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Olive Mill Wastewaters: Diversity of the Fatal Product in Olive Oil Industry and its Valorisation as Agronomical Amendment of Poor Soils: A Review

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Abstract: Discharge of olive mill wastewaters is known to have adverse effects on environment. Several scientific treatment technologies were proposed for these effluents. However, they are limited by some economical and technical constraints. Olive mill wastewaters direct agronomical recycling is both classical and innovative alternative for removal of these pollutant effluents. However, this procedure can still not be recommended as safe practice because data is often inconsistent and the physico-chemical characteristics of olive mill wastewaters are not the same depending on several intrinsic and extrinsic aspects. This review compiles and discusses the results of studies conducted worldwide over the last two decades on the effects of olive mill wastewaters as soil amendment. It particularly focuses on OMW associated eco-toxicity and the capability of the biotic and abiotic components of soils to overcome it. Moreover, it explores olive mill wastewaters bio-transformation in soils and also their potential environmental impact.

Key words: Olive mill wastewaters reuse, fertirrigation, soils amendment, abiotic stress, drought, cell stability, soil microorganisms, phenolics, oleuropein, hydroxytyrosol, *Olea europea*, date palm

INTRODUCTION

Because of the increase of olive oil market, olive oil industry is in constant growth worldwide and particularly in the Mediterranean countries (Fig. 1) . In this context, olive oil productive countries are confronted to a serious ecological problem because of the lack of applicable and economical solutions to the problems of the effluents (Olive Mill Wastewaters: OMW) generated by this industry.

In fact, OMW are liquid effluents, brown to reddish brown, acidic and highly loaded in organic matter and mineral elements (Cegarra *et al*., 1996; Paredes *et al*., 1999). Their chemical composition is quite variable. Among the different organic substances found in these effluents: phenolics, sugars, polysaccharides, proteins, lignins and fatty acids (Saiz-Jimenez *et al*., 1987; Aktas *et al*., 2001; Mulinacci *et al*., 2001; De Marco *et al*., 2007; Procida and Ceccon, 2006). Total microflora of OMW is essentially represented by fungi (Zenjari, 2002). Some bacteria from the *Comamonas*, *Ralstonia*, *Pseudomonas* and *Sphingomonas* genus were also isolated from OMW (Di Gioia *et al*., 2002).

The consumption of large quantities of water during the extraction of olive oil leads to a worldwide production of more than 30 million m^3 of OMW every year (Fig. 2) concentrated on a short period from October to January depending on the region. This represents a big ecological problem due to their potential toxicity and their seasonal production.

Although the extraction of olive oil continues to be principally achieved by the three steps procedure, a two phases centrifugation procedure was introduced since the nineteen's, particularly in Spain (Fig. 3). This system presents the advantage of extracting more oil with the consumption of less water and energy. It produces a residue (a humid paste) in less quantities but more loaded in organic matter and of which the disposal is equally considered as a serious ecological problem necessitating the adaptation of procedures studied for OMW (Roig *et al*., 2006).

Because of their acidity, their high salinity and their high organic load (Table 1), it is forbidden, at least in some European countries, to discharge OMW in nature. Recently, different options for OMW detoxification were proposed. Most of these methods aim the reduction of

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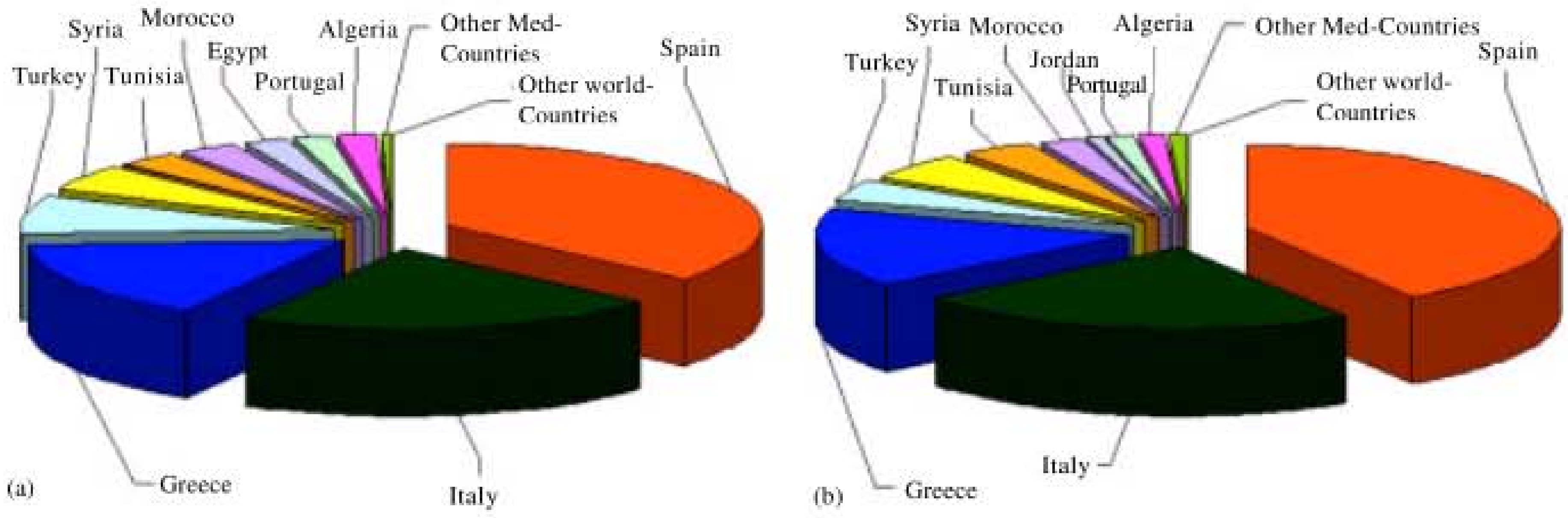

