

Review Article

The potential use of phenolic compounds recovered from olive mill wastewater in food model systems

ABSTRACT

In terms of economic growth, health benefits, and culture, the olive oil industry is a critical sector for many countries. However, olive mill wastewater (OMWW) is one of the most polluting by-products of the manufacture of virgin olive oil. Several studies have reported that OMWW is a valuable resource of usable compounds for recovery and valorization. Because of its high content of phenolic compounds, it may serve a significant function in food because of the phenolics' strong antioxidant value. The current paper provides a survey of OMWW's phenolic recovery methods, focusing on their application as active constituents in food products. In addition, this contribution provides an overview of key research describing the potentialities of OMWW phenolics in food model systems. The Scopus, Web of Science, and Science Direct databases were chosen as our paper references. Based on the available studies, traditional techniques like solvent extraction, membranes, and, more recently, innovative technologies that promise minimum impact on these phytochemicals' compounds are used to recover phenolics from OMWW. Various food products, such as vegetable oils, bakery products, milk beverages, and meat products, can be fortified. All of these applications are based on phenolics' antibacterial properties or their antioxidant abilities to minimize lipid peroxidation during cooking, deep-fat frying, and storage.

Keywords: olive mill byproducts, Olive mill wastewater, phenolic compounds, food matrix

Abbreviations

AGEs: Advanced Glycation Endproducts, **APCI:** Atmospheric Pressure Chemical Ionization, **BCFN:** Barilla Center For Food and Nutrition, **BHA:** Butylated HydroxyAnisole, **BHT:** Butylated HydroxyToluene, **CML:** CarboxyMethyl-Lysine, **ESI:** Electrospray Ionization, **FAB:** Fast Atom Bombardment, **FSI:** Food Sustainability System, **GAE:** Gallic Acid Equivalent, **HT:** HydroxyTyrosol, **LC-MS:** Liquid Chromatography-Mass Spectrometry, **MALDI:** Matrix Assisted Laser Adsorption Ionization, **MF:** MicroFiltration, **NF:** NanoFiltration, **OMW:** Olive Mill Waste, **OMWW:** Olive Mill WasteWater, **PG:** Propyl Gallate, **SCF:** SuperCritical Fluid, **SDGs:** Sustainable Development Goals, **SFS:** Sustainable Food System, **SPE:** Solid Phase Extraction, **RO:** Reverse Osmosis, **RP-SPE:** Reversed Phase-Solid Phase Extraction, **TBHQ:** Tert-ButylHydroQuinone, **UF:** UltraFiltration, **UHT:** Ultra High-Temperature, **VOO:** Virgin Olive Oil.

1. INTRODUCTION

Nowadays, multiple environmental negative impacts, such as biodiversity loss, soil deterioration, and water pollution, are all exacerbated by food production. In fact, the food system is responsible for 20–30% of global greenhouse gas emissions. Humanity has a monumental task in reorganizing food systems to provide healthy diets to all people in a sustainable manner. The 2030 Agenda for Sustainable Development encapsulates 17 Sustainable Development Goals to be achieved by 2030, including responsible production and consumption. The Sustainable Development Goals (SDGs) of the United Nations place a focus on a sustainable food system. The SDGs, which were adopted in 2015, call for fundamental changes in agriculture and food systems by 2030 in order to end hunger, establish food security, and improve nutrition. To achieve the SDGs, the global food system must be transformed [1].

Comment [Y1]: Delete, this is not a recent phenomenon

A sustainable food system (SFS) is one that ensures food security and nutrition for all while preserving the economic, social, and environmental foundations required for future generations to have food security and nutrition. This means that it is lucrative over time (economic sustainability); it helps society broadly (social sustainability); and it has a positive or neutral influence on the environment (environmental sustainability). Rapid population expansion, urbanization, increased income, changing consumption patterns, and globalization, as well as climate change and natural resource depletion, must all be considered [2].

Food system evolution has produced numerous favorable outcomes. As food industries have expanded, these outcomes have included the increase of off-farm employment opportunities and the expansion of food options beyond local staples, satisfying consumers' preferences in terms of taste, form, and quantity. However, the quick structural changes that have accompanied them have created new and serious issues, with potentially far-reaching implications for food security and nutrition. Many highly processed, high-calorie, and low-nutritional-value food items are now widely available and consumed, resulting in high levels of food loss and waste and an ecological footprint associated with the lengthening and industrialization of food supply chains [3].

The consumption of olive oil, which is widely known for its biological activities, is increasing throughout the world due to its health benefits and great nutritional properties [4]. Unfortunately, the challenge of treating and disposing of olive mill wastewater (OMWW) is wreaking havoc on the producing countries due to its high organic content. The agri-food industry has recently been under greater pressure to address social and environmental challenges in their supply chains throughout product lifecycles. Indeed, olive oil production necessitates the use of a considerable number of resources as well as the release of pollutants that have a substantial negative impact on the environment [5].

In order to examine the simultaneous influence of food systems on human health and the environment, the Barilla Center for Food and Nutrition (BCFN) has created the Food Sustainability Index (FSI). It assesses the long-term sustainability of 78 countries' food systems based on three core pillars: food loss and waste, agriculture, and nutritional problems. The index ranges from 0 to 100, with 100 representing the highest level of sustainability [6] (Fig. 1).

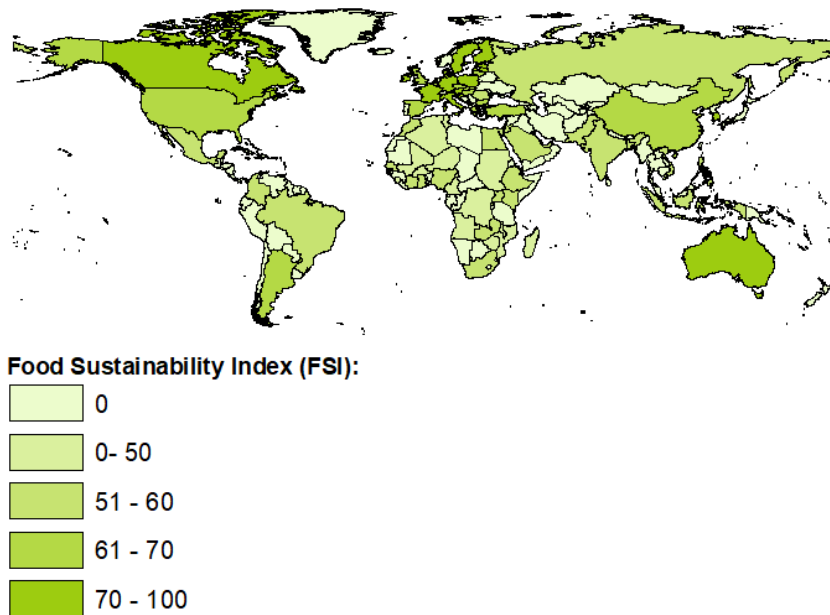


Fig. 1. Food Sustainability Index (FSI) (prepared using data of the Barilla Center for Food and Nutrition BCFN)

Olive mill wastewater (OMWW) has long been thought to be the most polluting and bothersome waste generated by olive mills. One of the most damaging effluents produced by the agro-food industry is said to be this complex medium. OMWW production is estimated to be over 30 million m³ worldwide, with 98% of it generated in the Mediterranean basin. Thus, the management of this liquid residue has been investigated, and some extensive and detailed studies reported that olive production systems conversion (i.e., two-phase instead of three-phase), detoxification methods, and recovery of compounds from OMWW can be considered to treat olive oil processing effluents [7].

In fact, this waste could be easily turned into a valuable source of antioxidant chemicals due to the high concentration of phenolic compounds, which can be added to a variety of foods to develop a functional product with better nutritional properties. In fact, phenolic compounds have been shown to inhibit metal-induced oxidation, scavenge free radicals, serve as reductants, and even preserve food quality. In addition, the antimicrobial activities of phenolics have been demonstrated, suggesting their usage as natural additives to extend the shelf life of foods. Recovering phenolic compounds not only gives financial and nutritional benefits, but also reduces the OMWW's environmental impact [8].

Olive oil extraction entails a number of steps, including olive washing, crushing, and malaxing, as well as the extraction itself, which is the most basic stage of the entire process. The extraction process will determine the amount and physical-chemical properties of the olive oil as well as those of the waste produced. The two processes for extracting the oil are

traditional pressing, which has been employed for millennia, and centrifugation, which the olive oil industry has taken over in recent decades [9].

This technology is referred to as three-phase because the centrifugal decanter separates three flows of matter: olive oil, pomace, and wastewater. This process, however, necessitates the addition of warm water to dilute the olive paste, thereby generating a significant amount of OMWW [9].

Fig. 2 depicts the extraction method applied to virgin olive oil production with regard to the byproducts obtained from the three-phase system process.

