consequence, dietary choice. An intriguing perspective comes from the observation that many of the preferred bitter foods such as coffee or tea, are also psychoactive (because of caffeine presence) and have healthy properties. The consumption of these foods was beneficial for humans so people somehow may have needed to learn to like them. In the same way, with the experience also bitter vegetables can turn to be accepted, although, whether the bitterness of these is actually liked or whether the other flavor characteristics are sufficient to overcome the dislike of bitterness, is not known as well (Forestell and Mennella, 2007; Mennella et al., 2009).

The consumer's point of view

In the framework described in the above paragraphs it is not surprising that many consumers negatively judges very bitter and pungent oils and that culture may even influence consumer preference for VOO (Jimenez-Guerrero et al., 2012). Bitterness and pungency sensations can persist for long time after deglutition, showing a clear after-taste that can strongly vary in intensity and duration depending from olive oils and from individual sensitivity (Esti et al., 2009; Caporale et al., 2006; Servili et al., 2009). Cicerale and co-workers (2009) have recently demonstrated that the irritation elicited by p-HPEA-EDA (oleocanthal) is mainly localized to the oropharynx, peaks at 15 s and lasts over 180 s post-exposure. Anymore they showed that oleocanthal irritation is more variable among individuals compared with the irritation elicited by CO_2 and the sweetness of sucrose, thus suggesting the existence of independent mechanisms underlying the taste by this molecule and justifying the high inter-individual variability associated to its anti-inflammatory effect (Bechaump et al., 2005).

The fact that most consumers dislike very bitter and pungent oils, highly limits the placement on the market of VOO having concentrations of phenolic acids higher than 200 mg/L (Fogliano and Sacchi, 2006). Nowadays bitter and pungent VOOs are considered niche products for gourmet estimators who ask for these specific sensory characteristics and are ready to pay a premium price for them. On the other hand producing polyphenol-rich VOO by increasing the concentration of less bitter phenyl alcohols (hydroxytirosol and tyrosol) though modulation of VOO storage time would reduce health benefits since storage is often accompanied by difficult to control lipid oxidative reactions ad since seicoridoid aglycons possesses biological properties that phenyl alcohols do not have.

Finally the addition of any kind of antioxidants or other ingredients to VOO should be ruled out because in this case according to the legislation the product cannot be named VOO.

Summary and future perspectives

Although VOO is the pillar fat of Mediterranean diet there is a large amount of people living in Mediterranean countries who does not consume it and prefers refined olive oil or even seeds' oils (Delgado and Guinard, 2011). The main reasons of this choice are both sensory and economic. The bitter and pungent taste of VOO is not preferred by most consumers and it is wrongly associated to a low quality product. From the chemical, technological and nutritional point of view, sweet VOOs have a low content of polyphenols and, as consequence, are low quality products. Thus a clear gap of knowledge exists between consumer perception of VOO quality and chemical/technological/nutritional quality. On the other hand although ways to naturally increase less bitter polyphenols in VOO are possible (through genetic improvement strategies, field selection of olive tree varieties, reducing water addition during process or using centrifugal decanter) are not easy to run and, even, not convenient from both economic and healthy point of view. Indeed, the disliking of bitterness and pungency by most people is due to genetic and physiological reasons, but humans can be educated to new tastes. The habit to a specific taste is a fundamental driver for liking it. Consumer choice is mainly driven by price, and unfortunately, VOO market price usually parallels its polyphenols content. In this respect, it is likely that market has also contributed to the taste preference of most people for "sweet" and "cheap" VOO.

The recent substantiation by EFSA of the health claim for VOO to protect blood lipids from oxidative damage (EFSA Journal 2011), as indicated by several scientific papers previously published (Covas, 2008; Frankel, 2011; Covas et al., 2006; Raederstorff et al., 2009; Castañer et al., 2011), may activate multilevel positive effects as depicted in **Figure 3**. Starting from marketing, producers aiming to obtain the health claim will need to modify the processing (choice of raw material and technology) to have VOO with a polyphenols concentration complying with EFSA directive. It can be expected that many commercial VOO will be marketed with the minimum content of phenol compounds necessary to get the health claim. This fact will get, in turn, to a sensory standardization of VOO towards more bitter and pungent VOO than those actually present on the market. The presence of the health claim on VOO having specific sensory characteristic, will represent a practice communication strategy: consumers will learn that bitter and pungent VOO are healthy products. As a consequence, it may increase the awareness of consumers on health benefits of VOO compared to other vegetable oils thus increasing their perception of VOO convenience and, consequently, its dietary intake. In the short period it can be expected that VOO market will go towards an increased consumer demand for a high quality product and the possibility to have a more advantageous quality/price ratio.

EU and National Authority should support this desirable process both subsidizing VOO producers to increase the content of polyphenols and promoting educational interventions. Consumers acceptance of most bitter and pungent VOO, will increase their liking for this taste and allow the general population to get the VOO polyphenols health benefits.

References

- Ahmed, C.B., Rouina, B.B., Sensoy, S., and Boukhriss, M. (2009). Saline water effects on fruit development, quality and phenolic composition of virgin olive oils cv Chemlali. *J. Agric. Food Chem*. **57**: 2803-2811.
- Amirante, P., Clodoveo M.L., Dugo G., Leone, A. and Tamborrino, A. (2006). Advance technology in virgin olive oil production from traditional and de-stoned pastes: Influence of the introduction of a heat exchanger on oil quality. *Food Chemistry*. **98** (4): 797-805.
- Anastasopoulos, E., Kalogeropoulos, N., Kaliora, A.C., Kountouri, A., and Andrikopoulos, N.K. (2011). The influence of ripening and crop year on quality indices, polyphenols, terpenic acids, squalene, fatty acid profile, and sterols in virgin olive oil (Koroneiki cv.) produced by organic versus non-organic cultivation method. *International Journal of Food Science and Technology* **46**: 170–178.
- Andrewes, P., Busch, J. L. H. C., de Joode, T., Groenewegen, A., and Alexandre, H. (2003). Sensory properties of virgin olive oil polyphenols: identification of deacetoxy-ligstroside aglycon as a key contributor to pungency. *J. Agric. Food Chem.* **51**: 1415-1420.
- Angerosa, F., and Di Giacinto, L. (1995). Caratteristiche di qualità dell'olio di oliva vergine in relazione ai metodi di frangitura. Nota II. *Riv. Ital. Sost. Grasse* **72**: 1–4.
- Angerosa, F., Mostallino, R., Basti, C., and Vito, R. (2001). Influence of malaxation temperature and time on the quality of virgin olive oil. *Food Chemistry* **72**: 19-28.
- Arranz, S., Silván, J.M., and Saura-Calixto, F. (2010). Nonextractable polyphenols, usually ignored, are the major part of dietary polyphenols: a study on the Spanish diet. *Mol Nutr Food Res.* **54**(11): 1646-1658.