Fig. 1: Olive (a) and olive oil (b) production (in metric tons) repartition among producer countries during the 1999-2003 (FAO, 2004: Food and Agriculture Organization of the United Nations, 2004)

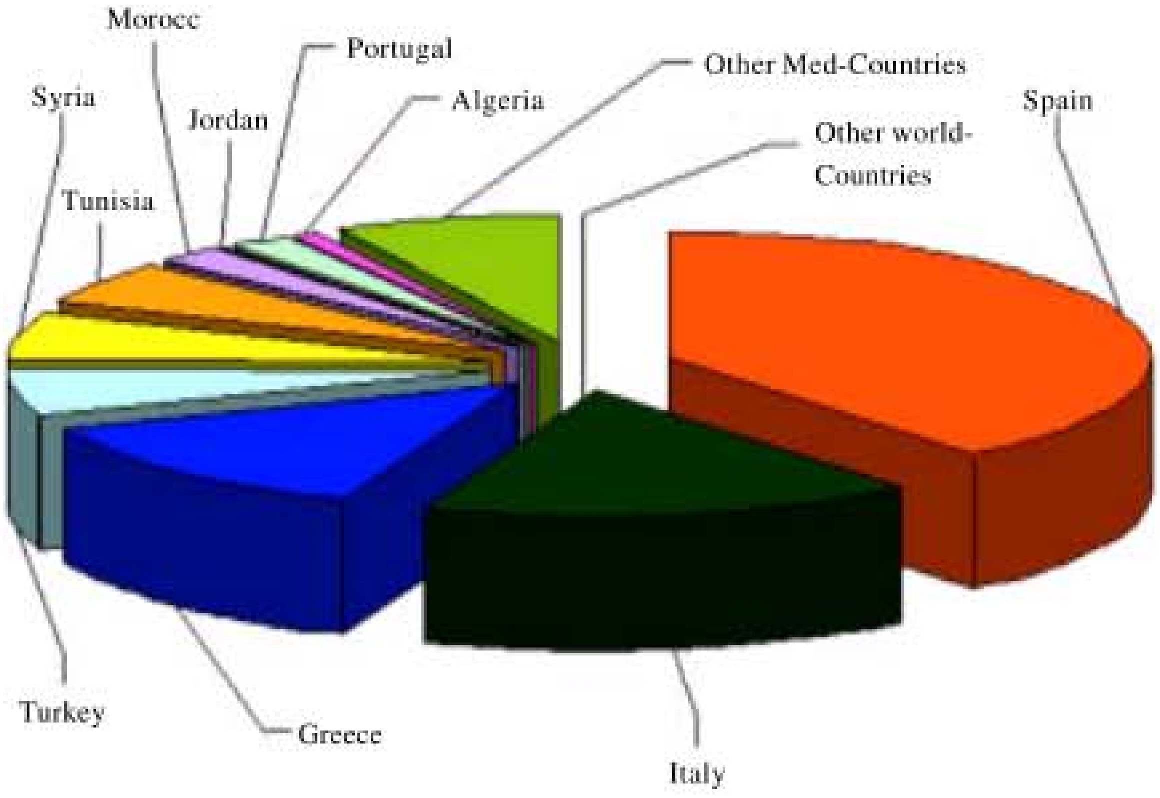

Fig. 2: Olive residue production (in metric tons) repartition among producer countries during the 1999-2003 (FAO, 2004: Food and Agriculture Organization of the United Nations, 2004)

