AGRONOMIC APPLICATION OF OLIVE MILL WASTE WATER: SHORT-TERM EFFECT ON SOIL CHEMICAL PROPERTIES AND BARLEY PERFORMANCE UNDER SEMIARID MEDITERRANEAN CONDITIONS

Raja Dakhli ^{(1)*}, Houcine Khatteli ⁽¹⁾, Lamouri Ridha ⁽²⁾, Houcine Taamallah ⁽¹⁾

(1) Laboratory of Eremology and Combating Desertification

 (2) Laboratory of Aridocultures
 Institute of Arid Areas, Médénine, Tunisia
 * Corresponding author E.mail: rajaedakhli@yahoo.fr

Abstract

The extraction of oil produces olive mill waste water "OMW" which has a very high polluting power resulting in high levels of COD (chemical oxygen demand), high salinity and a strong phenolic compounds causing environmental pollution. Thus, the exploitation of this waste without preliminary treatment is very limited considering the toxic effect on soil and plants. The search for new technologies or processes for recovery of the effluent is necessary. The spreading of OMWW in the sandy soil in arid conditions of Southern Tunisia is a potential alternative for this purpose. The identification of a recovery method of "OMW" as a fertilizer in agriculture is an initiative with both agronomic and environmental interest. In this study, the spreading of 15, 30 and 45 m³/ha for three consecutive years was tested in the presence of irrigated barley crop in order to assess the impact of the incorporation of these effluent on soil chemical and biological characteristics and plant behaviour. The results of the study showed that spreading amounts from 15 to 45 m³/ha for three consecutive years induced a considerable improvement of soil fertility. The pH and soil phosphorus content remain stable during the three years of study while the soil salinity increased for the 45 m³/ha treatment where it exceeded 6 dS /m. In addition, all the components of barley yield, except 1000 grain weight, were negatively affected by "OMW", in particular yield plots that received higher doses as 30 m³/ha and 45 m³/ ha. Obviously, the straw, spikels and grain yield, are negatively affected with relatively different degrees depending on the dose applied but also on the cumulative effect of successive applications during the three years of study.

Keywords: OMW, soil, Barley crop, yield.

Introduction

The cultivation of olive trees and the production and use of olive oil has been a well-known and established practice in the Mediterranean region for more than 7000 years. The consumption of olive oil is rapidly increasing worldwide.

Olive oil production is highly important for the economy, ecology and social life of many Mediterranean countries such as Spain, Italy, Greece and especially Tunisia, which accounts for about 8 % of olive oil world production (Doula et al., 2012; Dakhli, 2013a,b).

However, the extraction process generates a high amount of olive mill wastewater (OMW), with an average annual production of 700.000 T/an (Taamallah, 2007).

Enormous quantities produced annualy, reflect the importance of the olive oil sector in the Mediterranean and consequently the magnitude of the problems related to the disposal of large amounts of wastes produced during olive oil production.

OMW is an environmental problem. This sludge presents high salinity (16 ds/m), high values of chemical oxygen demand COD between 40 and 200 g/l (Saadi et al., 2007; Dakhli, 2015), phytotoxic properties and resistance to biodegradation due to the enormous supply of polyphenol and organic substance (10 to 12 g) (Rinaldi et al., 2003; Mekki et al., 2007).

Some OMW characteristics are favourable since this effluent is rich in water, organic substance, and in mineral nutrients, such as nitrogen, phosphorous, potassium, iron and magnesium (Lesage-Meessen et al., 2001).

Consequently, increasing attention has been given to the spread of OMW on agricultural lands as organic fertilizer and to recycle both the organic substance and the nutritive elements in the soil crop system (Komilis et al., 2005; Mekki et al., 2006; Mechri et al., 2008).

Some studies indicated a positive effect of OMW on physical, chemical and microbiological properties of soil. Pagliai et al. (2001) reported that OMW spreading had beneficial effects on topsoil.

Conversely, Mekki et al. (2006) found that OMW caused negative changes in microbial soil properties, decreasing or inhibiting microflora growth. Zenjari and Nejmeddine (2001) established that OMW spreading compromised soil fertility, altering physical and chemical soil properties.