- Artajo, L.S., Romero, M.P., Suárez, M., and Motilva, M.J. (2007). Partition of phenolic compounds during the virgin olive oil industrial extraction process. *European Food Research and Technology A*. (5-6): 617-625.
- Baccouri, B., Zarrouk, W., Khichene, D., Nouari, I., Ben Youssef, N., and Daoud, D. (2007). Influence of fruit ripening and crop yield on chemical properties of virgin olive oils from seven selected oleasters (Olea europea L.). *J Agron* **6**: 388–396.
- Bachmanov, A.A., and Beauchamp, G.K. (2007). Taste receptor genes. *Annu Rev Nutr* **27**: 389–414.
- Bartoshuk, L.M., and Beauchamp, G.K. (1994). Chemical senses. *Annu Rev Psychol* **45**: 419–449.
- Beauchamp, G. J., Keast, R. S. J., Morel, D., Lin, J., Pika, J., Han, Q., Lee, C.H., Smith, A. B., and Breslin, P. A. S. (2005). Ibuprofen-like activity in extra-virgin olive oil. *Nature* **437**: 45-46.
- Beauchamp, G.K., and Mennella, J.A. (2011). Flavor perception in human infants: development and functional significance. Digestion. **83**: 1-6.
- Behrens, M., and Meyerhof ,W. (2011). Gustatory and extragustatory functions of mammalian taste receptors. *Physiol Behav*. **105**(1): 4-13.
- Beltran, G., Aguilera, M.P., Del Rio, C., Sanchez, S., and Martinez, L. (2005). Influence of fruit ripening process on the natural antioxidant content of Hojiblanca virgin olive oils. *Food Chem* **89**: 207–215.
- Bendini, A., Cerretani, L., Carrasco-Pancorbo, A., Gómez-Caravaca, A. M., Segura-Carretero, A., Fernández-Gutiérrez, A., and Lercker, G. (2007). Phenolic molecules in virgin olive oils: a

survey of their sensory properties, health effects, antioxidant activity and analytical methods. An overview of the last decade. *Molecules* **12**: 1679–1719.

- Bucelli, P., Costantini, E. A. C. Barbetti, R., and Franchini, E. (2011). Soil Water Availability in Rainfed Cultivation Affects More than Cultivar Some Nutraceutical Components and the Sensory Profile of Virgin Olive Oil. *J. Agric. Food Chem* **59**(15): 8304-8313
- Caponio, F., Gomes, T., Summo, C., and Pasqualone, A. (2003). Influence of the type of olive-crusher used on the quality of extra virgin olive oils. *Eur. J. Lipid Sci. Technol*. **105**: 201–206.
- Caporale, G., Policastro, S., and Monteleone, E. (2004).Bitterness enhancement induced by cut grass odorant (cis-3-hexen-1-ol) in a model olive oil. *Food Quality and Preference* **15**: 219–227.
- Caporale, G., Policastro, S., Carlucci, A., and Monteleone, E. (2006). Consumer expectations for sensory properties in virgin olive oils. *Food Quality and Preference*. **17**(1–2): 116–125.
- Caruso, D., Visioli, F., Patelli, R., Galli, C., and Galli, G. (2001). Urinary excretion of olive oil phenols and their metabolites in humans. *Metab. Clin. Exp.* **50**: 1426-1428.
- Castañer, O., Fitó, M., López-Sabater, M.C., Poulsen, H.E., Nyyssönen, K., Schröder, H., Salonen, J.T., De la Torre-Carbot, K., Zunft, H.F., De la Torre, R., Bäumler, H., Gaddi, A.V., Saez, G.T., Tomás, M., Covas, M.I. and EUROLIVE Study Group. (2011). The effect of olive oil polyphenols on antibodies against oxidized LDL. A randomized clinical trial. *Clin Nutr*. 30(4): 490-493.

- Cerretani, L., Salvador, M. D., Bendini, A., and Fregapane, G. (2008). Relationship between sensory evaluation performed by Italian and Spanish official panels and volatile and phenolic profiles of virgin olive oils. *Chemosensory Perception*. **1**(4): 258-267.
- Cicerale, S., Lucas, L., and Keast, R. (2010). Biological activities of phenolic compounds present in virgin olive oil. *Int J Mol Sci*. **11**(2): 458-479.
- Cicerale, S., Lucas, L., and Keast, R. (2011). Antimicrobial, antioxidant and anti-inflammatory phenolic activities in extra virgin olive oil. *Curr Opin Biotechnol.*
- Cinquanta, L., Esti, M., and La Notte, E. (1997). Evolution of Phenolic Compounds in Virgin Olive Oil During Storage. *JAOCS*. **74**(10): 1259-1264.
- Conde, C., Delrot, S., and Geròs, H. (2008). Physiological, biochemical and molecular changes occurring during olive development and ripening. *Journal of Plant Physiology* **165**: 1545-1562.
- Corona, G., Spencer, J. P. E., and Dessi, M. A. (2009). Extra virgin olive oil phenolics: absorption, metabolism, and biological activities in the GI tract. *Toxicol. Ind. Health.* **25**: 285-293.
- Covas, M.I, Konstantinidou, V., and Fitó, M. (2009). Olive oil and cardiovascular health. *J Cardiovasc Pharmacol*. **54**(6): 477-482.
- Covas, M.I. (2007). Olive oil and the cardiovascular system. *Pharmacological Research* **55**: 175–186.
- Covas, M.I. (2008). Bioactive effects of olive oil phenolic compounds in humans: reduction of heart disease factors and oxidative damage. *Inflammopharmacology*. **16**(5): 216-218.