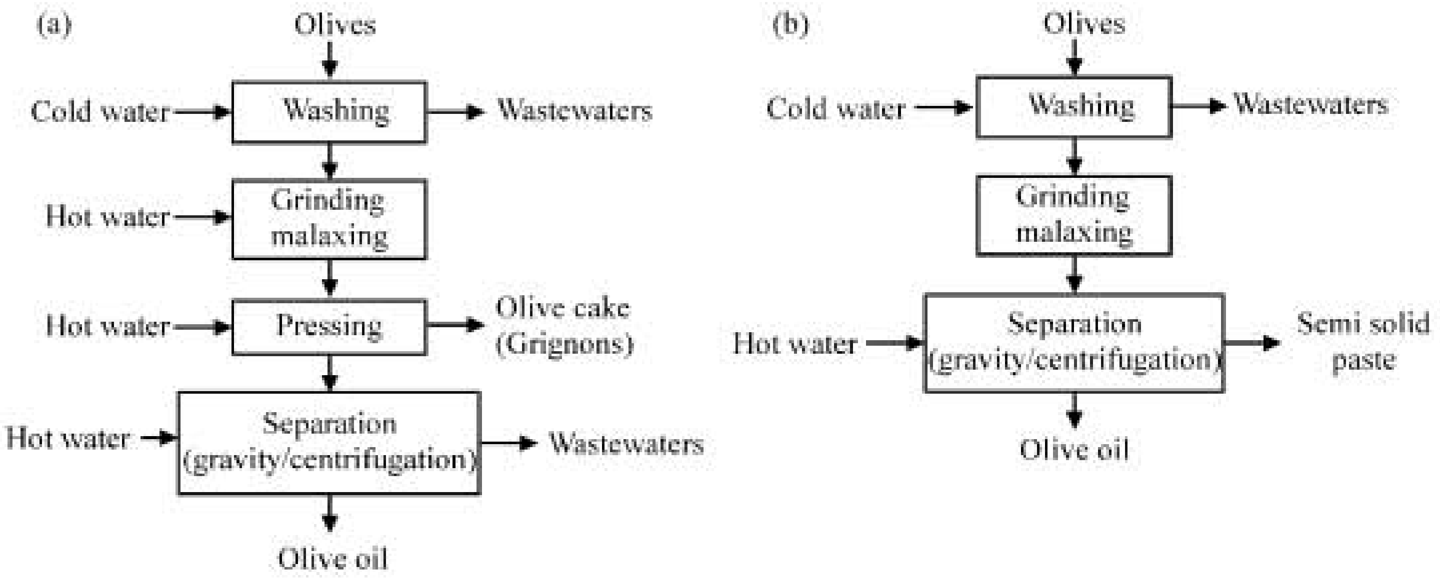

Fig. 3: Differences between olive oil 3-phases (a) and 2-phases (b) production systems

their toxicity by different procedures notably high and low technologies (Ramos-Cormenzana *et al*., 1995; Roig *et al*., 2006; Arvanitoyannis *et al*., 2007) for more complete information about the proposed valorisation/treatments procedures.

However, the application of these procedures remains uncertain for economical and technical reasons (Rozzi and Malpei, 1996) and particularly for the wide variability of OMW composition. For instance, the comparison of OMW phenolic load highlights important quantitative and qualitative differences among samples depending notably on extraction procedures, olives variety and the degree of the olives maturity as well as physiological state (Fig. 4).

Taking into account that the effects of phenolic compounds vary from toxicity to growth stimulation (Kamaya *et al*., 2006) and given that the biodegradation of

Table 1: General physico-chemical characteristics of OMW

	Cegarra *et al.* (1996)	Chatjipavlidis *et al.* (1996)	Montemurro *et al.* (2004)	Zenjari *et al.* (2006)	*Tejada and Gonzalez (2004)	*Alburquerque *et al.* (2006)
OM	nd	nd	nd	nd	150 g kg^{-1}	952.6 g kg^{-1}
pH	nd	5.4	5.11	4.68	3.9	4.97
EC	nd	10 dS cm^{-1}	nd	41 dS cm^{-1}	nd	3.01 dS cm^{-1}
Phenolics	0.86-1.6l g L^{-1}	nd	5.02 g L^{-1}	5.50 g L^{-1}	23 g kg^{-1}	nd
Lipids	0.26-4.79 g L^{-1}	nd	nd	nd	0.32 g kg^{-1}	116.3 g kg^{-1}
N	<800 mg L^{-1}	1360 mg L^{-1}	1.59 mg L^{-1}	2100 mg L^{-1}	10 g kg^{-1}	12.20 g kg^{-1}
P	<0240 mg L^{-1}	423 mg L^{-1}	nd	346 mg L^{-1}	8 g kg^{-1}	0.90 g kg^{-1}
K	24 g L^{-1}	6.10 g L^{-1}	nd	3.93 g L^{-1}	40 g kg^{-1}	15.9 g kg^{-1}