Recent studies have evaluated the long-term effect of OMW disposal on soil properties. Chaari et al. (2015); Kavvadias et al. (2014) observed a positive soil effect due to the supply of organic matter (OM) and macronutrients, but when a high dose (200 m³.ha⁻¹.year⁻¹) was provided for nine successive years, a cumulative effect of soil salinization became evident. Kavvadias et al. (2014) highlighted the effect on soil after 11 years of disposal of raw OMW. They reported that the uncontrolled and elevated disposal resulted in a source of pollution mainly on the soil surface. That is why it was necessary to establish soil quality standards in order to identify soils well-suited to OMW spreading.

Current research assesses the chemical and biological properties of soils after treatment with raw OMW. A study was performed on two different soils, located in Avellino province (Italy), that have been irrigated for 11 years with a low volume of OMW (30 m³. ha⁻¹. year⁻¹), corresponding to the annual OMW production of the factory mill and soils were analyzed after the last spreading performed in October 2013.

In this prespective, several studies indicated beneficial effects of OMW on soil fertility such as nutrient availability for crop growth (Casa et al., 2003; Cereti et al., 2004; Paredes et al., 2005; Ayoub et al., (2014).

In this context, several works have been carried out on the valorisation of these effluents in agriculture as fertilizer. Indeed, Taamallah (2007) demonstrate the richness in inorganic loads such as this high content of non-toxic organic compounds, macro-elements and micro-elements that indicated a significant fertilizing potential of the OMW that could be used advantageously in agronomy.

Indeed, they obtained an excellent fertilizing effect for the cultivation of Maize following a contribution of omww with productions twice superior to those given by the control using quantities of $300 \text{ m}^3/\text{ha/year}$ for 10 years. The same observation was made by Ros de Ursinos and Morisot (1981); Briccoli-Bati and Lombardo (1991).

In France, De Montpezat (1994) showed that a 330 m^3 contribution of OMW per hectare in an olive grove improves the yield of olive trees.

In Tunisia, several attempts to use OMW as fertilizer have been made and directed specifically towards the olive sector (Ben Rouina and Ammar, 1999). Ben Rouina and Taâmallah (2000) affirmed the utility of these effluents as a natural fertilizer for sandy soils.

The same authors and Ben Rouina et al., in (2001) worked on the olive tree and demonstrated that tree fruit shoots are longer and bear more floral clusters and flowers, the tree is increased with the contribution of OMW, in trees receiving doses of 100 m³/ha. However, at higher doses, these effluents caused a slight decrease in production, but, it remained higher than that given by trees with lower doses (50 m³/ha), which was 49.8 kg per tree.

However, some authors note the depressive effects of OMW on the soil following acidification, salinization and nitrogen immobilization (Morisot and Tournier, 1986). Others point to the richness of these effluents in relatively complex organic compounds which are difficult to biodegrade (Catalano et al., 1985), particularly phenolic compounds which have an inhibitory effect on plant growth (Balice et al. 1984; Balice et al. 1988; Cabrera et al. 1996).

Ros de Ursinos et al (1996) have demonstrated that the viscous and greasy character of OMW results in the formation of an oily deposit on the surface of the soil, thus causing it to be impermeabilized in a first stage and subsequently to suffocate.

On the other hand, the intensity of the blackish color of this residue and its cloudy appearance can increase the temperature and negatively influence the microbial activity of the soil (Fedeli and Camurati 1981).

In spite of these results, it can be said that at present, no satisfactory overall solution has been found and the questions concerning the harmful effects of OMW remain until now without clear answers.

The identification of OMW valorization as fertilizer, in agriculture remains a potential initiative with both agronomic and environmental interest.

It is in this perspective that we have proposed to carry out this research work which complements the previous work in order to contribute to solutions allowing to circumvent the possible harmful effects of OMW and to valorize the useful organic compounds for the restoration of degraded lands and improved crop productivity, in particular cereal crops in arid and semi-arid areas.

The potential beneficial effects of land application of OMW need to be clarified under field conditions.