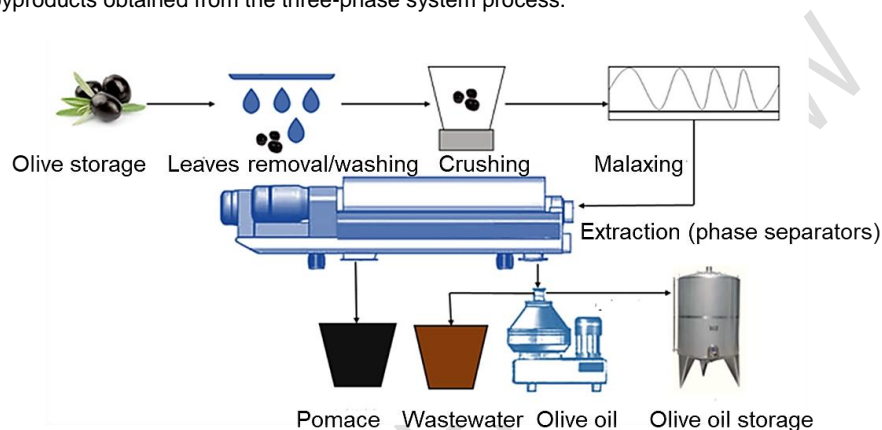


Fig. 2. Three-phase system process

The annual production of OMWW in the world is estimated to be between 10 and 30 million m³. The discontinuous technique (which is no longer widely employed) produces less but more concentrated waste (0.5-1 m³ per 1000 kg) than centrifugation (1-1.5 m³ per 1000 kg) [10].

There is a scarcity of precise and comprehensive data on the amount of waste generated by the olive mill industry and where it is generated. However, because OMWW contains various high-added-value compounds, the generation amount should be exploited.

OMWW is mainly made up of 83–94% water, 4–16% organic compounds, and 0.4-2.5% mineral salts. Sugars, tannins, phenolic compounds, polyalcohols, minerals, pectins, and lipids are present in OMWW. It has a high potassium concentration and considerable amounts of nitrogen, phosphorus, calcium, magnesium, and iron when compared to other organic wastes [11]. Despite the high concentration of phenolic compounds in the olive fruit, only 2% of the initial concentration is found in the VOO, with the remainder (about 53%) present in the OMW and the pomace (approximately 45%). This is owing to the hydrophilic character of phenolics and the contact between the water phase and the oil phase during the extraction [11]. It has a black color (due to lignin polymerization with phenolic chemicals), is acidic (pH of around 5), and has high electrical conductivity (Table 1).

Table 1. Physical and chemical properties of olive mill wastewater

	Unit	Range of values	References
pH	–	4.8–5.7	[12]
Conductivity	mS/cm	5–81	[12]
COD	g/L	16.5–156	[13]
BOD	g/L	13.4–37.5	[14]
Dry residue	g/L	11.5–90	[15]
Lipids	g/L	7	[15]
Sugar	g/L	1.3–4.3	[16]
Total nitrogen	g/L	0.06–0.9	[17]

COD: Chemical Oxygen Demand ; BOD: Biochemical Oxygen Demand

The presence of significant levels of organic chemicals in OMWW could explain their phytotoxic and bactericidal properties in part. As a result, significant research efforts have been focused on developing purification and treatment procedures that can recover the high-value phenolic chemicals from OMWW while reducing the volume and severe environmental effects of these hazardous effluents.

Solvent extraction, chromatographic separations, centrifugation, membrane processes [18], and, more recently, high voltage electrical discharges, pulsed electric fields, and ultrasounds [19, 20] have been proposed to recover phenolics from OMWW [21]. However, the two most frequently utilized methods for recovering phenolic chemicals are solvent extraction and membrane separation.

Despite the high cost of using significant amounts of organic solvents, solvent extraction is the most extensively utilized method for recovering phenolic compounds from OMWW [22, 23]. Several authors, including Kalogerakis et al. [24], employed ethyl acetate. Allouche et al. [25] described ethyl acetate as the most effective solvent for recovering phenols from OMWW produced using a three-phase mill.

Emmons and Guttersen's [26] method for obtaining oleuropein aglycon from OMWW entails adding citric acid to the raw material, boiling it to precipitate the solids, then extracting the oleuropein aglycon from the water immiscible component with a non-polar organic solvent mixture, preferably a 50/50 v/v hexane/acetone mixture. After that, the solvent is evaporated using a vacuum and/or heat.

De Marco et al. [28] used the SPE extraction method to recover biophenols from OMW, stating that reversed phase-solid phase extraction (RP-SPE) yielded about 1 g of pure hydroxytyrosol/1 L of OMWW.

Galanakis et al. [29] described a method for isolating dietary fibers from OMWW, including pectins and important phenolic compounds, as well as the utilization of isolated products as food additives and antioxidants, respectively. To begin with, OMWW is centrifuged to remove the fat and then concentrated by removing the water content. Following that, it is extracted with ethanol (up to 7% v/v) and an organic acid (in the range of 0.5 to 3% by weight of the extraction solution). One of the following organic acids can be used in the process: citric, tartaric, malonic, maleic, malic, oxalic, adipic, or fumaric. The remaining polyphenols in the dietary fibers are then extracted and filtered with ethanol at a

concentration of at least 85 percent (v/v). The polyphenol-containing liquid phase is clarified by filtration after dilution with 15-40% (v/v) ethanol.

The use of supercritical fluids (SCFs), notably supercritical CO₂, has been shown in recent studies to overcome the limitations of organic solvents such as toxicity and flammability. SCF extraction, on the other hand, comes with the drawback of requiring the use of expensive high-pressure equipment. Thus, phenolic chemicals were extracted from OMWW using a normal liquid solvent and a CO₂ supercritical fluid, as described by Lafka et al. [30]. The phenolic content was measured as caffeic acid equivalents on a dry basis (% w/w) and ranged from 0.43% to 1.29% using solvent extraction against 0.76% using supercritical CO₂.

For the recovery of phenolics from olive oil by-products, solvent extraction is used singly or preferably prior to membrane treatments. Membrane technologies are appealing for recovering phenolic compounds from olive mill wastewater because of their numerous advantages, including minimal energy consumption, no chemical requirements, and no phase shift. Despite the fact that conventional filtration membranes are still commonly used in the treatment of OMWW, microfiltration (MF), ultrafiltration (UF), nanofiltration (NF), and reverse osmosis (RO) membranes, mostly in sequential form, effectively cover the needs for the recovery, purification, and concentration of antioxidants in terms of their specific molecular weight cut-off values [20].

The large molecular weight range of OMWW chemicals makes high-purity recovery difficult. However, this can be solved via membrane technologies. Microfiltration membranes can hold microparticles as small as 0.1-10 μm, whereas UF membranes can hold macromolecules as small as 1-100 nm. NF membranes can separate molecules with a diameter of 0.5-5 nm, while RO membranes retain molecules with a diameter of less than 1 nm [31].

As a result, many membrane-based approaches for recovering natural antioxidants from OMWW have been investigated. Villanova et al. [32] proposed a method for recovering tyrosol and hydroxytyrosol from OMWW. Rough filtration (RF), microfiltration, ultrafiltration, nanofiltration, and reverse osmosis units, followed by column chromatography, are recommended for recovering tyrosol and hydroxytyrosol from polyphenolic fractions or their purified components of OMWW.

Membrane filtration was used by D'Antuono et al. [33] to recover OMWW phenolics from two Italian and three Greek olive varieties. MF, UF, and NF each produced three fractions. Each fraction had a distinct level of purity as well as a varying amount of phenolic compounds.

The fractions generated by a membrane method, specifically the reverse osmosis concentrate, after utilizing NF were characterized by Zagklis et al. [34]. Adsorption/desorption resins were used to further treat these phenolic compounds. Finally, vacuum evaporation was used to concentrate the recovered phenolic compounds, yielding a final extract with a phenolic concentration of 378 g GAE/L.

Bazzarelli et al. [35] recovered purified polyphenol-concentrated retentate, using a combination of microfiltration and nanofiltration membranes, as well as osmotic membrane distillation and membrane emulsification processes.

To obtain a dried phenolic-rich fraction of OMWW, Sedej et al. [36] used a two-step sequential filtration approach (ultrafiltration/reverse osmosis) and various drying procedures (spray drying, freeze drying, or infrared drying). In 3-phase OMWW, spray drying led to the highest total phenolic content, antioxidant activity, and phenolic compound content.

De Almeida et al. [37] used a combination of ultrafiltration and nanofiltration to develop a polyphenol-concentrated retentate that contained oleuropein, gallic acid, syringic acid, and HT.

Tight ultrafiltration and nanofiltration membranes offer excellent bioactive chemical recovery rates (>80%) and are the most convenient for polyphenol recovery [38].

Another study found that combining microfiltration, nanofiltration, and reverse osmosis produced an OMWW polyphenol-concentrated retentate with a high HT content (1.52 g/L), tyrosol content (0.52 g/L), and oleuropein content (0.51 g/L), as well as antioxidant hypolipidemic and hypoglycemic characteristics *in vitro* [31]. Similarly, to extract polyphenols and carbohydrates from OMWW, a pilot membrane design combining ultrafiltration, nanofiltration, and reverse osmosis membranes was developed [39].