* Two-steps paste; nd: Not determined

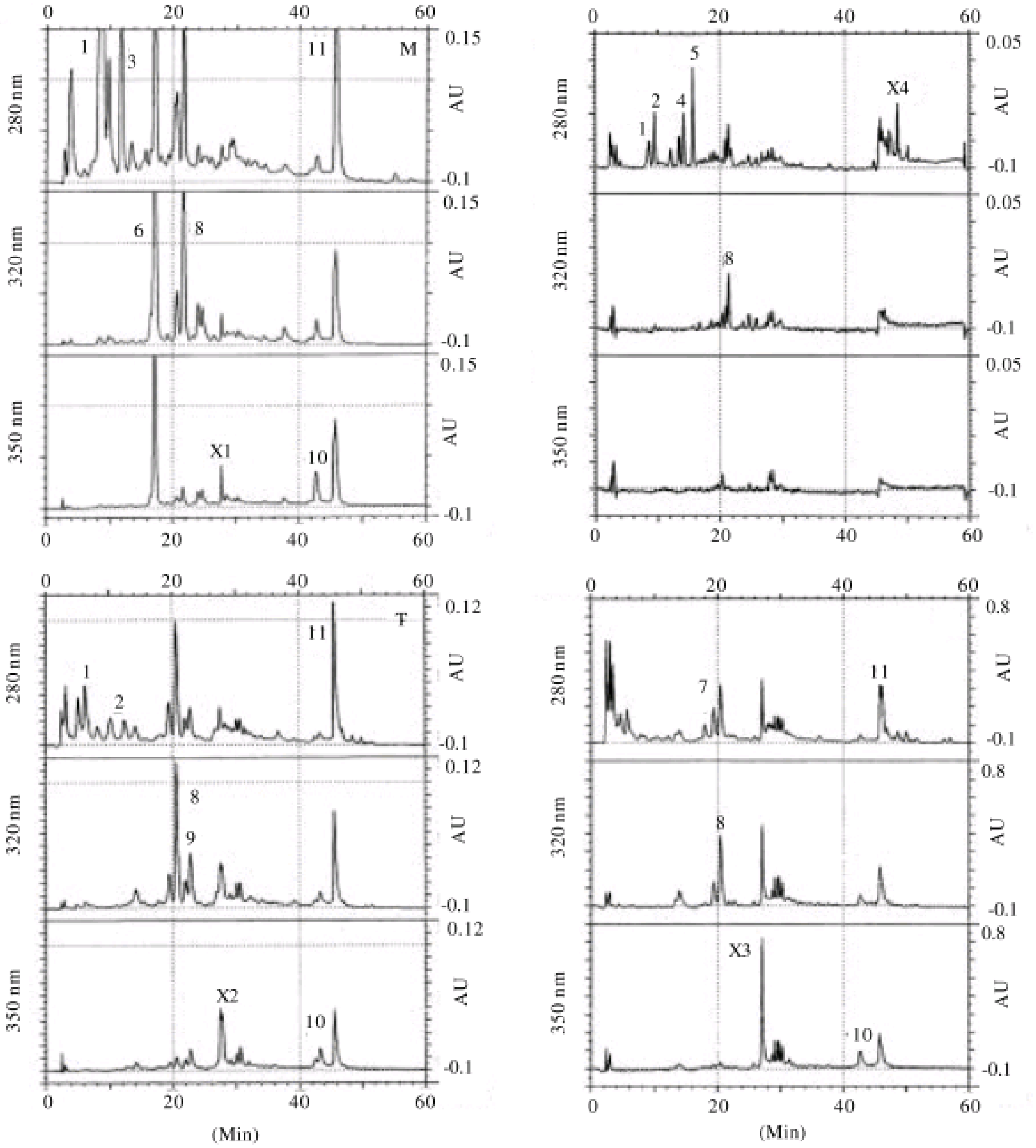

Fig. 4: Comparison of HPLC chromatograms (at 280, 320 and 350 nm) of phenolics in OMW samples from Morrocco (M), Italy (I), Tunisia (T) and Spain (S); 1: Tyrosol, 2 and 4: Protocatechuic acid derivatives, 3: Catechol, 5: Tyrosol derivative, 6: Caffeic acid, 7: Hydroxybenzoic acid, 8: P-coumaric acid, 9: Ferulic acid, 10: Luteolin derivative, 11: Oleuropein derivative, X1, X2 and X3: Non identified flavonoids, X4: Non identified flavanol

phenolics depends closely on their molecular structure (Aresta *et al*., 2003) as well as on their concentration, this heterogeneity of composition could be considered as one of the main limiting factors for the standardization of methods allowing the OMW reuse.

On the other hand, OMW contain an important level of organic matter, considerable quantities of nutrients and are a non negligible water source (Table 1). These properties suggest their recycling as fertilizers as useful and inexpensive alternative to maintain and even to restore the quality of Mediterranean agro-ecosystems (Tomati *et al*., 1995; Cegarra *et al*., 1996; Paredes *et al*., 2005). Nevertheless, the results concerning the agronomic valorisation of OMW are sometimes diverging or even contradictory. Therefore, this study reviews and discusses the more important results obtained on the subject.

EFFECTS OF OMW APPLICATION ON SOIL PROPERTIES

Organic wastes use as agricultural amendment is an effective strategy to regenerate degraded soils and an economical alternative that furnishes a locally available source of nutrients (Kowaljow and Mazzarino, 2007). Given that the majority of olive oil producer countries in the Mediterranean basin are exposed to the problem of desertification (Laraus, 2004), the use of OMW as agricultural amendment was proposed as one of the more suitable methods to solve the problem of their disposal and to reuse water for, at least, irrigation of cultures. In fact, soil amendment with OMW has for result the increase of organic carbon content and the improvement of the structure and nutrient levels and therefore the fertility of soil (Cabrera *et al*., 1996), the limitation of water loss by evaporation (Mellouli *et al*., 1998) and the reduction of soil porosity which suggests their possible use to reduce pesticides leaching (Cox *et al*., 1997).

These whole properties suggest strongly OMW use for degraded soils restoration in arid and semi-arid climates as well as the preservation of water tables. Nevertheless, several studies report soil properties deterioration as a result of OMW application. In this sense, it has been found by Gonzaflez-Vila *et al*. (1995) the increase of the lipid fraction in soil upper layer. Such accumulation of hydrophobic groups is shown to induce soil water-repellence (Tarchitzky *et al*., 2007). Besides, the content of phenolic compounds is increased after OMW application along with leaching in inferior layers (Sierra *et al*., 2001; Zenjari and Nejmeddine, 2001; Mekki *et al*., 2007). Moreover, increase in soil salinity along with sodium ions leaching is observed after OMW application (Cabrera *et al*., 1996; Sierra *et al*., 2001; Zenjari and Nejmeddine, 2001). OMW supply also implies mobilization of metals previously retained by soil notably zinc and copper (Madrid and Diaz-Barrientos, 1998). In several studies, a transient decrease or even an inhibition of major microbial enzymatic activities in the organic matter cycles notably the phosphatase and the β-glucosidase is reported (Piotrowska *et al*., 2006). However, an important feature of soil response to OMW is that modifications induced by OMW incorporation are generally temporary. Several studies showed that inhibitory effects were found principally in the first weeks after amendment and that soil was able to find again, at least partially, its initial state after an exposition period (Benitez *et al*., 2004; Piotrowska *et al*., 2006; Saadi *et al*., 2007). This soil feature depends closely on OMW applied amounts (Piotrowska *et al*., 2006) and its intrinsic physico-chemical and biological properties. In fact, the incorporation in soil of OMW-organic matter readily metabolizable activates soil microbial degradation. However, at high rate, a great accumulation of toxic compounds inhibit microbial growth and then slow down the microbial degradation process.