The present study was aimed at evaluating under field experiment, the short and medium term effects of different application rates of olive mill wastewater on several chemical soil properties and the evolution of phenolic compounds. Also, the assessment of the effects on the final yield of Barley crop will be addressed.

Material and Methods

Experimental site and OMW application

Experimental work was carried out in field experiment located at the IRA (Institute of Arid Regions) in southern Tunisia (governorate of Medenine). North latitude: 33° 16′ 21″, East longitude: 10° 19′ 30″. Climatic conditions were typically Mediterranean: with an average annual rainfall of 150 mm occurring mostly in autumn and spring and a mean annual air temperature ranging from 18 to 20 °C.

The soil chosen was isohumic with the following characteristics: sandy texture (90% sand, 3% loam, and 6% clay), pH (7.11), EC (2.32 dS.m⁻¹), % CaCO₃ (5.55) and % OM (0.98).

The fresh OMW was taken from a three-phase continuous extraction factory located in Saadane- southern Tunisia. The physicochemical characteristic of crude OMW are summarized in Table 1 and correspond to the mean values of 3 analyzes.

Characteristics	Data	Table 1
рН	4.80 ± 0.2	Physicochemical
Electrical conductivity (Ds/m)	10.0 ± 0.52	characteristics of olive mill
COD (g/L)	98.0 ± 2.1	waste water used in ferti-
BOD (g/L)	66.0 ± 2.4	irrigation .
Total organic carbon (g/L)	26.0 ± 2.4	(Values after represent ±
Total nitrogen kjeldahl (g/L)	1.60 ± 0.1	standard deviation).
Carbon/Nitrogen	16.25 ± 0.48	
Phenolic compounds (g/L)	8.80 ± 0.3	
Potassium (g/L)	6.10 ± 0.2	
Calcium (g/L)	1.10 ± 0.1	
Phosphorus-Olsen (g/L)	0.35 ± 0.02	
Mangesium (g/L)	0.42 ± 0.01	
Sodium (g/L)	1.57 ± 0.01	
Chlorides (mg/L)	0.65 ± 0.40	

The experimental design consisted of thirty six plots according to a complete randomized block design with three replicates per treatment. Plots with $2 \times 3 \text{ m}^2$

size, were delimited, mechanically ploughed and leveled to subsequently amend them with OMW (olive mill waste water).

Annually, since 2009, OMW spreading application was done in one application on the same plot for three consecutive years : 2009 (First year), 2010 (Second year) and 2011 (Third year). The three contributions were made on the same site and with the same experimental design.

Amendments were applied annually in December (from 2009 to 2012), spreading the waste on the soil surface, followed by arable- level homogenization. After a period of 15 days of rest deemed necessary for drying the soil, the incorporation of OMW was subsequently carried out by manual tillage.

Each year, OMW application was done at rates equivalent to 15, 30, and 45 m³ha⁻¹.

Also T0 plots were not amended and served as control : soil where OMW was never spreading. All the treatments were done in triplicate, distributing the plots alternatively.

The choice of the different doses was adopted following a series of optimizations made following our previous research work, the last one in 2009 in the framework of a master's work, which showed that doses above 50 m³ / ha of OMW negatively affect the behavior of barley (*Hordeum vulgare* L.). T1 = 15 m³/ ha, T2 = 30 m³/ ha and T3 = 45 m³/ ha were used for this purpose.

Four soil subsamples from each plot were taken randomly at 20-cm depth in December from 2009, 1 day after amendment, to 2012 at the end of the experiment (after : six months : After Barley harvest date). Samples were air dried and ground, and the fraction that passed through a 2-mm sieve was used for chemical analyses. A local variety of barley (*Hordeum vulgare* L.) was adopted in irrigated areas.

The main crop management dates over the 3-year period are reported in Table 2. *Hordeum vulgare* L., a very common Barley variety in the experimental area, was used in the 3 experimental years.