Other methods, like enzymatic processing, have been employed to recover phenolic compounds from OMWW. According to Khoufi et al. [40], the OMWW hydrolyzed with an enzymatic preparation from *Aspergillus niger* cultivated on wheat bran could be a valid source of bioactive-free phenolic compounds (especially hydroxytyrosol) with interesting applications. Another study discovered that pre-treating OMWW with *Aspergillus niger* and *Trichoderma atroviride* culture broths rich in α -glucosidase increased the amount of hydroxytyrosol produced. Similar investigations with *Trametes trogii* culture media, however, revealed a substantial oxidation of phenolic chemicals due to this strain's strong laccase activity [41]. In the same context, Hamza and Sayadi [42] investigated the efficacy of enzymatic pre-treatment (with *Aspergillus niger* α -glucosidase) and membrane technology (using MF and UF). Overall, this method was successful in recovering hydroxytyrosol.

The qualitative and quantitative determination of OMWW phenolic compounds is difficult due to the matrix's complexity and the heterogeneity of the fraction of interest. However, technological advancements in analytical instruments over the previous few decades are assisting in overcoming the aforementioned barrier. In this regard, the employment of high-resolution separation techniques that aid in the subsequent detection of individual components substantially assists in the improvement of the proposed methods' selectivity and sensitivity. On the other hand, the lack of commercially available pure standards for a large number of OMWW phenolics, as well as difficulties in completely resolving complex chromatographic profiles, has made the use of MS detection almost mandatory due to its ability to confirm identity and quantify overlapped peaks. Many studies have shown the effectiveness of the LC-MS for the identification of phenolic compounds in olive processing byproducts [43, 44]. In mass spectrometry, there are a variety of ionization processes that are entirely compatible with liquid chromatography (LC), such as atmospheric pressure chemical ionization (APCI), electrospray ionization (ESI), Fast Atom Bombardment (FAB) and matrix assisted laser desorption ionization (MALDI).

OMWW is a rich source of polyphenols, with secoiridoid derivatives such as hydroxytyrosol, the dialdehydic form of decarboxymethyl oleuropein aglycone, tyrosol, and verbascoside being particularly important [45]. Olive polyphenols are partitioned between the water and oil phases during the extraction of olive oil. However, because they are water-soluble substrates with strong polarity, the bulk percentage is missing in the oil phase. The extraction and identification of phenolics from OMWW seems promising because they're powerful natural antioxidants that have sparked a lot of interest in the cosmetic, food, and pharmaceutical industries. OMWW has been shown to contain almost forty distinct phenolic chemicals [4].

The ability to characterize OMWWs in terms of phenolic content is critical for developing effective re-evaluation approaches. As a result of the complexity of OMWWs, analytical procedures capable of providing a complete qualitative and quantitative screening of their composition have been developed. Several analytical approaches have been published in the literature for the identification and quantification of specific phenolic compounds in OMWW. Molecular formulas and the method of extraction and identification of the phenolic compounds in OMWW are listed in Table 2.

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Table 2. The Phenolic Compounds Most Commonly Found in Olive Mill Wastewaters

Phenolic compound	Molecular formula	Method of extraction	Method of identification	References
3,4-dihydroxyphenylglycol (DHPG)	C ₈ H ₁₀ O ₄	Solvent extraction	MALDI-TOF-MS	[46]
Apigenin	C ₁₅ H ₁₀ O ₅	Ultrasound-Assisted Solid Liquid Extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Apigenin-7- <i>O</i> -glucoside	C ₂₁ H ₂₀ O ₁₀	Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Apigenin-7- <i>O</i> -rutinoside	C ₂₇ H ₃₀ O ₁₄	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
β-hydroxy-isoverbascoside	C ₂₉ H ₃₆ O ₁₆	Membrane technology coupled to low-pressure gel filtration chromatography on a Sephadex LH-20 Membrane extraction	HPLC-DAD-MS/MS LC-DAD-ESI-MS ⁿ	[49] [33]
Caffeic acid	C ₉ H ₈ O ₄	Solvent extraction Membrane extraction	RPLC-DAD RPLC-ESI-MS LC-DAD-ESI-MS ⁿ	[48] [33]
Caffeoyl-6'-secologanoside	C ₂₅ H ₂₈ O ₁₄	Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]

(Cafselogoside)		Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
Chlorogenic acid (Caffeoyl-quinic acid)	$C_{16}H_{18}O_9$	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
Chrysoeriol	$C_{16}H_{12}O_6$	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
Comselogoside	$C_{25}H_{27}O_{13}$	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47]
		Membrane extraction	LC-DAD-ESI-MS ⁿ	[33]
Demethyloleuropein	$C_{24}H_{30}O_{13}$	Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47]
Dihydro-oleuropein	$C_{25}H_{36}O_{13}$	Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47]
Elenolic acid (EA)	$C_{11}H_{14}O_6$	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47]
		Solvent extraction	HPLC-ESI-Q-TOF-MS	[50]

Elenolic acid (EA) derivative (decarboxylated form of hydroxyelenolic acid)	C ₁₀ H ₁₄ O ₅	Solvent extraction	MALDI-TOF MS	[46]
		Solvent extraction	HPLC-ESI-Q-TOF-MS	[50]
Elenolic acid glucoside (Oleoside methyl ester)	C ₁₇ H ₂₄ O ₁₁	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Gallic acid	C ₇ H ₆ O ₅	Solvent extraction	HPLC-ESI-Q-TOF-MS	[50]
Hydroxytyrosol (3,4-DHPEA)	C ₈ H ₁₀ O ₃	Adsorption resin technology	LC-ESI-MS/MS	[51]
		Solvent extraction	RPLC-DAD	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	RPLC-ESI-MS UPLC-DAD-ESI-QTOF-HRMS	[47]
		Membrane extraction	LC-DAD-ESI-MS ⁿ	[33]
		Solvent extraction	MALDI-TOF-MS	[46]
		Solvent extraction	HPLC-ESI-QTOF-MS	[50]
Hydroxytyrosol glucoside	C ₁₄ H ₂₀ O ₈	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Membrane extraction	LC-DAD-ESI-MS ⁿ	[33]

		ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Hydroxytyrosil	C ₁₉ H ₂₆ O	Solvent extraction	RPLC-DAD	[48]
acyclodihydroelenolate		Membrane extraction	RPLC-ESI-MS LC-DAD-ESI-MS ⁿ	[33]
Ligstroside	C ₂₅ H ₃₂ O ₁₂	Solvent extraction	RPLC-DAD	[48]
		ultrasound-assisted solid liquid extraction (USLE) method	RPLC-ESI-MS UPLC-DAD-ESI-QTOF-HRMS	[47]
Luteolin	C ₁₅ H ₁₀ O ₆	Solvent extraction	RPLC-DAD	[48]
		ultrasound-assisted solid liquid extraction (USLE) method	RPLC-ESI-MS UPLC-DAD-ESI-QTOF-HRMS	[47]
Luteolin-4'- <i>O</i> -glucoside	C ₂₁ H ₂₀ O ₁₁	Solvent extraction	RPLC-DAD	[48]
		ultrasound-assisted solid liquid extraction (USLE) method	RPLC-ESI-MS UPLC-DAD-ESI-QTOF-HRMS	[47]
Luteolin-7- <i>O</i> -glucoside	C ₂₁ H ₂₀ O ₁₁	Solvent extraction	RPLC-DAD	[48]
		ultrasound-assisted solid liquid extraction (USLE) method	RPLC-ESI-MS UPLC-DAD-ESI-QTOF-HRMS	[47]
Luteolin-7- <i>O</i> -rutinoside	C ₂₇ H ₃₀ O ₁₅	Solvent extraction	RPLC-DAD	[48]
			RPLC-ESI-MS	

Luteolin- <i>O</i> -rutinoside	C ₂₇ H ₃₀ O ₁₅	Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Oleacin (3,4-DHPEA-EDA)	C ₁₇ H ₂₀ O ₆	Solvent extraction	MALDI-TOF MS	[46]
		Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Olenoside A and B	C ₁₁ H ₁₄ O ₅	Solvent extraction	HR-ESI-MALDI-TOF-MS	[52]
Oleocanthal (<i>p</i> -HPEA-EDA)	C ₁₇ H ₂₀ O ₅	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Oleoside	C ₁₆ H ₂₂ O ₁₁	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
		Membrane extraction	LC-DAD-ESI-MS ⁿ	[33]
Oleuropein	C ₂₅ H ₃₂ O ₁₃	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]

			LC-DAD-ESI-MS ⁿ	
		Membrane extraction		[33]
Oleuropein aglycone (3,4-DHPEA-EA)	C ₁₉ H ₂₂ O ₈	Solvent extraction	MALDI-TOF-MS	[46]
		Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47]
Oleuropein aglycone derivative	C ₁₆ H ₂₆ O ₁₀	Solvent extraction	RPLC-DAD	[48]
		Membrane extraction	RPLC-ESI-MS LC-DAD-ESI-MS ⁿ	[33]
Oleuropein glucoside isomers	C ₃₁ H ₄₂ O ₁₈	Solvent extraction	RPLC-DAD	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	RPLC-ESI-MS UPLC-DAD-ESI-QTOF- HRMS	[47]
Oleuroside	C ₂₅ H ₃₂ O ₁₃	Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47]
<i>p</i> -coumaric acid	C ₉ H ₈ O ₃	Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47]
		Membrane extraction	LC-DAD-ESI-MS ⁿ	[33]