- Covas, M.I., Nyyssönen, K., Poulsen, H.E., Kaikkonen, J., Zunft, H.J., Kiesewetter, H., Gaddi, A., de la Torre, R., Mursu, J., Bäumler, H., Nascetti, S., Salonen, J.T., Fitó, M., Virtanen, J., Marrugat, J., and EUROLIVE Study Group. (2006). The effect of polyphenols in olive oil on heart disease risk factors: a randomized trial. *Ann Intern Med.* **145**(5): 333-341.
- Covas, M.I., Ruiz-Gutierrez, V., de la Torre, R., Kafatos, A., Lamuela-Raventos, R.M., Osada, J., Owen, R.W., and Visioli, F. (2006). Minor Components of Olive Oil: Evidence to Date of Health Benefits in Humans. *Nutrition Reviews.* **64**(10): S20-S30.
- Cowart, B.J., Beauchamp, G.K., Mennella, J.A. (2004). Development of taste and smell in the neonate. **In** : Fetal and Neonatal Physiology, pp. 1819–1827. Polin, R.A., Fox, W.W., and Abman, S.H., Eds., Saunders, Philadelphia.
- D'Angelo, S., Manna, C., Migliardi, V., Mazzoni, O., Morrica, P., Capasso, G., Pontoni, G., Galletti, P., and Zappia, V. (2001). Pharmacokinetics and metabolism of hydroxytyrosol, a natural antioxidant from olive oil. *Drug. Metab. Dispos.* **29**: 1492-1498.
- Dabbou, S ., Dabbou, S., Selvaggini, R., Urbani, S., Taticchi, A., Servili, M., and Hammami, M. (2011). Comparison of the Chemical Composition and the Organoleptic Profile of Virgin Olive Oil from Two Wild and Two Cultivated Tunisian Olea europaea. *Chemistry & Biodiversity* **8**: 191-202.
- De la Torre, K., Jauregui, O., Gimeno, E., Castellote, A.I., Lamuela-Raventós, R.M., and López-Sabater, M.C. (2005). Characterization and quantification of phenolic compounds in olive oils by solid-phase extraction, HPLC-DAD, and HPLC-MS/MS. *J Agric Food Chem*. **53**: 4331–4340.

- De la Torre, R. (2008). Bioavailability of Olive Oil Phenolic Compounds in Humans. *Inflammopharmacology*. **16**: 1–3.
- de la Torre-Carbot, K., Chavez-Servın, J.L., Jauregui, O., Castellote, A.I., Lamuela-Raventos, R.M., Fito, M., Covas, M.I., Munoz-Aguayo, D., and Lopez-Sabater, M.C. (2007). Presence of virgin olive oil phenolic metabolites in human low density lipoprotein fraction: Determination by high-performance liquid chromatography–electrospray ionization tandem mass spectrometry. *Analytica Chimica Acta* **583**: 402–410.
- de la Torre-Carbot, K., Chávez-Servín, J.L., Jaúregui, O., Castellote, A.I., Lamuela-Raventós, R.M., Nurmi, T., Poulsen, H.E., Gaddi, A.V., Kaikkonen, J., Zunft, H.F., Kiesewetter, H., Fitó, M., Covas, M.I., and López-Sabater, M.C. (2010). Elevated circulating LDL phenol levels in men who consumed virgin rather than refined olive oil are associated with less oxidation of plasma LDL. *J Nutr.* **140**(3): 501-508.
- Delgado, C., and Guinard, J.X. (2011) How do consumer hedonic ratings for extra virgin olive oil relate to quality ratings by experts and descriptive analysis ratings? *Food Quality and Preference* **22** (2): 213-225.
- Dell'Agli, M., Fagnani, R., Galli, G.V., Maschi, O., Gilardi, F., Bellosta, S., Crestani, M., Bosisio, E., De Fabiani, E., and Caruso, D. (2010). Olive oil phenols modulate the expression of metalloproteinase 9 in THP-1 cells by acting on nuclear factor-kB signaling. *J Agric Food Chem* **58**: 2246-2252.
- De Stefano, G., Piaquadio, P., Servili, M., Di Giovacchino, L., and Sciancalepore, V. (1999). Effect of extraction system on thephenolic composition of virgin olive oils. *Fat/Lipids* **101**: 328–332.

- Di Giovacchino, L., Sestili, S., and Di Vincenzo, D. *(*2002*).* Influence of olive processing on virgin olive oil quality. *European Journal of Lipid Science and Technology* **104***:* 587–601.
- Di Giovacchino, L., Solinas, M., and Miccoli, M. (1994). Effect of extraction systems on the quality of virgin olive oil. *J. Am. Oil Chem. Soc.* **71**: 1189–1194.
- EFSA Journal (2011). **9**(4): 2033-2057.
- El Riachy, M., Priego-Capote, F., Leon, L., Rallo, L., and de Castro, M.D.L. (2011). Review Article. Hydrophilic antioxidants of virgin olive oil. Part 2: Biosynthesis and biotransformation of phenolic compounds in virgin olive oil as affected by agronomic and processing factors. *Eur. J. Lipid Sci. Technol*. **113**: 692–707.
- Esti, M., Contini, M., Moneta, E., and Sinesio, F. (2009). Phenolics compounds and temporal perception of bitterness and pungency in extra-virgin olive oils: Changes occurring throughout storage. *Food Chemistry* **113**: 1095–1100
- Evangelisti, F., Zunin, P., Calcagno, C., Tiscorina, E., and Petacchi, R. (1994). Dacus oleae infestation and its consequences on the phenolic compounds of virgin olive oil. *Riv. It. Sost. Grasse.* **74**: 507–511.
- Fini, L., Hotchkiss, E., Fogliano, V., Graziani, G., Romano, M., De Vol, E.B., Qin, H., Selgrad, M., Boland, C.R., and Ricciardiello, L. (2008). Chemopreventive properties of pinoresinol-rich olive oil involve a selective activation of the ATM-p53 cascade in colon cancer cell lines. *Carcinogenesis*. **29**(1): 139-46.
- Fito, M., de la Torre, R., and Covas, M.I. (2007). Olive oil and oxidative stress. *Mol. Nutr. Food Res.* **51**: 1215 – 1224.