Concerning OMW impact on soil pH, controversial effects were obtained. Thus, in contrast to a decrease of soil pH due to OMW amendment found by Morisot and Tournier (1986) and Zenjari and Nejmeddine (2001), it was showed by Sierra *et al*. (2001) that the acidity of OMW is compensated by the alkalinity of soil carbonates. In fact, these become bicarbonates, moved and accumulated in the deeper horizons. In an other study, it was found that OMW-amended soil shows pH increase probably due to sodium ion brought by OMW that generates $NaCO_3$ of more alkaline hydrolysis than soil initial $CaCO_3$ (Sierra *et al*., 2007). This suggests a crucial effect of soil native properties on OMW-soil interactions. Edaphic properties must then be taken in consideration when spreading OMW. In this sense, phenolics' leaching is also shown to be negligible in soils rich in carbonate and clay materials (Sierra *et al*., 2007). Also, nitrogen mineralization is more relevant in soils with high clay content (Cabrera *et al*., 2005).

In the other hand, Saiz-Jimenez *et al*. (1987) report soil enrichment in humic-like substances due to OMW. A partial incorporation of components from OMW in soil humic fraction is confirmed by Gonzaflez-Vila *et al*. (1995). Besides, Brunetti *et al*. (2007) report compositional, structural and functional changes in characteristics of soil humic substances due to partial incorporation of humic-like components from applied olive mill wastes. The effects of such modifications on soil characteristics are a little, if even, studied and increasing of non-humified organic matter fraction by olive mill wastes could lead to a decrease of soil organic matter stability (López-Piñeiro *et al*., 2007). In fact, although mineralization procedures trend to attenuate changes in soil humic substances (Senesi *et al*., 2007), humic substances, particularly phenolics, are shown to exert allelopathic effects that have drastic consequences on plants growth (Pellissier, 1994).

OMW LAND APPLICATION EFFECTS ON MICROFLORA

The impact of OMW on soil microflora could be considered according to two general points of view: (1) the enrichment with a readily metabolizable carbon source and (2) the incorporation of inhibiting compounds. It is generally recognized that the toxicity of these effluents is due to their phenolic compounds, principally monomeric ones (Capasso *et al*., 1995; Perez *et al*., 1992). Other compounds, notably fatty acids, contribute also to the toxic potential of OMW (Gonzalez *et al*., 1990). Detrimental effects of soil salinity toward micro-organisms were also reported by Wichern *et al*. (2006).

The response of microflora to OMW is the result of the interaction between positive and negative factors that depends tightly on OMW concentration and period after application. Thus, the application of raw OMW induces an initial increase in the respiration and the number of unities forming colonies in most of microflora groups (Paredes *et al*., 1987; Mekki *et al*., 2006a). However, only weak number of total viable microflora is observed with high dose application (Mekki *et al*., 2006a).

In addition, the modification of soil conditions induces changes in the distribution of microbial populations which could lead to shift in nutrient cycle patterns (Mechri *et al*., 2007a). Thus, OMW application enhances copiotrophs predominance (Kotsou *et al*., 2004). In this sense, Hu *et al*. (1999) showed that high carbon availability may inhibit oligotrophs in natural soils. OMW application is equally in favour of fungi populations (Mekki *et al*., 2006a). In fact, fungi are known by their substantial resistance to recalcitrant substances notably OMW (Sampedro *et al*., 2004; Aranda *et al*., 2007). The increase of the C/N ratio (Paredes *et al*., 1987) as well as increased salinity (Wichern *et al*., 2006) may also lead to a shift towards fungi. Furthermore, modification in soil fungal homeostasis was also reported following OMW supply (Tardioli *et al*., 1997).

A selective toxicity of OMW with respect to some micro-organisms was also revealed (Paredes *et al*., 1986). This species-specific toxicity may have important effects on soil fertility. In fact, Mekki *et al*. (2006a) showed the toxicity of OMW toward nitrifying bacteria that play a critical role in nitrogen cycle (Oved *et al*., 2001; Mendum and Hirsch, 2002). Also, Paredes *et al*. (1987) showed an increase in the number of denitrifiers due to OMW application, which could also contribute to soil nitrogen loss. In the other hand, due to their high carbon content as compared to their nitrogen content, OMW offer convenient conditions for the growth of a nitrogen fixating bacterial consortium notably *Azotobacter* (Balis *et al*., 1996). The increase in the phosphate solubilisers levels is equally observed (Paredes *et al*., 1987).