Table2. Main Barley crop cycle activities during three.						
	2009/2010	2010/2011	2011/2012			
OMW application	14 December 2009	08 December 2010	28 December 2011			
Sowing	29 December 2009	24 December 2010	12 January 2012			
Harvest	14 April 2010	12 April 2011	07 April 2012			

Parameters Analysis

Chemical Soil Analysis. The following parameters were monitored in the soil layer (0-20 cm), periodically during three years: pH and EC25 (1:2.5 aqueous extracts), TOC (Total Organic C) was determined by Dichromate oxidation, total N by the Kjeldahl method (Kjeldahl, 1883) available P with bicarbonate extraction according to the method of OlsenWatanabe, PC (phenolic compounds) (water soluble and extracted with sodium pyrophosphate 0.4 N/sodium hydroxide 0.1 N, 1:1) quantified by Folin-Ciocalteau method (Box, 1983), N and total K were determined by fire Spectrophotometer and Chlorides ions was estimated using method of titration with Chlorhydric acid.

In order to investigate the influence of waste application on crop yield : number of spikes/ m^2 was estimated. At harvest, grain and straw yield, 1000 grain weight were recorded.

The statistical analysis was performed either for each year or globally for the 3-year period using SAS (System for Windows version 9) and STATIGRAPHICS plus (version 5.1).

Results and discussions

Divergent outcomes were reported in literature of the direct OMW disposal influence on soil properties (Mekki et al., 2006; Chaari et al., 2014), as well its use as a source of OM and nutrients.

Effect of OMW on organic soil composition

The studied soil was initially poor in organic matter (OM). OMW addition increased immediately TOC content for all samples collected.

TOC content ranged from 0.602-1.011 after 1 day from first OMW application to 0.229-1.256 after 1 day from third spreading (Table 3).

These results are in agreement with those of Cabrera et al (1996), which showed that the annual contribution to a sandy soil initially containing 0.45% organic matter of 37 or 61 L/m² of OMW for three years resulting in a respective increase in organic matter of 1.62% and 1.98% respectivly. Abichou (2011) showed that after the first application, the level of soil organic matter initially very low is markedly improved for the different doses of OMW.

This organic carbon increase is proportional to the OMW spread quantities. This is of the greatest importance in semiarid conditions where agricultural soils poor in organic matter are dominant, and are being subjected to intense processes of degradation (Madejón et al., 2003; Sierra, 2007; Taamallah. 2007; López et al., 2008). This increase tends to evolve positively from one year to the next due to the cumulative quantities of OMW spreading during three years of study. Whereas, the total organic carbon content decreased significantly in the amended soils over time, from time 1 to time 2 for all samples. Indeed, for the first year of study, we recorded a highly significant decrease by 27% for T1 (15 m³/ha), 43% for T2 (30 m³/ha) and 55% for T3 (45 m³/ha).

This decrease is respectively by 34%, 45% and 58% for the second year and by 50%, 58%, 73% for the last year. Reduction is correlated with OMW applied dose.

This could be explained by the fact that OMW rich in organic matter have greatly stimulated the soil microbial activity, which has led consequently to a rapid degradation of the organic compounds supplied by these effluents.

At the end of experiment, a relative stability of TOC content of soil has been maintened at an average 0.4% to 0.5% (Table 3). Despite the relatively low nitrogen content of OMW (1.6 g/l), a significant improvement of this element compared to the control was observed for all samples. (Table 3).

Treatment	Voor	Time 1	Time 2	Table 3
	year	: After 1 day	After 6 months	Effect of different

EQA – Environmental quality / Qualité de l'Environnement	/ Qualità ambientale, 27 (2018) 1-17
--	--------------------------------------