- Fogliano , V., and Sacchi, R. (2006) Oleocanthal in olive oil: between myth and reality. *Mol. Nutr. Food Res.* **50**: 5-6.
- Forestell, C.A., and Mennella, J.A. (2007). Early determinants of fruit and vegetable acceptance. *Pediatrics* **120**: 1247–1254.
- Frankel E.N. (2011). Nutritional and biological properties of extra virgin olive oil. *J Agric Food Chem.* **59**(3): 785-792.
- Galvano, F., La Fauci, L., Graziani, G., Ferracane, R., Masella, R., Di Giacomo, C., Scacco, A., D'Archivio, M., Vanella, L., and Galvano, G. (2007). Phenolic compounds and antioxidant activity of Italian extra virgin olive oil Monti Iblei. *J Med Food.* **10**(4): 650-656.
- Gambacorta, G., Faccia, M. , Previtali, M.A., Pati, S., La Notte, E., and Baiano, A. (2010). Effects of Olive Maturation and Stoning on Quality Indices and Antioxidant Content of Extra Virgin Oils (cv. Coratina) during Storage. *Journal Of Food Science*. **75**(3):C229-235.
- Garcia, J. M., Yousfi, K., Mateos, R., Olmo, M., and Cert, A. (2001). Reduction of oil bitterness by heating of olive (Olea europaea) fruits. *J. Agric. Food Chem*. **49**: 4231-4235.
- Garcia-Mesa, J.A., Pereira-Caro, G., Fernandez-Hernandez, A., Garcia-Ortiz Civantos, C., and Mateos, R. (2008). Influence of lipid matrix in the bitterness perception of virgin olive oil. *Food Quality and Preference*. **19**: 421–430.
- García-Villalba, R., Carrasco-Pancorbo, A., Nevedomskaya, E., Mayboroda, O.A., Deelder, A.M., Segura-Carretero, A., and Fernández-Gutiérrez, A. (2010). Exploratory analysis of human urine by LC-ESI-TOF MS after high intake of olive oil: understanding the metabolism of polyphenols. Anal Bioanal Chem. **398**(1): 463-475.

- Glendinning, J.I. (1994). Is the bitter rejection response always adaptive? *Physiol Behav* **56**: 1217–1227.
- Gòmez-Caravaca, A.M., Cerretani, L., Bendini, A., Segura-Carretero, A., Fernàndez-Gutiérrez, A., Del Carlo, M., Compagnone, D., and Cichelli, A. (2008). Effects of Fly Attack (Bactrocera oleae) on the Phenolic Profile and Selected Chemical Parameters of Olive Oil. *J. Agric. Food Chem*. **56**: 4577–4583.
- Gomez-Rico, A., Salvador, M. D., Moriana, A., and Perez, D. (2007). Influence of different irrigation strategies in a traditional Cornicabra cv. olive orchard on virgin olive oil composition and quality. *Food Chem.* **100**: 568–578.
- Gutierrez, F., Albi, M. A., Palma, R., Rios, J. J., and Olias, J. M. (2000). Bitter taste of virgin olive oil: correlation of sensory evaluation and instrumental HPLC analysis. *J. Food Sci.* **54**: 68-70.
- Gutierrez-Rosales, F., Perdiguero, S., Gutierrez, R., and Olias, J.M. (1992). Evaluation of the Bitter Taste in Virgin Olive Oil. *JAOCS* **69**(4)
- Gutierrez-Rosales, F., Rios, J. J., and Gomez-Rey, M. L. (2003). Main polyphenols in the bitter taste of virgin olive oil. Structural confirmation by on-line high-performance liquid chromatography electrospray ionization mass spectrometry. *J. Agric. Food Chem*. **51**: 6021-6025.
- Hardin-Fanning, F. (2008). The Effects of a Mediterranean-Style Dietary Pattern on Cardiovascular Disease Risk. *Nurs Clin N Am.* **43**: 105–115.

- Inarejos-Garcia, A.M., Androulaki, A., Desamparados Salvador, M., Fregapane, G., and Tsimidou, M.Z. (2009). Discussion on the objective evaluation of virgin olive oil bitterness. *Food Research International*. **42**: 279–284
- Jiménez-Guerrero, J.F., Gázquez-Abad, J.C., Mondéjar-Jiménez, J.A. and Huertas-García, R. (2012). Consumer Preferences for Olive-Oil Attributes: A Review of the Empirical Literature Using a Conjoint Approach. **In** : Olive Oil – Constituents, Quality, Health Properties and Bioconversions, pp. 233-243. Boskou, D., Eds, InTech.
- Kalua CM, Bedgood DR Jr, Bishop AG, and Prenzler PD. Changes in virgin olive oil quality during low-temperature fruit storage. J Agric Food Chem. 2008 Apr 9;56(7):2415-22
- Kalua, C.M., Bedgood, D.R., Bishop, A.G., and Prenzler, P.D. (2006). Changes in volatile and phenolic compounds with malaxation time and temperature during virgin olive oil production. *J. Agric. Food Chem.* 54: 7641-7651.
- Khanal, P., Oh, W.K., Yun, H.J., Namgoong, G.M., Ahn, S.G., Kwon, S.M., Choi, H.K., and Choi, H.S. (2011) p-HPEA-EDA, a phenolic compound of virgin olive oil, activates AMP-activated protein kinase to inhibit carcinogenesis. *Carcinogenesis* **32**: 545-553.
- Krebs, J.R. (2009). The gourmet ape: evolution and human food preferences. *Am J Clin Nutr* **90**: 707S–711S
- Lavelli V, and Bondesan L. Secoiridoids, tocopherols, and antioxidant activity of monovarietal extra virgin olive oils extracted from destoned fruits. J Agric Food Chem. 2005 Feb 23;53(4):1102-7.
- Lozano-Sànchez, J., Cerretani, L., Bendini, A., Segura-Carretero, A., and Fernàndez-Gutiàrrez, A. (2010). Filtration process of extra virgin olive oil: effect on minor components,

oxidative stability and sensorial and physicochemical Characteristics. *Trends in Food Science & Technology* **21**: 201-211.