OMW FERTILIZATION EFFECTS ON CROPS

Several researchers investigated OMW application to soil and tested their impacts as organic fertilizer showing positive or negative effects. Considering their enriching effect on the organic matter, the ability of OMW application to improve soil fertility seems very profitable (Tomati *et al*., 1996; Cegarra *et al*., 1996). Nenetheless, the results of different studies are often contradictory which does not allow a legal and safe fertilization method sitting (Table 2). In fact, although application of raw OMW was proved profitable with few species (Montemurro *et al*., 2004; Hanifi and El Hadrami, 2008a, b), raw OMW are often associated with phytotoxic effects (Ben Rouina *et al*., 1999; Rinaldi *et al*., 2003; El-Hadrami *et al*., 2004; Mekki *et al*., 2006b).

Several researchers attribute OMW toxicity to their phenolic compounds (Capasso *et al*., 1995; Aliotta *et al*., 2000). In fact, it was described that some phenolic compounds such as p-coumaric acid and ferulic acid present phytotoxic effects toward plants consisting in the disturbance of their physiological processes (Li *et al*., 1993; Yang *et al*., 2002, 2004). Besides, soil acidification is also known to affect negatively crops by damaging roots and involving nutrients depletion (Tan *et al*., 1992). In the same manner, salt is widely known for its toxicity due to both direct effects of Na^+ and Cl^- ions and to their osmotic effect (Mahajan and Tuteja, 2005; Parida and Das, 2005). Salt could also alter plants nutrition (Cabrera *et al*., 1996; Murillo *et al*., 2000) and deteriorate soil physical conditions. Salinized soil becomes hard and compact reducing thus water and air infiltration and water holding capacity (Choudhary *et al*., 2006; Walker and Bernal, 2008). Salt presents a serious problem taking into account its anarchic use for olive conservation notably in small traditional mills (Fig. 5). In addition, several other components, such as lipids and humic-like substances can also contribute to the toxicity of OMW (Linares *et al*., 2003).

Table 2: Effect of OMW amendment on some cultivable crops

Crops studied	OMW amount	Soil properties	Effects of OMW amendment	Reference
Maize (*Zea mays*)	30 m^3 ha^{-1} (three-phases discontinuous system)	Not determined	Growth amelioration, sensitivity of first development stages	Hanifi and El Hadrami (2008b)
Date palm (*Phoenix dactylifera*)	150 m^3 ha^{-1} $year^{-1}$ (three-phases discontinuous system)	Calcareous soil	Growth and yield amelioration	Hanifi and El Hadrami (2008a)
Tomato (*Lycopersicon esculentum*), chickpea (*Cicer arietinum*), bean (*Vicia faba*), wheat (*Triticum durum*) and barley (*Hordeum vulgare*)	30 m^3 ha^{-1} (three-phases discontinuous system)	Not determined	Germination and growth reduction, leaf necrosis	Mekki *et al*. (2006b)
Maize, wheat, chickpea and tomato	25 and 50% of soil holding capacity (three-phases discontinuous system)	Sand peat (2:1, v/v)	Significant reduction of yield for all crops, chlorophyll content reduction	El Hadrami *et al.* (2004)
Rye-grass	80 m^3 ha^{-1} (OWW obtained using the centrifuge method)	Not determined	Increase of growth parameters by 18.2 to 41.1%; temporary reduction of leaf greenness	Montemurro *et al.* (2004)
Durum wheat	50 m^3 ha^{-1} (OWW obtained using the centrifuge method)	Silty-loam soil, permeable, shallow (0.6 m depth) and with good native fertility	No significant difference as regard to yield some necrosis of the leaves and caused a slow emission of secondary stems general wheat tolerance	Rinaldi *et al*. (2003)
Olive tree	0, 2, 4, 6 and 8 L pot^{-1} containing 16 kg of soil	Sandy soil	Phytotoxicity and plants death at high doses (6 and 8 L pot^{-1})	Ben Rouina *et al.* (1999)

Fig. 5: Olives' conservation using sodium chloride in a traditional mill

In the other hand, OMW fate in soil is complex and could be associated with nutrients depletion. In fact, nutrients availability in OMW amended soil is contradictory and depends on nutrient nature and time after OMW amendment (Piotrowska *et al*., 2006). Thus, a transient immobilisation of soil nitrate was observed after OMW amendment (Sierra *et al*., 2007). Besides, the enrichment in K^+ and Na^+ impairs the composition of the exchange complex and induces calcium and magnesium leaching (Cabrera *et al*., 1996). What's more, OMW are able to mobilize heavy metals previously retained by soil notably zinc and copper (Madrid and Barrientos-Diaz, 1998). OMW can also affect plants nutrition indirectly. In fact, OMW offer adequate conditions for the growth of phosphate solubilisers (Paredes *et al*., 1987) and nitrogen fixating bacteria (Balis *et al*., 1996) allowing the improvement of soil contents in this essential elements. On the other hand, Mekki *et al*. (2006a) showed OMW toxicity toward nitrifying bacteria responsible of the first step in the nitrification in which the ammonia is transformed in nitrate via the nitrite. Such inhibition, added to other losses notably immobilization and volatilization (Sierra *et al*., 2007; Paredes *et al*., 1987) could have drastic impact on crops. In addition, organic acids contained in OMW were used successfully to solubilize rock phosphate. However, this effect was transitory and extractable soil phosphate was later decreased (Mechri *et al*., 2007b).