<i>p</i> -Hydroxybenzoic acid (4-hydroxybenzoic acid)	C ₇ H ₆ O ₃	Solvent extraction	HPLC-ESI-Q-TOF-MS	[50]
Protocatechuic acid	C ₇ H ₆ O ₄	Solvent extraction	HPLC-ESI-Q-TOF-MS	[50]
Quercetin	C ₁₅ H ₁₀ O ₇	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
Quercetin-3- <i>O</i> -rhamnoside	C ₂₁ H ₂₀ O ₁₁	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
Quinic acid	C ₇ H ₁₂ O ₆	Membrane extraction	LC-DAD-ESI-MS ⁿ	33
Rutin	C ₂₇ H ₃₀ O ₁	Solvent extraction	RPLC-DAD	[48]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF- HRMS	[47 α]
		Solvent extraction	LC-DAD-ESI-MS ⁿ	[33]
Tyrosol	C ₈ H ₁₀ O ₂	Adsorption resin technology	LC-ESI-MS ²	[51]
		Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]
		Solvent extraction	LC-DAD-ESI-MS ⁿ	[33]
		Solvent extraction	HPLC-ESI-Q-TOF-MS	[50]
Tyrosol glucoside	C ₁₄ H ₂₀ O ₇	Solvent extraction	RPLC-DAD RPLC-ESI-MS	[48]

		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]
Verbascoside	C ₂₉ H ₃₆ O ₁₅	Solvent extraction	LC-DAD-ESI-MS ⁿ	[33]
		Ultrasound-assisted solid liquid extraction (USLE) method	UPLC-DAD-ESI-QTOF-HRMS	[47]

MALDI-TOF-MS: matrix-assisted laser desorption/ionization (MALDI) time-of-flight mass spectrometry, UPLC-DAD-ESI-QTOF-HRMS: ultra-high pressure liquid chromatography system with diode array and electrospray ionization quadrupole time-of-flight high resolution mass spectrometry, RPLC-DAD: reversed phase HPLC-photodiode array detection, RPLC-ESI-MS: RPLC-electrospray ionisation mass spectrometry, LC-DAD-ESI-MSⁿ: liquid chromatography/diode array detection/electrospray ion trap tandem mass spectrometry, LC-ESI-MS/MS : liquid chromatography-electrospray ionization tandem mass spectrometry

Table 3. Range of contents of phenolics quantified in OMWW

Phenolic compound	Contents range	Method of quantification	Reference
Apigenin	2.5–6.5 ^a	RP-HPLC-UV-MS	[53]
Caffeic acid	0.014–0.017 ^c	RP-HPLC-UV-MS	[53]
Elenolic acid (EA)	4.9–11.7 ^a	RP-HPLC-UV-MS	[53]
Gallic acid	3.86–6.71 ^a	HPLC-ESI-QTOF-MS	[50]
	22.2–61.0 ^a	RP-HPLC-UV-MS	[53]
Hydroxytyrosol (3,4-DHPEA)	483.0–1733.2 ^a	HPLC-ESI-QTOF-MS	[50]
	157.2–245.1 ^a	RP-HPLC-UV-MS	[53]
	544–1560 ^a	HPLC-DAD	[54]

	1230–1290 ^a	RPLC-DAD RPLC-ESI-MS	[48]
	0.25–18.2 ^b	LC-ESI-MS/MS	[51]
Hydroxytyrosol glucoside	1300–1700 ^a	RPLC-DAD RPLC-ESI-MS	[48]
Ligstroside	0.0087–0.0092 ^c		[4]
Luteolin	2.5–36.2 ^a	RP-HPLC-UV-MS RPLC-DAD	[53]
	270–510 ^a	RPLC-ESI-MS	[48]
Luteolin-7- <i>O</i> -glucoside	0–0.0214 ^c		[4]
Luteolin-hexoside	3.2–24.2 ^a	RP-HPLC-UV-MS	[53]
Oleacin (3,4-DHPEA-EDA)	11300–45951 ^a	HPLC-DAD	[54]
Oleuropein derivative	5400–7600 ^a	RP-HPLC-UV-MS RP-HPLC-UV-MS RPLC-DAD	[53] [48]
Oleuroside	200–400 ^a	RP-HPLC-UV-MS RPLC-DAD	[48]
<i>p</i> -coumaric acid	15.9–21.8 ^a	RP-HPLC-UV-MS	[53]
<i>p</i> -hydroxybenzoic acid (4-hydroxybenzoic acid)	1.75–6.15 ^a	HPLC-ESI-QTOF-MS	[50]

Protocatechuic acid	2.77–5.29 ^a	HPLC-ESI-QTOF-MS	[50]
	25.3–136.7 ^a	RP-HPLC-UV-MS	[53]
Rutin	440–640 ^a	RPLC-DAD	[48]
		RPLC-ESI-MS	
Tyrosol	218.4–581.0 ^a	HPLC-ESI-QTOF-MS	[50]
	1180–1560 ^a	RP-HPLC-UV-MS	[48]
	0.19–4.32 ^b	RPLC-ESI-MS	[51]
Vanillic acid	1.68–62.7 ^a	HPLC-ESI-QTOF-MS	[50]
	0.0174–0.0198 ^b		[4]
Verbascoside	14496–24100 ^a	HPLC-DAD	[54]
	1620–1760 ^a	RP-HPLC-UV-MS	[48]

Contents are given as ^a: mg/Kg , ^b: mg/mL, and ^c: weight %

1
2

The cultivar, pedoclimatic conditions, maturity of the fruit, processing conditions, and degree of hydrolysis of OMWW (related to its storage conditions) are all factors that influence the concentrations of the phenolic compounds in the OMWW [55]. Due to these various factors, including the phenolic recovery approach and the analytical technique used, both qualitative and quantitative profiles are dependent. 4-hydroxybenzoic acid, protocatechuic acid, and vanillic acid were all discovered in OMWW [56, 57, 58]. Furthermore, the presence of polymeric phenols with a lignin-like structure, which make up the effluent's most resistant component, has been attributed to the effluent's characteristic brownish-black color [59]. The inherent unpredictability of the wide variety of analytical parameters and methodologies used to extract and evaluate the phenolic compounds, as described above, could possibly explain the diversity of phenolics in OMWW. Nonetheless, the phenolic concentration of OMWW from different origins varies greatly, complicating the identification and quantification procedures.

In addition to its phenolic content, OMWW includes numerous valuable nutrients, including sugar, proteins, and phosphate. OMWW has also been shown to contain soluble dietary fibres, particularly pectin substances with excellent gelling properties [29].

This article presents a literature review of major research outlining OMWW's phenolic recovery techniques and the possible applications of OMWW's phenolics for the development of functional food products. The purpose of this research is to present a critical overview of pertinent scientific studies centered on OMWW's phenolics and their possible application in food model systems. It summarizes current knowledge in the valorization of **byproducts** of a vital agro-food product, such as **VOO**.

2. Methodology

This study was conducted in order to find and document scientific papers that are relevant to the topic. The papers included in this review are strictly limited to scientific research published in peer-reviewed journals. The study was conducted in three phases: preparation, execution, and analysis. The review strategy was devised in the planning phase. The authors came to a conclusion about the criteria used to record the papers and the key questions addressed in the review.

Following this stage of conceptualization, the authors proceeded on to the actual work. In this step, data mining was achieved through a review of scientific databases, the recording of all OMWW-related papers, and the final selection of those whose contexts fit the current review's purpose. The Scopus, Web of Science, and Science Direct databases were chosen as our paper references. Papers were found in these databases by searching for titles, abstracts, and keywords.

"OMWW (olive oil mill wastewater)," "OMWW's phenolics," "recovery of phenolics from OMWW," and "application of OMWW's phenolics in food model systems" were the main keywords employed in this procedure. The process was completed with a third phase, which was the analysis of the results. Furthermore, in order to provide a reliable and qualitative database of papers, the scientific work reported above is confined to articles in peer-reviewed scientific journals published in English.

In this regard, the manuscript has made no attempt to shed light on research that has been done and published in conference proceedings, PhD or master's theses, diploma

Comment [Y2]: Reduce the introduction too legthy

Comment [Y3]: space

Comment [Y4]: full meaning

dissertations, working papers, or textbooks. As a result, limiting the study to scientific journals ensures that the analysis is thorough and unbiased.

Thus, the authors examined the recorded papers and classified them into groups based on their content, as defined during the planning phase. The publications have been categorized according to the year of publication, the tested concentration of recovered phenolics, the food matrix, and the impact they investigate. The analysis includes papers that have been published until the end of 2021.

Comment [Y5]: let themethodology be brief too

3. Results and Discussion

Recently, OMWW has been proposed for the formulation of functional foods with a wide range of effects. Indeed, phenolics identified in olive mill by-products have been extensively reported in the literature for their bioactive potential. Their valorization not only aids in environmental conservation but also provides natural bio-ingredients for re-use in food and non-food applications. Natural phenolic compounds, particularly those recovered from food industry by-products, are promising antioxidants for use in various foods as substitutes for synthetic antioxidants. From both an environmental and food technology standpoint, the use of OMWW as a valuable source of phenolic compounds capable of adding functional value to food items is of great interest.

In fact, Visioli et al. [60] investigated three phenolic extracts obtained from OMWW that demonstrated a high antioxidant capacity when compared to the most commonly used antioxidants in foods, but with varying degrees of activity related to phenolic content and purity.