- Manach, C., Scalbert, A., Morand, C., Remesy, C., and Jimenez, L. (2004). Polyphenols: food sources and bioavailability. *Am J Clin Nutr*.**79**:727–747.
- Margolskee, R.F., Dyer, J., Kokrashvili, Z., Salmon, K.S., Ilegems, E., and Daly, K. (2007). T1R3 and gustducin in gut sense sugars to regulate expression of Na + -glucose cotransporter 1. *Proc Natl Acad Sci USA.* **104**: 15075– 15080.
- Mateos, R., Cert, A., Perez-Camino, M. C., and Garcia, J.M. (2004). Evaluation of virgin olive oil bitterness by quantification of secoiridoid derivatives. *J. Am. Oil Chem. Soc.* **81**: 71-75.
- Mattila, P., Pihlava, J.-M., and Hellstrom, J. (2005). Contents of phenolic acids, alkyland alkenylresorcinols, and avenanthramides in commercial grain products. J *Agric Food Chem*. **53**: 8290–8295.
- Mennella, J.A., Forestell, C.F., Morgan, L., and Beauchamp, G.K. (2009). Early milk feeding influences taste acceptance and liking during infancy. *Am J Clin Nutr* **90**: 780S–788S
- Miró-Casas, E., Covas, M.I., Farre, M., Fito, M., Ortuño, J., Weinbrenner, T., Roset, P., and de la Torre, R. (2003a). Hydroxytyrosol disposition in humans. Clin Chem. **49**: 945-52.
- Miró-Casas, E., Covas, M.I., Fito, M., Farré-Albadalejo, M., Marrugat, J., and de la Torre, R. (2003b). Tyrosol and hydroxytyrosol are absorbed from moderate and sustained doses of virgin olive oil in humans. *Eur. J. Clin. Nutr.* **57**: 186-190.
- Miró-Casas, E., Farré Albaladejo, M., Covas, M.I., Rodriguez, J.O., Menoyo Colomer, E., Lamuela Raventós, R.M., and de la Torre, R. (2001). Capillary gas chromatography-mass

spectrometry quantitative determination of hydroxytyrosol and tyrosol in human urine after olive oil intake. *Anal Biochem.* **294**(1): 63-72.

- Montedoro, G, Servili, M., Baldioli, M., Selvaggini, R., Miniati, E., and Macchioni A. (1993). Simple and hydrolyzable compounds in virgin olive oil. 3. Spectroscopic characterizations of the secoiridoid derivatives. *J. Agric. Food Chem.* **41**: 2228–2234.
- Montedoro, G.F., Garofalo, L, and Sensidoni, A. (1985). Infestazione di olive da Dacus oleae e caratteristiche qualitative degli oli vergini. *Riv. It. Sost. Grasse*. **62**: 565–567.
- Morales, M.T, Cálvente, J.J., and Aparicio, R. (1996) «Influence of olive ripeness on the concentration of green aroma compounds in virgin olive oil».—Flavour Fragr. J.. 11,171-178
- Motilva, M. J., Romero, M. P., Alegre, S., and Girona, J. (1999). Effect of regulated deficit irrigation in olive oil production and quality. *Acta Hort.* **474**: 377–380.
- Motilva, M.J., Tovar, J.M., Romero, P.M., Alegre, S., and Girona, J. (2000). Influence of regulated deficit irrigation strategies applied to olive trees (Arbequina cultivar) on oil yield and oil composition during the fruit ripening period. *Journal of the Science of Food and Agriculture* **80**: 2037–2043.
- Nabhan, G.P. (2004). Why some like it hot. Washington, Island Press, DC.
- Ovaskainen, M.L., Torronen, R., Koponen, J.M., Sinkko, H., Hellstrom, S., Reinivuo, H., and Mattila, P. (2008). Dietary Intake and Major Food Sources of Polyphenols in Finnish Adults. *J. Nutr.* **138**: 562–566.
- Owen, R.W., Mier, W., Giacosa, A., Hule, W.E., Spiegelhalder, B., and Bartsch, H. (2000). Phenolic compounds and squalene in olive oils: the concentration and antioxidant potential of

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total phenols, simple phenols, secoroids, lignans and squalene. *Food Chem Toxicol*. **38**: 647–659.

- Parenti, A., Spugnoli, P., Masella, P., and Calamai, L. (2008). The effect of malaxation temperature on the virgin olive oil phenolic profile under laboratory-scale conditions. *Eur. J. Lipid Sci. Technol*. **110**: 735–741.
- Patumi, M., d'Andria, R., Marsilio, G., and Fontanazza, G. (2002). Olive and olive oil quality after intensive monocone olive growing (Olea europaea L., cv. Kalamata) in different irrigation regimes. *Food Chem.* **77**: 27–34.
- Pereira, J. A., Alves, M. R., Casal, S., and Oliveira, M.B.P.P. (2004). Effect of olive fruit fly infestation on the quality of olive oil from cultivars. Cobrancosa, Madural and Verdeal Transmontana. *Ital. J. Food Sci*. **3**: 355–365.
- Radtke, J., Linseisen, J., and Wolfram, G. (1998). [Phenolsäurezufuhr Erwachsener in einem bayerischen Teilkollektiv der Nationalen Verzehrsstudie] Phenolic acid intake of adults in a Bavarian subgroup of the national food consumption survey. *Z Ernahrungswiss.* **37**: 190–197.
- Raederstorff D. (2009). Antioxidant activity of olive polyphenols in humans: a review. *Int J Vitam Nutr Res.* **79**(3): 152-165.
- Ranalli, A., Contento, S., Schiavone, C., and Simone, N. (2001). Malaxing temperature affects volatile and phenol composition as well as other analytical features of virgin olive oil. *Eur. J. Lipid Sci. Technol*. **103**: 228-238.
- Romero, C., Medina, E., Vargas, J., Brenes, M., and De Castro, A. (2007). In vitro activity of olive oil polyphenols against Helicobacter pylori. *J Agric Food Chem.* **55**: 680-686.