Total organic carbon (TOC) Average*						doses of OMW on
T0 (0 m^3 /ha)	1	0.370 ± 0.120	Α	0.176 ± 0.050	А	soil organic
T1 (15 m^3 /ha)	1	0.602 ± 0.050	В	0.328 ± 0.140	В	composition during
T2 $(30 \text{ m}^3 / \text{ha})$	1	0.850 ± 0.590	С	0.413 ± 0.180	С	three successive
T3 (45 m^3 /ha)	1	1.011 ± 0.610	D	$0.458 \pm 0,070$	D	years of studies
$T0 (0 \text{ m}^3/\text{ha})$	2	0.232 ± 0.140	Α	0.148 ± 0.030	А	. · ·
T1 (15 m^3 /ha)	2	0.781 ± 0.330	В	0.437 ± 0.050	В	
T2 (30 m ³ /ha)	2	0.951 ± 0.520	С	0.493 ± 0.060	В	
T3 (45 m^3 /ha)	2	1.103 ± 0.770	D	0.523 ± 0.370	С	_
T0 (0 m^3 /ha)	3	0.229 ± 0.170	Α	0.127 ± 0.050	А	-
T1 (15 m ³ /ha)	3	$.0.802 \pm 0.430$	В	0.400 ± 0.070	В	
T2 (30 m ³ /ha)	3	1.028 ± 0.560	С	0.447 ± 0.190	В	
T3 (45 m ³ /ha)	3	1.256 ± 0.890	D	0.499 ± 0.030	В	
		Total organic N	litro	gen (TON) Averag	ge*	
T0 (0 m ³ /ha)	1	0.012 ± 0.011	А	$0,036 \pm 0,028$	А	
T1 (15 m^3 /ha)	1	$0.019 \pm 0,012$	В	$0,\!047 \pm 0,\!036$	В	
T2 (30 m^3 /ha)	1	$0.026 \pm 0,019$	С	$0,058 \pm 0,052$	С	
T3 (45 m^3 /ha)	1	$0.039 \pm 0,015$	D	$0,067 \pm 0,035$	D	<u>.</u>
$T0 (0 \text{ m}^3/\text{ha})$	2	0.076 ± 0.023	А	0.063 ± 0.026	А	
T1 (15 m^3 /ha)	2	0.109 ± 0.049	В	0.077 ± 0.032	В	
T2 (30 m^3 /ha)	2	0.133 ± 0.054	С	0.105 ± 0.037	С	
T3 (45 m^3 /ha)	2	0.163 ± 0.088	D	0.117 ± 0.062	D	
T0 (0 m ³ /ha)	3	0.078 ± 0.029	А	0.026 ± 0.014	А	
T1 (15 m^3 /ha)	3	0.118 ± 0.056	В	0.048 ± 0.033	В	
T2 (30 m^3 /ha)	3	0.147 ± 0.068	С	0.061 ± 0.043	В	
T3 (45 m^3 /ha)	3	0.188 ± 0.084	D	0.089 ± 0.058	В	
			Avera	age *		
T0 (0 m ³ /ha)	1	$1,237 \pm 0,320$	А	$1,882 \pm 0,820$	А	
T1 (15 m^3 /ha)	1	$2,855 \pm 0,630$	В	$2,729 \pm 0,730$	В	
T2 (30 m_{2}^{3} /ha)	1	$3,696 \pm 0,870$	С	$3,589 \pm 0,590$	С	
T3 (45 m^3 /ha)	1	$6,522 \pm 0,740$	D	$6,862 \pm 0,780$	D	
T0 (0 m ³ /ha)	2	$1,528 \pm 0,230$	А	$1,619 \pm 0,690$	А	
T1 (15 m^3 /ha)	2	$3,557 \pm 0,540$	В	$2,274 \pm 0,830$	В	
T2 (30 m^3 /ha)	2	$4,258 \pm 0,820$	С	$3,460 \pm 0,770$	С	
T3 (45 m^3 /ha)	2	$7,916 \pm 0,960$	D	$6,910 \pm 0,970$	D	
T0 (0 m ³ /ha)	3	$1,547 \pm 0,790$	А	$0,120 \pm 0,610$	А	
T1 (15 m_{3}^{3} /ha)	3	$3,601 \pm 0,650$	В	$1,240 \pm 0,640$	В	
T2 (30 m_3^3 /ha)	3	$7,876 \pm 0,660$	С	$2,410 \pm 0,830$	С	
T3 (45 m^3 /ha)	3	$9,980 \pm 0,790$	D	$4,740 \pm 0,870$	D	

At the end of the third year, we were able to deduce that each new contribution of OMW was followed by a restoration of the stock of soil total organic carbon of the previous year.