According to Kachouri and Hamdi [61], the incorporation of OMWW (fermented by *Lactobacillus plantarum*) into olive oil facilitated the decrease of phenolics in wastewater residuals and the rise of phenolics in olive oil. This is due to *Lactobacillus plantarum*'s ability to depolymerize high-molecular-weight phenolics in OMWW, which allows them to potentially transfer from wastewater to oil. These researchers discovered that combining OMWW with fermented *L. plantarum* with plain or non-fermented OMWW resulted in a considerable increase in polyphenol concentration in the oil, with 703 and 112 mg/L of oil, respectively. Individual phenolic compounds, particularly oleuropein content, showed a similar pattern in this investigation, with 401.8 and 140.4 mg/L in oil samples with and without fermented *L. plantarum*, respectively.

The antioxidant potential of OMWW extract was compared to that of BHA and BHT. Peroxide values were lower when OMWW extract was added at 500 ppm compared to BHA [62].

In comparison to vitamin E and C, hydroxytyrosol, recovered from OMWW, has shown higher antiradical effects and has thus been utilized to prevent lipid oxidation in fish products [63].

Servili et al. [54] increased the concentration of phenolic compounds in VOO and EVOO by adding phenolics recovered from OMWW. The crude phenolic extract was obtained by membrane filtration, and it was then extracted with ethyl acetate and ethanol. The crude phenolic extract was also added during the extraction process, specifically before the malaxation stage. In comparison to negative control, adding 5 or 10% of this phenolic concentrate resulted in a considerably increased content of total phenolic compounds, particularly 3,4-DHPEA and 3,4-DHPEA-EDA.

The effect of OMWW phenolic compounds added to milk beverages on beneficial bacteria in yogurt and similar products was investigated. The addition of phenolic extract had minimal effect on the concentration of *Lactobacillus* and *Streptococcus* bacteria during fermentation [65].

The use of OMWW for the creation of a functional beverage was proposed by Zbakh et al. [66]. Commercial products contain a variety of chemicals, including antioxidants like ascorbic acid, chelators like ethylenediaminetetraacetic acid (EDTA), and acidifiers like citric acid. When OMWW extract was used in beverages, no additional antioxidants were required.

Troise et al. [67] evaluated the antioxidant activity of OMWW phenolic extract in UHT milk samples and investigated its potential for the inhibition of the Maillard reaction (MR) by adding phenolic extract at 0.1 and 0.05% w/v. The authors reported an inhibition of reactive carbonyl species formation in samples prior to heat treatment. They also revealed greater stability of tested samples without any sensorial negative attributes.

To extend the shelf-life at 4°C, a phenolic concentrate from OMWW was employed to treat the surface of fresh chicken breasts. The samples were dipped for 1 minute in the crude solution, then cleaned and packaged on a rigid tray with a gas permeable pellicle [68]. *Enterobacteriaceae* and *Pseudomonas* spp. showed a delay in growth in the dipping samples. In addition, the bactericidal activity of OMWW phenolic extract was assessed on a variety of spoilers, starters, and food-borne bacteria (*Staphylococcus* spp., *Listeria* spp., *Escherichia* spp., *Salmonella* spp., *Pseudomonas* spp., *Lactobacillus* spp., and *Pediococcus* spp.) in order to propose them as natural additives for extending food shelf life. Resistance to phenolics was lowest in *Staphylococcus aureus* and *Listeria monocytogenes*. Fasolato et al. [68] found that gram-negative bacteria (*S. Typhimurium* and *Pseudomonas* spp.) were unaffected by the tested doses, but starter cultures (*Staphylococcus xylosum*) proliferated at a much slower rate. Fasolato et al. [68] discovered that a 38.6 g/L concentration of phenolic extract was efficient in extending the shelf life of fresh chicken breast. When compared to the control, the results revealed a delay in growth of both *Enterobacteriaceae* and *Pseudomonas* spp., as well as a 2 day improvement in shelf life.

Using a surface treatment, Chavez-Lopez et al. [69] did a similar study in which OMWW phenolics were applied to fermented sausages to suppress mold populations. The researchers discovered that soaking the product in a 2.5% phenolic solution inhibited some fungi [69].

Previous research has demonstrated that a polyphenol-rich extract from OMWW can preserve the α -tocopherol content while frying refined olive oil and prevent the generation of undesirable volatile compounds [70]. Esposto et al. [70] investigated the impact of purified phenolic concentrate recovered from OMWW in regulating oil stability in a frying process. The phenolic extract was added to a refined olive oil at different concentrations (100, 200, 400, and 1200 mg/kg of polyphenols) and its effects were compared to those of a refined olive oil containing BHT and an EVOO with a high phenolic content. When added at a concentration of at least 400 mg polyphenols/Kg, this OMWW extract, which mostly comprises tyrosol, hydroxytyrosol, 3,4-DHPEA-EDA, and verbascoside, was found to have a stronger ability to retain α -tocopherol content than BHT, decreasing oxidation.

The effectiveness of commercially available phenolics recovered from OMWW (powder form containing 10% polyphenols and 5% hydroxytyrosol) in comparison to other antioxidants (e.g., ascorbic acid, tocopherol mixture, and α -tocopherol) in inhibiting microbial growth of bread and rusks during storage has been studied [71]. Concentrations were chosen to protect against oxidative deterioration as well as microbiological spoilage. The bread and

rusk samples were stored for 20 days and 12 weeks, respectively, and subjected to various microbiological tests on a regular basis (e.g., total coliforms, yeast-moulds, and *Bacillus* spp.). According to the findings, antioxidants were able to induce antibacterial qualities in bakery items and hence extend their shelf life. The tested concentrations of 200 mg/kg flour was shown to be the most effective in extending the shelf life of both bread and rusk samples.

Phenolic compounds from OMWW act as natural food antioxidants, oxidative stabilizers, and texture enhancers in food emulsions as well as antimicrobial molecules in meat products [72].

Chavez-López et al. [73] used OMWW-extracted phenolics to improve quality parameters and increase the shelf life of fermented sausages. In both *in vitro* and *in vivo* experiments, the extracts reduced fungal growth and spore germination in fermented sausages in a dose-dependent manner. *Cladosporium cladosporioides*, *Penicillium aurantiogriseum*, *Penicillium commune*, and *Eurotium amstelodami* were all significantly inhibited *in situ* after being treated with 2.5 % OMWW phenolic extract.

De Leonardis et al. [74] recommended combining lard with phenolics as a "novel food," demonstrating that OMWW's natural antioxidants were particularly effective in preventing lard from oxidizing. The phenolic extract considerably enhanced the oxidative stability of fat and, when evaluated on mouse cell lines, the applied levels (100-200 ppm) were not cytotoxic (embryonic fibroblasts).

Other researchers incorporated pure OMWW phenolic compounds into white meat hamburgers at concentrations of 750 mg/kg or 1500 mg/kg to see how they impacted product shelf life after 11 days at 4 °C. The inhibition of total mesophilic count was effective at all concentrations tested, notably at the highest concentration [75].

The effect of OMWW-extracted polyphenols in white meat burgers packaged in PVC on increasing sensory and sanitary qualities was studied by Veneziani et al. [75]. The incorporation of the phenolic extract at different concentrations (0.75 and 1.50 g/kg) inhibited the growth of mesophilic aerobic bacteria in comparison to both the control and the sample treated with the lowest concentration, demonstrating a dose-dependent behavior effect with a 24 hour extension of shelf life.

Galanakis et al. [76] investigated the antioxidant effect of OMWW phenolic extract in combination with other antioxidants, finding that it reduced oxidative deterioration during bread and rusk baking and had an antimicrobial effect against *S. aureus*, *B. subtilis*, *E. coli*, and *P. aeruginosa* (at 200 mg/kg of flour).

Cedola et al. [77] fortified bakery products by adding OMWW that had previously been submitted to ultrafiltration and evaluated the chemical composition and sensory quality attributes of the resulting products. Both the bread dough and the spaghetti formulation at a final concentration of 30% w/w were made with ultrafiltered OMWW. The results showed that adding OMWW to bread and pasta improved their chemical composition considerably without compromising their sensory characteristics.

Roila et al. [78] used phenolic extract (250 g/mL and 500 g/mL) to prevent the growth of *Pseudomonas fluorescens* and *Enterobacteriaceae* in mozzarella cheese.

According to Troise et al. [79], spray-dried OMWW decreases the Maillard reaction in a cookie model system, demonstrating that OMWW is a versatile additive capable of

interfering at multiple stages of the Maillard reaction. Multiple mechanisms promoted the control of lysine and asparagine changes in the cookie model system because of the chemical nature of secoiridoids. At three different concentrations, OMWW inhibited the production of protein-bound Amadori compounds, CML dicarbonyls, acrylamide, and 5-hydroxymethylfurfural in cookies at three different concentrations, demonstrating the efficacy of secoiridoids-based functional ingredients to prevent the formation of AGEs.

In a butter formulation, extracts of both OMWW were added at various concentrations (2, 4, 6, and 8 mg/100 g of butter), revealing that the highest concentration confers resistance to oxidative stress during storage at 25°C for 3 months, inhibiting the growth of *S. aureus*, total coliforms, yeast, and molds [80].