- Rotondi, A., Alfei, B., Magli, M., and Pannelli, G. (2010). Influence of genetic matrix and crop year on chemical and sensory profiles of Italian monovarietal extra-virgin olive oils. *J Sci Food Agric* **90**: 2641–2648.
- Sacchi, R., Savarese, M., Del Regno, A., Paduano, A., Terminiello, R., and Ambrosino, M.L. (2008) Shelf-life of vegetable oils bottled in different polyethylenterephthalate (PET) containers. *Packaging Technology and Science*. **21**: 269-277.
- Salvador, M.D., Aranda, F., and Fregapane, G. (2001). Influence of fruit ripening on 'Cornicabra' virgin olive oil quality – A study of four successive crop seasons. *Food Chemistry* **73**: 45-53.
- Sangiovanni, E., Colombo, E., Fumagalli, M., Abbiati, F., Caruso, D., and Dell'agli, M. (2012). Inhibition of NF- κB Activity by Minor Polar Components of Extra-Virgin Olive Oil at Gastric Level. *Phytother Res*. doi: 10.1002/ptr.4600.
- Saura-Calixto, F., Serrano, J., and Goni, I. (2007). Intake and bioaccessibility of total polyphenols in a whole diet. *Food Chem.***101**:492–501.
- Sclafani, A. (2007). Sweet taste signaling in the gut. *Proc Natl Acad Sci USA* **104**: 14887–14888.
- Servili, M., Esposto, S., Fabiani, R., Urbani, S., Taticchi, A., Mariucci, F., Selvaggini, R., and Montedoro, G.F. (2009). Phenolic compounds in olive oil: antioxidant, health and organoleptic activities according to their chemical structure. *Inflammopharmacology*. **17**: 76–80
- Servili, M., Selvaggini, R., Esposto, S., Taticchi, A., Montedoro, G., and Morozzi, G. (2004). Health and sensory properties of virgin olive oil hydrophilic phenols: agronomic and

technological aspects of production that affect their occurrence in the oil. *Journal of Chromatography A* **1054**: 113–127.

- Servili, M., Selvaggini, R., Taticchi, A., Esposto, S., and Montedoro, G. (2003). Volatile Compounds and Phenolic Composition of Virgin Olive Oil: Optimization of Temperature and Time of Exposure of Olive Pastes to Air Contact during the Mechanical Extraction Process. *J.Agric. Food Chem.* **51**: 7980-7988.
- Servili, M., Taticchi, A., Esposito, S., Urbani, S., Selvaggini, R., and Montedoro, G. (2008). Influence of the decrease in oxygen during malaxation of olive paste on the composition of volatiles and phenolic compounds in virgin olive oil. *J. Agric. Food Chem.* **56**: 10048-10055.
- Servili, M., Taticchi, A., Esposto, S., Urbani ,S., Selvaggini, R., and Montedoro G. (2007). Effect of olive stoning on the volatile and phenolic composition of virgin olive oil. *J Agric Food Chem.* **55**(17): 7028-7035.
- Siliani, S., Mattei, A., Innocenti, L. B., and Zanoni, B. (2006). Bitter taste and phenolic compounds in extra virgin olive oil: An empirical relationship. *Journal of Food Quality* **29**: 431–441.
- Sinesio, F., Moneta, E., and Esti, M. (2005). The dynamic sensory evaluation of bitterness and pungency in virgin olive oil. *Food Quality and Preference* **16**: 557–564.
- Skevin, D., Rade, D., Strucelj, D., Mokrovcak, Z., Nederal, S., and Bencic, D. (2003). The influence of variety and harvest time on the bitterness and phenolic compounds of olive oil. *Eur. J. Lipid Sci. Technol.* **105**: 536-541.
- Smith, A. B., Han, Q., Breslin, P. A. S., and Beauchamp, G. K. (2005). Synthesis and assignment of absolute configuration of (-)-oleocanthal: a potent, naturally occurring non-

steroidal anti-inflammatory and antioxidant agent derived from extra virgin olive oils. *Org. Lett*. **7**: 5075-7078.

- Stefanoudaki, E., Williams, M., Chartzoulakis, K., and Harwood, J. (2009). Effect of Irrigation on Quality Attributes of Olive Oil. *J Agric Food Chem*. **57**(15): 7048-7055.
- Tamendjari, A., Angerosa, F., and Bellal, M. M. (2004). Influence of Bactrocera oleae infestation on olive oil quality during ripening of Chemlal olives. *Ital. J. Food Sci*. **3**: 343–354.
- Tamendjari, A., Angerosa, F., Mettouchi, S., and Bellal, M.M. (2009). The effect of fly attack (Bactrocera oleae) on the quality and phenolic content of Chemlal olive oil. *Grasas y aceites.* **60**(5): 507-513.
- Temime, S.B., Wael, T., Bechir, B., Leila, A., Douja, A., and Mokhtar, Z. (2006). Changes in olive oil quality of Chetoui variety according to origin of plantation. *Journal of Food Lipids*. **13**: 88-99.
- Tovar, M.J., Motilva, M.J., and Romero, M.P. (2001). Changes in the phenolic composition of virgin olive oil from young trees (Olea europaea L. cv. Arbequina) grown under linear irrigation strategies. *J. Agric. Food Chem*. **49**: 5502–5508.
- Tsimidou, M. (1998). Polyphenols and quality of virgin olive oil in retrospect. *Ital. J. Food Sci*. **2**: 99–116.
- Tuck, K. L., Freeman, M. P., Hayball, P. J., Stretch, G. L., and Stupans, I. (2001). The in vivo fate of hydroxytyrosol and tyrosol, antioxidant phenolic constituents of olive oil, following intravenous and oral dosing of labelled compounds to rats. *J. Nutr.* **131**: 1993-1996