At the first sampling time, a slightly total organic nitrogen (TON) increase was observed in the amended plots compared with the control, whereas six months after the OMW treatment the amended soils showed higher values of TON than the untreated ones. The t-test pointed out a significantly increase of TON in the OMW treated soil from (time 1) to (time 2).

After the second and third application, OMW resulted in a marked increase in TON content. Indeed, just after OMW disposal, values were found to be 10-fold higher than the observed ones after the first spreading operation.

This considerable increase can be attributed to the effect of cumulative quantities of OMW after two successive years and especially to microorganisms activity.

However, we have seen a marked decrease in TON at the end of the crop cycle compared to that at the beginning of the experiment. This diminution could be due to the element uptake by microorganisms and plants' active metabolism.

OMW incorporation contribute to establish a balance between the input and output of organic and mineral nitrogen pools with conservation of a relatively acceptable rate of this element.

As regard phenolic compounds, the OMW treatment determined statistically significant increases compared to the controls for all samples. This increase was amplified from one year to the next following the cumulative effect of three successive OMW applications (Table 3). Polyphenols are difficult to decompose (Obied et al., 2005) and present phytotoxicity (Aggelis et al., 2003) or suppression of soil microorganisms (Kotsou et al., 2004). Moreover, the high level of polyphenols in OMW can also pollute surface and groundwater resources (Aggelis et al., 2003).

Moreover, over the time, a statistically significant decreases were consistently detected in the OMW treated soils 6 months from OMW application. Such decrease could be explained by the fact that phenols are broken down by specific bacteria, yeasts (Di Serio et al., 2008), and fungi. These microorganisms greatly degrade (Kissi et al., 2001; Kachouri et al., 2005; Ergül et al., 2009) OMW and follow first-order kinetics, which depend on environmental conditions such as soil moisture and temperature.

Polyphenols are the main limiting factor for spreading OMW because of their phytotoxic and antibacterial actions. Thus, polyphenol degradation and incorporation into the soil humic fraction depend on environmental conditions (Sierra et al 2007).

Effect of OMW on soil mineral composition

Excepting some non significant fluctuations (increase, decrease) recorded after the first and second OMW spreading, the pH remains predominantly neutral.

As a result, the buffering capacity of the soil counterbalances the acidity of OMW, mainly due to the presence of organic acids. Similar results were obtained by Chaari et al. (2015) after a regular application of higher doses in a 9 years' study.

These results are also consistent with those of Levi-Minzi et al. (1992), which demonstrated that, despite their high acidity, OMW do not modify the pH due to the buffer capacity of the soil which is relatively rich in limestone (general case of southern Tunisia soils).

In this case, this buffering capacity can promote the activity of soil microflora by reducing the antimicrobial power of OMW phenolic compounds (Dakhli, 2015).

OMW resulted in a significant increase in soil salinity compared to the control.

Thus, EC of the soil, its sodium and chloride content were significantly affected by the application of OMW, showing increases one day after spreading and decreases six months later in the treated soils compared with the controls. Our findings were consistent with previous works, reporting EC increases during the irrigation times and decreases in between them (Cabrera et al., 1996; Mekki et al., 2007; Sierra et al., 2007; Chartzoulakis et al., 2010; Kavvadias et al., 2014; Moraetis et al., 2011).

Soil salinity increase affects the soil structure and porosity, moisture holding capacity of soils and the water infiltration, the biological activity and diversity of soil organisms, and plant nutrient availability.

Kexch and Total K were significantly affected by the OMW treatment and just after third spreading disposal, values were found to be 8- to 10-fold higher than the observed ones after first and second OMW supply (Tables 3). By contrast, at second sampling, low decreases were recorded (a mean of 31% and 34% respectively for Total K and Exch K). In addition, six months after spreading soil both K forms values were two-fold lower than the values observed at time 1.