The incorporation of OMWW phenolic extract into foodstuffs will contribute to resolving the issue of OMWW's high pollution charge and maximize the extract's utility. The concentration of the added OMWW phenolic extract should be carefully evaluated in order to achieve the intended effect while avoiding unfavorable effects in terms of product stability and sensory impact. Despite the fact that the bitter and pungent sensory note of virgin olive oil is desired—even within a particular range set by consumer acceptance—there is little study on the negative effects of adding an excessive amount of OMWW food products.

Because of their complex matrix and the composition of their bioactive chemicals, the design of functional foodstuffs necessitates research into the stability and interactions of phenolic compounds with other dietary ingredients. More research is also needed on the sensory impact of OMWW. Table 4 lists some examples of the potential use of phenolics recovered from OMWW in food model systems.

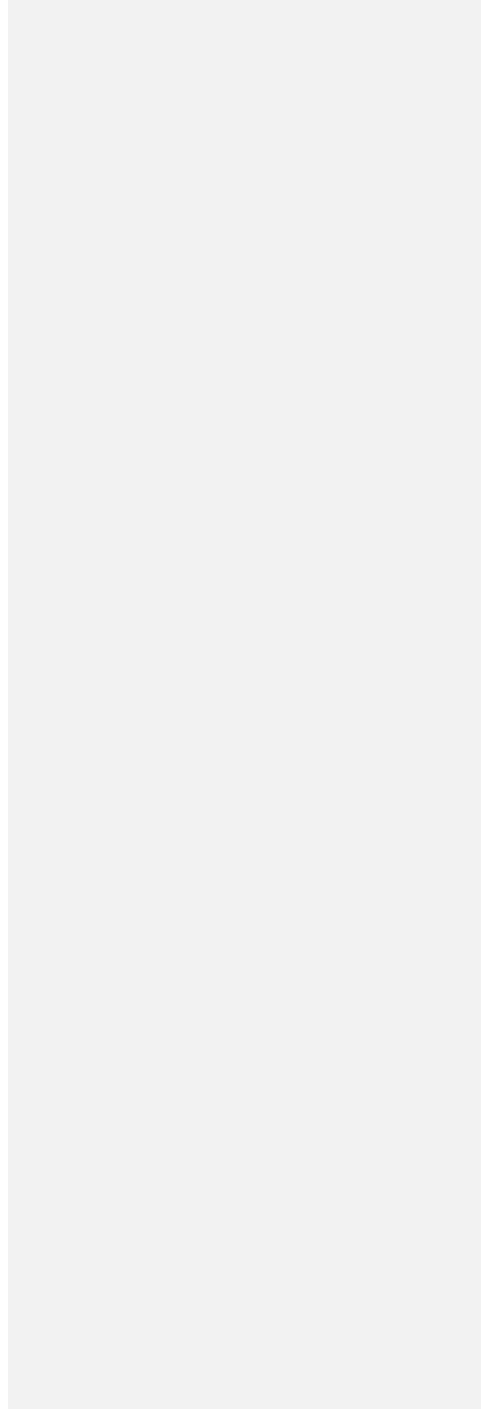
Table 4. Using recovered phenolics in food model systems

Food matrix	Additive	Tested Concentration	Result	Mechanisms	References
Fermented Sauges	Polyphenols	The samples are dipped in 2.5–5% W/V.	Antifungal activity	At the concentrations employed, OMWW has antifungal action that is species dependent.	[69]
White meat burgers	Purified phenolic extract	0.75-1.50 g/kg	Antibacterial activity	Retarding the growth of aerobic mesophilic bacteria	[75]
Fresh breast of chicken	Crude phenolic concentrate	38.6 g/L	Antioxidant and antibacterial activities	TBAR levels that are much lower Growth of <i>Enterobacteriaceae</i> and <i>Pseudomonas</i> spp. is delayed by 2 days.	[75]
Lard	Crude phenolic extract	100-200 ppm	Antioxidant activity	Prevention against oxidation of lard Extension of shelf life	[74]
Milk	Olive oil mill wastewater polyphenol powders	0.1 and 0.05 % W/V	Functional milk	Increasing product stability	[67]

Milk beverage	crude phenolic concentrate	100-200 mg/mL	Fortified beverage	Formulation of fortified beverage	[54]
Butter	crude phenolic extract	2, 4, 6, and 8 mg/100g	Antioxidant activity	Conferring resistance against oxidation	[80]
Cheese	crude phenolic concentrate	250 and 500 µg/mL	Antimicrobial activity	Increasing shelf life	[78]
Cooked and cold stored meat ball	Crude phenolic extract	50-100 mg/L	Antioxidant and activity	Inhibition of lipid peroxidation	[29]
Bread and pasta	Purified phenolic extract	900 ml of OMWW for bread and 30% w/w for pasta	Antioxidant activity and food fortification	Enhancing chemical composition without compromising the sensory properties	[77]
Cooked beef and pork	Crude phenolic extract	50-100 mg/Kg of meat	Antioxidant activity	Improving lipid stability during cooking	[81]
Raw and cooked fresh pork sausages	Crude phenolic extract	750, 1500 mg/kg	Antioxidant activity	Inhibition of lipid oxidation and inhibition of oxidative degradation of cholesterol	[82]
Fermented salami	Purified phenolic extract	0.15%	Antibacterial activity	<i>Listeria monocytogenes</i> growth inhibition after 45 days of fermentation	[75]

Comment [Y6]: Reduce the result and discussion too, its to lengthy

UNDER PEER REVIEW



7. Conclusions and future directives

Environmental concerns related to olive processing wastes have been extensively reported. Given the seriousness of the environmental impact of olive processing wastes, many options for valuing OMWW have been proposed. However, many elements should be considered when choosing the appropriate valuing method, including the overall amount of effluent, investment costs, the industrial or agricultural environment, and most importantly, the legislation. OMWW phenols are far too precious to be depleted or released into the environment. As a result, recovering phenols and repurposing them in various products and markets should be prioritized and more investigated.

Detoxification, production system change, and recovery of important components have been the most popular treatments to date. Traditional techniques such as solvent extractions, membranes, and, more recently, innovative technologies are used to recover phenols from olive mill waste and other food processing by-products. The potential of using components from olive mill waste dates back several years. All of these efforts have resulted in the industrial recovery of phenols from OMWW, as well as their use as natural preservatives and bioactive substances. Various foods, such as vegetable oils, table olives, lard, bakery products, milk, drinks, and meat products, have been investigated. The antioxidant potential of phenolics, as well as their antimicrobial activity, have been demonstrated in all of these applications.

In the food industry, phenolic compounds have the potential to be used as biopreservatives. In fact, phenolic compounds have been thoroughly researched for use in food products to extend the shelf life of foodstuffs. Natural food ingredients have been increasingly popular in recent years. The use of phenolic compounds instead of synthetic antioxidants, such as butylated hydroxyanisole (BHA, E-320), butylated hydroxytoluene (BHT, E-321), tert-butylhydroquinone (TBHQ, E-319), and propyl gallate (PG, E-310), are an interesting alternative to replacing chemical additives in the food matrix. Due to their possible harmful impact on human health, the chemicals are subjected to a maximum concentration limit in foods. The value of natural food preservatives with antioxidant and antibacterial characteristics for food production and consumer health has been largely investigated. Because of the detrimental effects of synthetic chemicals, natural antioxidants and antimicrobials have gained acceptance as replacements. Natural antioxidants and antimicrobials, on the other hand, necessitate more research so that the optimal doses can be properly applied to foods without affecting sensory properties.

While considerable progress has been made in terms of OMWW valorization approaches, there is still much more study to be done. In fact, the stability of phenolic compounds in OMWW-based foodstuffs during processing and storage should be thoroughly investigated. More efforts should be made in an attempt to develop various alternatives to avoid possible inactivation by direct addition. Spraying, coating, and dipping treatments prior to packaging can be considered as viable solutions. A different approach would be to utilize one or more chemicals that could have synergistic effects at lower doses without affecting the food's sensory qualities.

REFERENCES

1. Herrero M, Thornton PKT, Mason-D' Cruz D, Palmer J, Bodirsky BL, Pradhan P, Barrett CB, Benton TG, Hall A, Pikaar I, Bogard JR, Bonnett GD, Bryan BA, Campbell BM, Christensen S, Clark M, Fanzo J, Godde CM, Rockström J. Articulating the effect of food systems innovation on the Sustainable Development Goals. *Lancet Planet. Health* 2021; 5(1):e50-e62.