- Tuck, K. L., Hayball, P. J., and Stupans, I. (2002). Structural characterization of the metabolites of hydroxytyrosol, the principal phenolic component in olive oil, in rats. *J. Agric. Food Chem.* **50**: 2404-2409.
- Tura, D., Failla, O., Bassi, D., Pedo, S., and Serraiocco, A. (2008). Cultivar influence on virgin olive (Olea europea L.) oil flavor based on aromatic compounds and sensorial profile. *Scientia Horticulturae* **118**: 139-148.
- Tura, D., Gigliotti, C., Pedo, S., and Failla, O. (2007). Influence of cultivar and site of cultivation on levels of lipophilic and hydrophilic antioxidants in virgin olive oils (Olea europea L.) and correlations with oxidative stability. *Sci. Hort*. **112**: 108–119.
- Valls, R.M., Soler, A., Girona, J., Heras, M., Romero, M.P., Covas, M.I., Solà, R., Masana, L., and Motilva, M.J. (2010). Effect of the long-term regular intake of virgin olive oil on the phenolic. *Journal of Pharmaceutical and Biomedical Analysis* **53**: 68–74
- Vekiari, S.A., and Koutsaftakis, A. (2002). The effect of different processing stages of olive fruit on the extracted olive oil polyphenol content. *Grasas y Aceites* **53** (3): 304-308.
- Vichi, S., Romero, A., Tous, J, and Caixach, J. (2011). The Activity of Healthy Olive Microbiota during Virgin Olive Oil Extraction Influences Oil Chemical Composition.. *J. Agric. Food Chem*. **59**: 4705–4714.
- Visioli, F., and Galli, C. (1995). Natural antioxidants and prevention of coronary heart disease: the potencial role of olive oil and its minor constituents. *Nutr Metab Cardiovasc Dis.* **5**: 306–314.
- Visioli, F., Galli, C., Bornet, F., Mattei, A., Patelli, R., Galli, G., and Caruso, D. (2000). Olive oil phenolics are dose-dependently absorbed in humans. *FEBS Lett.* **468**: 159-160.

- Vissers, M. H., Zock, P. L., Roodenburg, A. J., Leenen, R., and Katan, M. B. (2002). Olive oil phenols are absorbed in humans. *J. Nutr*. **132**: 409-417.
- Vitaglione, P., Napolitano, A., and Fogliano, V. (2008). Cereal dietary fibre: a natural functional ingredient to deliver phenolic compounds into the gut. *Trends in Food Science and Technology*. **19**(9): 451-463.
- Youssef, N.B., Zarrouk, W., Carrasco-Pancorbo, A., Ouni, Y., Segura-Carretero, A., Fernàndez-Gutìerrez, A., Daouda, D., and Zarrouk, M. (2010). Effect of olive ripeness on chemical properties and phenolic composition of ch´etoui virgin olive oil. *J Sci Food Agric* **90**: 199–204.

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LEGENDS TO FIGURES

Figure 1: Chemical structures of the main VOO phenolics. The major VOO polyphenols are secoiridoids, simple phenols, and lignans. Secoiridoids are the most abundant and mainly include 3,4-DHPEA-EDA and pHPEA-EDA (i.e. the dialdehydic form of elenoic acid (EA) linked either to hydroxytyrosol or to tyrosol, respectively) as well as 3,4-DHPEA-EA (i.e. the isomer of oleuropein aglycon). These compounds derive from olive oleuropein glucoside and ligstroside glucoside. During ripening specific glycosidases hydrolyse the sugar moiety and the aglycons can be majorly retained in the oil phase during the extraction process. During VOO storage, seicoridoids undergo to two principle degradation phenomena leading to the dialdehydic compounds or to hydroxytyrosol, tyrosol and elenolic acid. Among lignans pinoresinol and 1-acetoxypinoresinol are majorly represented.