These potential risks of OMW brought some countries to establish laws limiting the maximum OMW quantity tolerated in agricultural application to 30 m^3 in Catalonia and 80 and 50 m^3 for centrifugation and press respectively in Italy and Portugal (Sierra *et al*., 2007). These application rates are negligible in term of irrigation given that water irrigation rate is about 1 m^3 m^{-2} from which about 50% is brought by rainfall. Besides, considering the strong sensitivity of some development stages notably the first development ones, it is recommended to avoid OMW spreading on agricultural soils during these periods (Hanifi and El Hadrami, 2008b). However, it must be brought in mind the possibility that some phytotoxic compounds could be lately released by the degradation of olive mill wastes in soil (Nastri *et al*., 2006).

OLIVE MILL WASTEWATERS: A POTENTIAL BIO-PESTICIDE

Increasing interest is recently given to the use of natural products as bio-control agents for substituting chemical products. In this sense, OMW effects against

Table 3: OMW possible effects on the interaction plant-pathogen

Variables	Factor influencing OMW impact on plant-pathogens interaction	Resulting effect	Reference
Direct effects	OMW phenolics	Inhibition / impairing of pathogen growth	Capasso *et al.* (1995), Kistner *et al.* (2004), Soler-Rivas *et al.* (2006)
	OMW indigenous microflora	Antagonisms toward pathogens	Yangui *et al.* (2008)
	Enhancement of pathogen growth coupled to phytotoxicity	Aggravation of disease incidence	Bonanomi *et al.* (2006)
	*Not indentified thermo-resistant factor	Induction of systemic acquired resistance and immunisation of plants to subsequent foliar attack	Kavroulakis *et al.* (2005)
Indirect effect	Enrichment in organic molecule readily metabolizable	Creation of environment unfavourable to pathogen and probably the improvement of soil microflora population suppressive against *Rhizoctonia solani*	Kotsou *et al.* (2004)

*Olive residue compost

weeds was tested on the orobanche. The use of high doses was effective with regard to the reduction of orobanche stems numbers and dry matter. In addition, the stems emergence of orobanche to soil surface was delayed by OMW application. These effects were equally accompanied by an improvement of bean culture biomass and yield (Saffour *et al*., 2004). Besides, OMW use against weeds in corn, wheat and sunflower cultures showed an important reduction of weeds cover but some toxic effect were observed particularly in term of corn and sunflower germination (Boz *et al*., 2003).

Moreover, the addition of these effluents as a supplement to the growth substrate of *Pleurotus pulmonarius* was able to impair the growth of *Pseudomonas tolaasii*, the pathogen of this mushroom, but without completely preventing its development (Soler-Rivas *et al*., 2006). In this same sense, the amendment of the nutritive solution of hydroponic cultivated tomatoes by OMW eliminates bacterial growth (Kistner *et al*., 2004). Also, OMW were used as bio-control agent against the damping-off caused by *Rhizoctonia solani* and *Fusarium solani* (Yangui *et al*., 2008). Besides, the aqueous extract of two-steps paste was successfully employed in a sustainable farming system to inhibit/eliminate *Fusarium oxysporum lycopersici* as well as other pathogenic fungi (Ranalli *et al*., 2004).

In fact, a strong antimicrobial activity was credited to these effluents due to their phenolics notably catechol, methylcatechol and hydroxytyrosol (Perez *et al*., 1992; Capasso *et al*., 1995; Soler-Rivas *et al*., 2006). OMW indigenous bacteria are also proved to exert antagonist effect against *Rhizoctonia solani* and *Fusarium solani* (Yangui *et al*., 2008). In addition, OMW organic matter plays a crucial role in the plants-pathogens interactions outcome. Kotsou *et al*. (2004) showed that soil treatment by OMW created an environment that improves selectively copiotrophs communities which would be the probable cause of soil suppressiveness against *Rhizoctonia solani*. Nevertheless, other study showed that the application of olive mill dry residue affects favourably pathogenic fungi by furnishing organic constituent easily metabolizable and concomitantly affect negatively plants by phytotoxic effects; this further increases fungal diseases incidence (Bonanomi *et al*., 2006) (Table 3).

OMW FATE IN SOIL AND ENVIRONMENTAL CONSEQUENCES

OMW decomposition in soil is mediated by several factors namely biotic and abiotic ones and controlled by soil properties, effluent composition and climatic conditions. Generally, OMW mineralization in nature is slow and difficult because of their low nitrogen content (Paredes *et al*., 1987) and their toxicity usually attributed to their phenolic compounds either monomeric (Cappasso *et al*., 1992) or polymeric ones (Sayadi *et al*., 2000). The detrimental effects of these compounds is ascribed to their direct toxicity (Capasso *et al*., 1995; Soler-Rivas *et al*., 2006) and the inactivation of microbial exo-enzymes concerned in degradation as well as the combination with organic molecules to produce recalcitrant complexes (Benoit and Starkey, 1968; Lewis and Starkey, 1968; Bradley *et al*., 2000; Kanerva *et al*., 2006).

These whole effects and others could have drastic repercussions on soil fertility which affect negatively plants nutrition and micro-organisms activity. In fact, a transient immobilisation of soil nitrate was observed and mainly attributed to the high C/N content of OMW but also to the activation of soil microbial activities that lead to an increase of nitrogenated soluble molecules demand (Bengtson and Bengtsson, 2005). Likewise, beside a transitory and medium term (4 months) increase in available phosphorus due to OMW application (Sierra *et al*., 2007), it was observed that soil extractable phosphorus decrease presumably due to soil insolubilization in the form of Ca-P or the inhibition of microbial activities responsible of phosphate release (Mechri *et al*., 2007b).