2. Ilieva RT. Urban Food Systems Strategies: A Promising Tool for Implementing the SDGs in Practice. *Sustainability* 2017; 9(1707):1-35.
3. Campi M, Dueñas M, Fagiolo G. Specialization in food production affects global food security and food systems sustainability. *World Dev.* 2021; 14:105411.
4. Dermeche S, Nadour M, Larroche C, Moulti-Mati F, Michaud P. Olive mill wastes: Biochemical characterizations and valorization strategies. *Process Biochem.* 2013; 48: 1532-1552.
5. Yanguí A, Abderraba M. Towards a high yield recovery of polyphenols from olive mill wastewater on activated carbon coated with milk proteins: Experimental design and antioxidant activity. *Food Chem.* 2018;262:102-109.
6. BCFN data. https://www.barillacfn.com/en/food_sustainability_index/
7. Hamid A, Bibi Z, Ghafoor I, Anwar Z, Khattak JZK. Approach of Bioremediation in Olive Oil and Dairy Industry: A Review. *Res. J. Appl. Sci. Eng. Tech.* 2012;5(6): 2255-2263.
8. Zbakh H, El Abbassi A. Potential use of olive mill wastewater in the preparation of functional beverages: A review. *J. Funct. Foods* 2012;4(1):53-65.
9. Roig A, Cayuela ML, Sanchez-Monedero M. An overview on olive mill wastes and their valorisation methods. *Waste Manag.* 2006;26:960-969.
10. Casa R, D'Annibale A, Pieruccetti F, Stazi S, Sermanni GG, Cascio BL. Reduction of the phenolic components in olive-mill wastewater by an enzymatic treatment and its impact on durum wheat (*Triticum durum* Desf.) germinability. *Chemosphere* 2003;50:959-966.
11. El-Abbassia A, Saadaoui N, Kiai H, Raiti J, Hafidi A. Potential applications of olive mill wastewater as biopesticide for crops protection. *Sci. Total Environ.* 2017;576:10-21.
12. Paraskeva CA, Papadakis VG, Tsarouchi E, Kanellopoulou DG, Koutsoukos PG. Membrane processing for olive mill wastewater fractionation. 2007;213:218-229.
13. Asses N, Ayed L, Bouallagui H, Sayadi S, Hamdi M. Biodegradation of different molecular-mass polyphenols derived from olive mill wastewaters by *Geotrichum candidum*. *Int. Biodeterior. Biodegrad.* 2009;63:407-413.
14. Karpouzias DG, Ntougias S, Iskidou E, Rousidou C, Papadopoulou KK, Zervakis GI, Ehalotis C. Olive mill wastewater affects the structure of soil bacterial communities. *Appl. Soil Ecol.* 2010;45:101-111.
15. El-Abbassia A, Khayet M, Kiai H, Hafidi A, García-Payo MC. Treatment of crude olive mill wastewaters by osmotic distillation and osmotic membrane distillation. *Sep. Purif. Technol.* 2013;104:327-332.
16. Mekki A, Dhoub A, Sayadi S. Review: Effects of olive mill wastewater application on soil properties and plants growth. *Int. J. Recycl. Org. Waste Agric.* 2013; 2(15):1-7.
17. Khoufi S, Louhichi A, Sayadi S. Optimization of anaerobic co-digestion of olive mill wastewater and liquid poultry manure in batch condition and semi-continuous jet-loop reactor. *Bioresour. Technol.* 2015;182:67-74.

18. Rahmanian N, Jafari SM, Galanakis CM. Recovery and removal of phenolic compounds from Olive Mill wastewater. *J. Am. Oil Chem. Soc.* 2014;91:1-18.
19. Roselló-Soto E, Koubaa M, Moubarik A, Lopes RP, Saraiva JA, Boussetta N, Grimi N, Barbaa FJ. Emerging opportunities for the effective valorization of wastes and by-products generated during olive oil production process: Non-conventional methods for the recovery of high-added value compounds. *Trends Food Sci. Technol.* 2015;45(2):296-310.
20. Cassano A, Conidi C, Galanakis CM, Castro-Muñoz R. Recovery of polyphenols from olive mill wastewaters by membrane operations. *In Membrane Technologies for Biorefining* 2016: 163-187.
21. Galanakis CM. The universal recovery strategy. *In Food Waste Recovery Processing Technologies and Industrial Techniques* 2015: 59-81.
22. Goula AM, Gerasopoulos D. Integrated olive mill waste (OMW) processing toward complete by-product recovery of functional components. *In Olives and Olive Oil as Functional Foods: Bioactivity, Chemistry and Processing* 2017: 78-204.
23. Gómez-Caravaca AM, Segura-Carretero A, Martínez-Férez A, Ochando-Pulido JM. Recovery of Phenolic Compounds From Olive Oil Mill Wastewaters by Physicochemical Methodologies. *In Food Bioconversion* 2017:467-489.
24. Kalogerakis N, Politi M, Foteinis S, Chatzisyneon E, Mantzavinos D. Recovery of antioxidants from olive mill wastewaters: A viable solution that promotes their overall sustainable management. *J. Environ. Manage.* 2013;128:749-758.
25. Allouche N, Fki I, Sayadi S. Toward a high yield recovery of antioxidants and purified hydroxytyrosol from olive mill wastewaters. *J. Agric. Food Chem.* 2004;52(2):267-273.
26. Emmons W, Gutteresen C. Isolation of oleuropein aglycon from olive vegetation water. Google Patents. 2005.
28. De Marco E, Savarese M, Paduano A, Sacchi R. Characterization and fractionation of phenolic compounds extracted from olive oil mill wastewaters. *Food Chem.* 2007;104(2):858-867.
29. Galanakis CM, Tornberg E, Gekas V. Clarification of high-added value products from olive mill wastewater. *J. Food Eng.* 2010;99:90-197.
30. Lafka T-A, Lazou AE, Sinanoglou VJ, Lazos ES. Phenolic and antioxidant potential of olive oil mill wastes. *Food Chem.* 2011;125:92-98.
31. Tundis R, Conidi C, Loizzo MR, Sicari V, Cassano A. Olive mill wastewater polyphenol-enriched fractions by integrated membrane process: A promising source of antioxidant, hypolipidemic and hypoglycaemic compounds. *Antioxidants* 2020;9(7):602.
32. Villanova L, Villanova L, Fasiello G, Merendino A. Process for the recovery of tyrosol and hydroxytyrosol from olive mill wastewaters and catalytic oxidation method in order to convert tyrosol in hydroxytyrosol. Google patents 2008.
33. D'Antuono I, Kontogianni VG, Kotsiou K, Linsalata V, Logrieco AF, Tasioula-Margari M, Cardinali A. Polyphenolic characterization of olive mill wastewaters, coming from Italian and Greek olive cultivars, after membrane technology. *Food Res. Int.* 2014;65(PC):301-310.

34. Zagklis DP, Vavouraki AI, Kornaros ME, Paraskeva CA. Purification of olive mill wastewater phenols through membrane filtration and resin adsorption/desorption. *J. Hazard. Mater.* 2015;285:69-76.
35. Bazzarelli F, Piacentini E, Poerio T, Mazzei R, Cassano A, Giorno L. Advances in membrane operations for water purification and biophenols recovery/valorization from OMWWs. *J. Membr. Sci.* 2016;497:402-409.
36. Sedej I, Milczarek R, Wang SC, Sheng R, De Jesús Avena Bustillos R, Dao L, Takeoka G. Membrane-filtered olive mill wastewater: Quality assessment of the dried phenolic-rich fraction. *J. Food Sci.* 2016;81(4):E889–E896.
37. De Almeida MS, Martins RC, Quinta-Ferreira RM, Gando Ferreira LM. Optimization of operating conditions for the valorization of olive mill wastewater using membrane processes. *Environ. Sci. Pollut. Res.* 2018;25(22):21968–2198.
38. Castro-Muñoz R, Cassano A, Conidi C. Membrane-based technologies for meeting the recovery of biologically active compounds from foods and their by-products. *Crit. Rev. Food Sci. Nutr.* 2019;59:2927–2948.
39. Sygouni V, Pantziaros AG, Iakovides IC, Sfetsa E, Bogdou PI, Christoforou EA, Paraskeva CA. Treatment of two-phase olive mill wastewater and recovery of phenolic compounds using membrane technology. *Membranes* 2019;9(2):27.
40. Khoufi S, Aloui F, Sayadi S. Extraction of antioxidants from olive mill wastewater and electro-coagulation of exhausted fraction to reduce its toxicity on anaerobic digestion. *J. Hazard. Mater.* 2008;531-539.
41. Hamza M, Khoufi S, Sayadi S. Fungal enzymes as a powerful tool to release antioxidants from olive mill wastewater. *Food Chem.* 2012;131:1430-1436.
42. Hamza M, Sayadi S. Valorisation of olive mill wastewater by enhancement of natural hydroxytyrosol recover. *Int. J. Food Sci.* 2014;50(3):826-833.
43. Quirantes-Piné R, Lozano-Sánchez J, Herrero M, Ibáñez E, Segura-Carretero A, Fernández-Gutiérrez A. HPLC–ESI–QTOF–MS as a Powerful Analytical Tool for Characterising Phenolic Compounds in Olive-leaf Extracts. *Phytochem. Anal.* 2012;24(3):213-223.
44. Bonacci S, Di Gioia ML, Costanzo P, Maiuolo L, Tallarico S, Nardi M. Natural Deep Eutectic Solvent as Extraction Media for the Main Phenolic Compounds from Olive Oil Processing Wastes. *Antioxidants* 2020;9(513):1-14.
45. De Marco E, Savarese M, Paduano A, Sacchi R. Characterization and fractionation of phenolic compounds extracted from olive oil mill wastewaters. *Food Chem.* 2007;104(2):858-867.
46. Ventura G; Calvano CD, Abbattista R, Bianco M, De Ceglie C, Losito I, Palmisano F, Cataldi TRI. Characterization of bioactive and nutraceutical compounds occurring in olive oil processing wastes. *Rapid Commun. Mass Spectrom.* 2019;33:1670-1681.
47. Klen TJ, Wondra AG, Vrhovšek U, Vodopivec BM. Phenolic Profiling of Olives and Olive Oil Process-Derived Matrices Using UPLC-DAD-ESI-QTOF-HRMS Analysis. *J. Agric. Food Chem.* 2015;63:3859-3872.