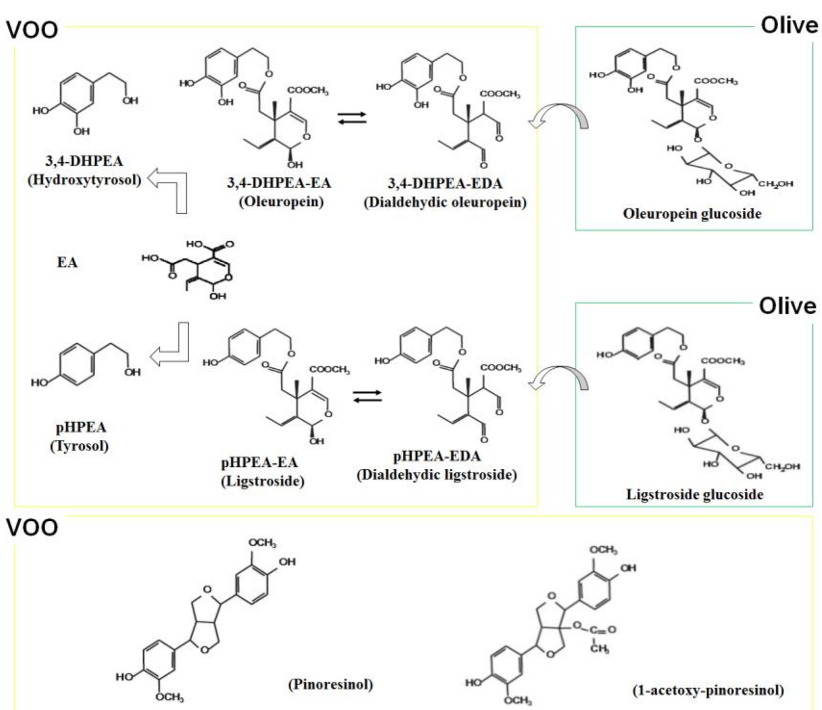

Figure 2: Metabolism of oleuropein and ligstroside aglycon. Oleuropein and ligstroside aglycon - (OH)/TyrEDA - may be partly hydrolysed in the stomach or in the small intestine. The correspondent phenolic acids (hyroxytyrosol and tyrosol, (OH)/Tyr, rapidly pass through the portal vein in the liver. Here they are conjugated to glucuronic acid (GA) and the (hydroxy)/tyrosol-glucuronides, (OH)/Tyr-glu, are formed. (OH)Tyr may be further methylated by cathechol-O-methyltransferase to form (OH)Tyr-glu-met and (OH)Tyr-met (homovanillic alcohol). The latter may be further oxidized to homovanillic acid (HVA) and then sulphated together with (OH)/Tyr, thus HVA-sulf and (OH)/Tyr-sulf may be formed. Liver metabolites pass into the bloodstream and they can be finally filtered by kidney into urine. Blood (OH)/Tyr-glu can also derive from glucuronidation during the passage of (OH)/Tyr through the intestinal mucosa. The possibility of oleuropein and ligstroside aglycons to resist to stomach/intestinal hydrolysis and to be directly absorbed through the small intestine has been demonstrated by the finding of (OH)/Tyr-EDA metabolites (mainly hydrogenated) in the urine. Once in the bloodstream (OH)/TyrEDA may be hydrolysed and their constituents - (OH)/Tyr and EA - may contribute to the cumulus from other body compartments or pass in their original form or metabolised in urine, (OH)/TyrEDA-metabolite (Visioli et al., 2000; Caruso et al., 2001; Vissers et al., 2002; Miro´-Casas et al., 2001; Miro´-Casas et al., 2003a; Mirò-Casas et al., 2003b; Tuck et al., 2002; D'Angelo et al., 2001; Tuck et al., 2001; de la Torre-Carbot et al., 2007).

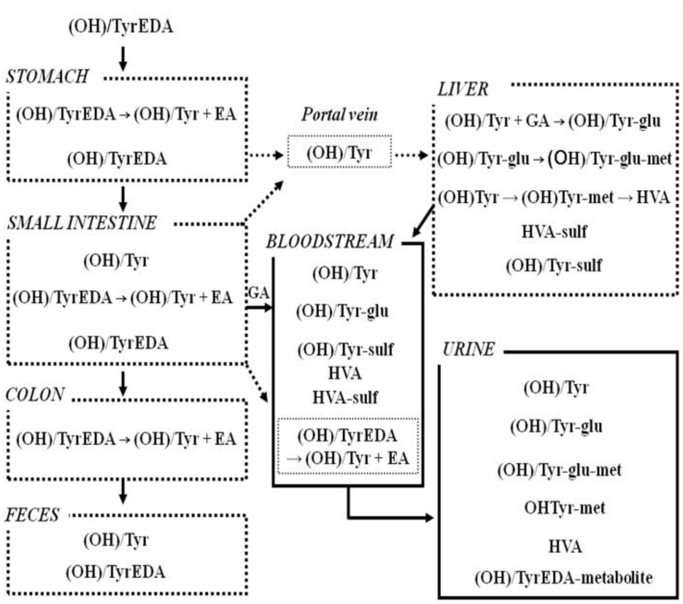

Figure 3: Summary and future perspectives. Opportunity for consumers to get the maximum health benefits from VOO is conditioned by their choice. Scientific evidence indicate that VOO bitterness and pungency can be the sensory drivers towards a healthy choice. Actually consumer preferences do not fit with this indications, being convenience perception linked to sweet taste and cheap cost. However, human physiology and behaviour indicate that habituation to the bitter and pungent VOO is possible and the recent substantiation by EFSA of the health claim for VOO polyphenols may represent the effective strategy to direct consumer choice towards the healthiest one. In fact, it is expected that bitter and pungent VOO will increase in the market as producers will be interested in obtaining the health claim. Taste of these VOO associated to the health claim on the packaging may be the most effective communication strategy to change consumer perception of VOO convenience. A possibly increased consumer demand for high quality products may get to more advantageous quality/price ratio.

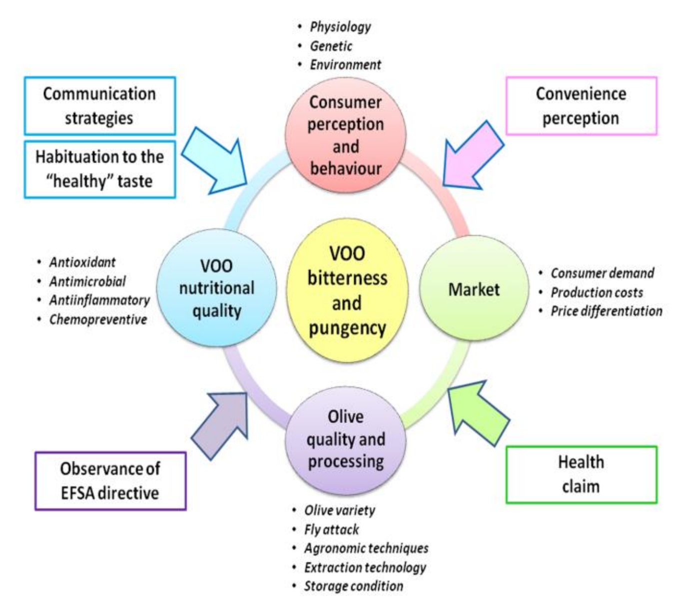

Table 1: Phenolic compounds typical of Mediterranean diet.

Food	Food daily intake	Phenol concentration	Phenol intake	Chemical class	Ref
Fruits	258 g totally 191 g edible	8.8 g/kg dry weight	550 mg/d	▪ Phenolic acids ▪ Flavonoids ▪ Proanthocyanidins (monomers and polymers)	Arranz et al., 2010
Vegetables	279 g totally 230 g edible	3.3 g/kg dry weight	197 mg/d	▪ Flavonoids ▪ Lignans ▪ Proanthocyanidins (monomers and polymers)	Arranz et al., 2010
Cereals	140 g	2.1 g/kg dry weight	353 mg/d	▪ Phenolic acids ▪ Lignans (mainly bound to dietary fibre)	Arranz et al., 2010
VOO	25-50 mL	100-300 mg/kg	5-10 mg/d	▪ Secoiridoids	Cicerale et al., 2010