Table 4: OMW phenolics possible transformations in soil

Factor influencing OMW phenolics	Resulting effect	Reference
Soil minerals (Mn, Fe)	Polymerisation to less toxic molecules	Colarieti *et al*. (2002), Colarieti *et al*. (2006), Greco *et al*. (2006)
Soil fungi (*Fusarium*)	Transformation of monomeric phenolics to lignin	Sampedro *et al*. (2004, 2005)
Not identified	release of phytotoxic compounds by the degradation of olive mill wastes in soil	Nastri *et al*. (2006)
Not indentified	the generation of a toxic phenolic fraction	Mekki *et al*. (2007)
Soil fungus (*Aspergilus flavus*)	Degradation of oxidized OMW	Kachouri *et al*. (2005)

In the other hand, many studies demonstrate that toxicity in OMW-amended soil tends to disappear after few months (Saadi *et al*., 2007). Although phenolics are recognized to be major factor influencing OMW toxicity, few studies interested particularly to OMW-phenolics transformations in soil (Table 4). Actually, three phenomena govern the fate of these compounds in soil: degradation, polymerisation and leaching. Their adsorption by soil was also reported (Sierra *et al*., 2007). Slight migration in depth of phenolic compounds is showed (Mekki *et al*., 2007); but this migration represents an eminent risk for the water table particularly in soil conditions that favour leaching (Spandre and Dellomonaco, 1996). In the other hand, soil biotic and abiotc components exert important capacities as natural catalyst to promote the oxidation and the polymerisation of OMW phenolic compounds in less toxic products (Colarieti *et al*., 2006; Greco *et al*., 2006). Actually, OMW monomeric phenolics are proved to polymerize under the action of soil minerals notably Mn and Fe (Colarieti *et al*., 2002). Such minerals are commun soil constituents that catalyse several reactions of organic matter humification. Some soil micro-organisms, notably fungi, are also known for their aptitude to diminish OMW toxicity notably by transforming monomeric phenolics to lignin (Sampedro *et al*., 2004, 2005; Aranda *et al*., 2007). This capacity is principally credited to the enzymatic activities produced by these fungi notably the peroxidases and laccases that polymerize monomeric phenolics (Tsioulpas *et al*., 2002; Casa *et al*., 2003; D'Annibale *et al*., 2004). Other degradation mechanisms involving hydrogen peroxide, oxalic acid and siderophores could be in relation with this action of fungi (Tsioulpas *et al*., 2002; D'Annibale *et al*., 2004; Aranda *et al*., 2007).

These polymerized products show similar properties to humic substances (Dec et Bollag, 1990; Casa *et al*., 2003). It seems that this fraction could serve as precursor for the formation of humic acids in soil (Saiz-Jimenez *et al*., 1987). This ecosystem aptitude to transform and reduce phenolic load of OMW would explains the restoration of soils after application of OMW (Benitez *et al*., 2004; Saadi *et al*., 2007). In fact, phenolics toxicity was correlated to their lipophilicity (Wang *et al*., 2001) and structure (Aresta *et al*., 2003). It was suggested that the polymerisation of phenolics reduces their aptitude to cross the cytoplasmic membrane (Casa *et al*., 2003).

However, the possibility that some fungi diminish monomeric phenolics while transforming them in polymers of similar toxicity cannot be excluded (Tsioulpas *et al*., 2002). In this sense, the toxicity of olive residues toward tomato and soy plants did not diminish in spite of a decrease in phenolic compounds (Sampedro *et al*., 2004). Additionally, the generation of a toxic phenolic fraction after OMW land application is detected by Mekki *et al*. (2007).

In the other hand, although high molecular weight phenolics are recognized as being poorly biodegradable, the degradation of high molecular-mass phenolics notably lignins is equally possible by some microorganisms (Vargas-Garcýa *et al*., 2007). However, fungi ability to depolymerise these phenolics seems to be limited. The increase of the molecular-mass of phenolics leads to decrease in levels of depolymerization by fungal cultures (Sayadi *et al*., 2000). The nature and the manner in which the products of this degradation influence the living organisms are not understood. In fact, the toxicity of OMW is often attributed to their monomeric phenolics and the degradation of the polymerized fractions to monomers could lead to toxicity.

CONCLUSION

Generally, OMW beneficial effects are due to their high concentration in nutrients, especially potassium, while, the negative effects are associated with their high content in mineral salt, their acidity and the presence of toxic compounds especially phenolics. Although the organic fraction of OMW seems to be gradually degraded and incorporated in soil humic substances, salt accumulation remains a constraint in OMW agronomical valorisation essentially in dry lands where the high evaporation leads to enhance the salinization of soils. OMW spreading must take into account the cumulative effect of salt that would lead in long term to soil degradation. Technical and scientific interventions are still needed for the safe OMW re-use. A more effective extraction of the fatty acids before soil application would contribute to the decrease of OMW toxic effects. An upstream control of salt use would be critical to prevent land secondary salinity. Besides, use of halophytes notably as associated crops or in agro-forestry systems could contribute to reducing this OMW effect. Besides, more interest should be given to the study of soil transformations of OMW toxic fraction and eventual long-term accumulation of residual toxic molecules.

Sustainable OMW agronomical reuse may have a high potential to improve some crop production with minimized adverse environmental effects. This will require a global approach taking into account olive processing, soil properties and crop management.

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