48. Obied HK, Bedgood DR, Prenzler PD, Robards K. Chemical screening of olive biophenol extracts by hyphenated liquid chromatography. *Anal. Chim. Acta* 2007;603:176-189.
49. Cardinali A, Pati S, Minervini F, D'Antuono I, Linsalata V, Lattanzio V. Verbascoside, isoverbascoside, and their derivatives recovered from olive mill wastewater as possible food antioxidants. *J. Agric. Food Chem.* 2012;60:1822-1829.
50. Mattonai M; Vinci A, Degano I, Ribechini E, Franceschi M, Modugno F. Olive mill wastewaters: Quantitation of the phenolic content and profiling of elenolic acid derivatives using HPLC-DAD and HPLC/MS² with an embedded polar group stationary phase. *Nat. Prod. Res.* 2019;33:3171-3175.
51. Bazoti FN, Gikas E, Skaltsounis AL, Tsbopoulos, A. Development of a liquid chromatography-electrospray ionization tandem mass spectrometry (LC-ESI MS/MS) method for the quantification of bioactive substances present in olive oil mill wastewaters. *Anal. Chim. Acta* 2006, 573-574, 258-266.
52. Damak N, Allouche N, Hamdi B, Litaudon M, Damak M. New secoiridoid from olive mill wastewater. *Nat. Prod. Res.* 2012;26:125-131.
53. Russo M, Bonaccorsi IL, Cacciola F, Dugo L, De Gara L, Dugo P, Mondello L. Distribution of bioactives in entire mill chain from the drupe to the oil and wastes. *Nat. Prod. Res.* 2020:1-6.
54. Servili M, Esposito S, Veneziani G, Urbani S, Taticchi A, Di Maio I, Selvaggini R, Sordini B, Montedoro GF. Improvement of bioactive phenol content in virgin olive oil with an olive-vegetation water concentrate produced by membrane treatment. *Food Chem.* 2011;124: 1308-1315.
55. Obied HK, Allen MS, Bedgood DR, Prenzler PD, Robards K. Investigation of Australian Olive Mill Waste for Recovery of Biophenols. *J. Agric. Food Chem.* 2005;53:9911-9920.
56. Capasso R, Evidente A, Scognamiglio F. A simple thin layer chromatographic method to detect the main polyphenols occurring in olive oil vegetation waters. *Phytochem. Anal.* 1992;3:270-275.
57. Aramendia MA, Borau V, Garcia I, Jimenez C, Lafont F, Marinas JM, Urbano FJ. Qualitative and quantitative analyses of phenolic compounds by high performance liquid chromatography and detection with atmospheric pressure chemical ionization mass spectrometry. *Rapid Commun. Mass Spectrom.* 1996;10:1585-1590.
58. Della Greca M, Monaco P, Pinto G, Pollio A, Previtiera L, Temussi F. Phytotoxicity of low-molecular-weight phenols from olive mill waste waters. *Bull. Environ. Toxicol.* 2001;67:352-359.
59. Hamdi M. Future prospects and constraints of olive mill wastewaters use and treatment: a review. *Bioprocess Eng.* 1993;8:209-214.
60. Visioli F, Romani A, Mulinacci N, Zarini S, Conte D, Vincieri FF, Galli C. Antioxidant and Other Biological Activities of Olive Mill Waste Waters. *J. Agric. Food Chem.* 1999;47: 3397-3401.
61. Kachouri F, Hamdi M. Enhancement of polyphenols in olive oil by contact with fermented olive mill wastewater by *Lactobacillus plantarum*. *Process Biochem.* 2004;39:841-845.
62. Fki I, Allouche N, Sayadi S. The use of polyphenolic extract; purified hydroxytyrosol and 3,4-dihydroxyphenyl acetic acid from olive mill waste water for the stabilization of refined oils: a potential alternative to synthetic antioxidants. *Food Chem.* 2005;93:197-204.

63. Fernández-Bolaños J, Rodríguez G, Rodríguez R, Guillénand R, Jiménez A. Extraction of interesting organic compounds from olive oil waste. *Grasas y aceites* 2006;57(1):95-106.
64. Servili M; Rizzello CG; Taticchi A, Esposito S, Urbani S, Mazzacane F, Di Maio I, Selvaggini R, Gobetti M, Di Cagno R. Functional milk beverage fortified with phenolic compounds extracted from olive vegetation water, and fermented with c-aminobutyric acid (GABA)-producing and potential probiotic lactic acid bacteria. *Int. J. Food Microbiol.* 2011;147:45-52.
66. Zbakh H, El Abbassi A. Potential use of olive mill wastewater in the preparation of functional beverages: A review. *J. Funct. Foods* 2012;4:53-65.
67. Troise AD, Fiore A, Colantuono A, Kokkinidou S, Peterson DG, Fogliano V. Effect of olive mill wastewater phenol compounds on reactive carbonyl species and Maillard reaction end-products in ultrahigh-temperature-treated milk. *J. Agric. Food Chem.* 2014;62:10092-10100.
68. Fasolato L, Cardazzo B, Balzan S, Carraro L, Taticchi A, Montemurro F, Novelli E. Minimum bactericidal concentration of phenols extracted from oil vegetation water on spoilers, starters and food-borne bacteria. *Ital. J. Food Saf.* 2015;4(2):4519.
69. Chaves-López CC, Serio A, Mazzarrino G, Martuscelli M, Scarpone E, Paparella A. Control of household mycoflora in fermented sausages using phenolic fractions from olive mill wastewaters. *Int. J. Food Microbiol.* 2015;207:49-56.
70. Esposito S, Taticchi A, Di Maio I, Urbani S, Veneziani G, Selvaggini R, Servili, M. Effect of an olive phenolic extract on the quality of vegetable oils during frying. *Food Chem.* 2015;176:184-192.
71. Galanakis CM, Tsatalas P, Charalambous Z, Galanakis IM. Control of microbial growth in bakery products fortified with polyphenols recovered from olive mill wastewater. *Environ. Technol. Innov.* 2018;10:1-15.
72. Araújo M, Pimentel FB, Alves RC, Oliveira MBP. Phenolic compounds from olive mill wastes: Health effects, analytical approach and application as food antioxidants. *Trends Food Sci. Technol.* 2015;45(2):200-211.
73. Chaves-López CC, Serio A, Mazzarrino G, Martuscelli M, Scarpone E, Paparella A. Control of household mycoflora in fermented sausages using phenolic fractions from olive mill wastewaters. *Int. J. Food Microbiol.* 2015, 207, 49-56.
74. De Leonadis A, Macciola V, Lembo G, Aretini A, Nag A. Studies on oxidative stabilisation of lard by natural antioxidants recovered from olive-oil mill wastewater. *Food Chem.* 2007; 100(3):998-1004.
75. Veneziani, G.; Novelli, E.; Esposito, S.; Taticchi, A.; Servili, M. Applications of recovered bioactive compounds in food products. In *Olive Mill Waste*; Academic Press: Cambridge, MA, USA, 2017; pp. 231–253.
76. Galanakis CM, Tsatalas P, Charalambous Z, Galanakis IM. Control of microbial growth in bakery products fortified with polyphenols recovered from olive mill wastewater. *Environ. Technol. Innov.* 2018;10:1-15.
77. Cedola A, Cardinali A, D'Antuono I, Conte A, Del Nobile MA. Cereal foods fortified with by-products from the olive oil industry. *Food Biosci.* 2019;33:100490.

78. Roila R, Valiani A, Ranucci D, Ortenzi R, Servili M, Veneziani G, Branciari R. Antimicrobial efficacy of a polyphenolic extract from olive oil by-product against “Fior di latte” cheese spoilage bacteria. *Int. J. Food Microbiol.* 2019;295:49-53.

79. Troise AD, Colantuono A, Fiore A. Spray-dried olive mill wastewater reduces Maillard reaction in cookies model system. *Food Chem.* 2020;1:126793.

80. Mikdame H, Kharmach E, Mtarfi NE, Alaoui K, Ben Abbou M, Rokni Y, Majbar Z, Taleb M, Rais Z. By-products of olive oil in the service of the deficiency of food antioxidants: The case of butter. *J. Food Qual.* 2020:2020:1-10.

81. De Jong S, Lanari MC. Extracts of olive polyphenols improve lipid stability in cooked beef and pork: Contribution of individual phenolics to the antioxidant activity of the extract. *Food Chem.* 2009;116(4):892-897.

82. Balzan S, Taticchi A, Cardazzo B, Urbani S, Servili M, Di Lecce G, Zabalza IB, Rodriguez Estrada MT, Novelli E, Fasolato L. 2017. *LWT-Food Sci. Technol.* 2017;85(A):89-95.

Comment [Y7]: No paragraph throughout the write up.

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