Both Kexch and total K were higher in the treated soils in comparison with the controls irrespective of time after disposal and spreading period. These increases are directly caused by the high content of both K forms in the OMW and are in agreement with what has been previously observed by several authors (Mechri et al., 2008; Di Serio et al., 2008; Chartzoulakis et al., 2014; Kavvadias et al., 2010). These K increases are beneficial for crop productivity and health according to the different plant requests and uptake efficiency (Arienzo et al., 2009) and can have ecological and economical advantages avoiding or reducing the use of K fertilisers (Di Serio et al., 2008). Furthemore, Rusan et al (2016) demonstarded that K drastically increased with all OMW treatments compared with the control, where the highest value for soil K was 2926 mg kg-1 for the UOMW. The increase in soil K contents with OMW application can be attributed to their high content in the OMW used in the irrigation (2441.8 mg L^{-1}). The contribution of OMW to soil total potassium content is certainly positive. However, the accumulation of excessive amounts recorded represents a potential source of toxicity that can threaten the activity of soil microorganisms and the mineral nutrition of barley. From the results obtained, we can conclude that OMW constitute a potential source of potassium showing a high fertilizing potential that can be valued for the rehabilitation of poor soil in this element, taking into consideration that higher concentrations of this element could cause an imbalance of cation exchange complex and consequently a plant nutritional disturbance.

Effect of different doses of OMW on barley yield components

OMW impact on Barley yield components during 3 years' period was illustrated in Table 4.

Treatment	year	Number of spikes/m ²	Yield straw (T/ha)	Grain yield (T/ha)	1000 Grains Weight (g)	
T0 (0 m^3 /ha)	1	$265.7 \pm 64.7 \text{ A}$	2.230 ±0.79 A	1.26 ±0.48 A	59.58 ±2.77 A	
T1 (15 m ³ /ha)	1	243.3 ±49.3 A	$2.058\pm0.52\;A$	1.044±0.27 B	5896±1.70 A	
T2 (30 m ³ /ha)	1	175.5 ±42.2 A	$1.653 \hspace{0.2cm} \pm \hspace{0.2cm} 0.84 \hspace{0.2cm} B$	0.766±0.24 C	59.97 ±5.95 A	
T3 (45 m ³ /ha)	1	151.0 ± 40.8 B	$1.545 \ \pm 0.47 \ B$	0.675 ±0.47 C	59.84 ±5.14 A	
T0 (0 m^3 /ha)	2	187.2 ±35.5 A	1.233 ±0.22 A	0.412 ±0.21 A	58.52 ±1.75 A	
T1 (15 m ³ /ha)	2	100.7 ±34.5 B	0.966 ±0.17 B	$0.171 \pm 0.08 \text{ B}$	56.19 ±2.34 A	
T2 (30 m ³ /ha)	2	85.2 ±18.4 C	0.925 ± 0.14 B	$0.150 \pm 0.04 \text{ B}$	55.37 ±2.93 A	
T3 (45 m ³ /ha)	2	82.5 ±30.5 C	0.915 ±0.21 B	0.127 ±0.09 C	54.39 ±1.77 A	
T0 (0 m^3 /ha)	3	127.0 ±27.3 A	0.580 ±0.18 A	0.159 ±0.03 A	57.37 ±1.18 A	
T1 (15 m ³ /ha)	3	51.2 ±22.0 B	0.481 ±0.09 A	0.100 ± 0.02 B	54.51 ± 1.80 A	
T2 (30 m ³ /ha)	3	50.5 ± 12.6 B	0.360 ±0.17 B	0.061 ±0.01 C	54.44 ±3.62 A	
T3 (45 m ³ /ha)	3	18.83 ±8.79 C	$0.342\pm0.08~B$	$0.031\pm0.01~D$	53.96 ±5.11 A	
For each year different letters indicate a significant difference among the means at LSD test (P $>$ 0:001).						

Table 4. Effect of different doses of OMW on Barley yield components during threesuccessive years of studies.

*Number of spikes/m*². The number of spikes/m² over the 3-year period decreased from the first to the last years compared to the controls, in particular for 30 m³/ha and 45 m³/ha. This amplification of damage can be attributed to the cumulative effect of successive OMW applications. At the third year of experiment, this reduction was by 60% for T1 (15 m³/ha) and T2 (30 m³/ha) and by 85% for T3 (45 m³/ha).

1000 Grains Weight. for the first year, this parameter was not affected by OMW application compared to the control for all samples. Indeed, no statistically significant difference was observed. However, for the next two years, this parameter showed a slight reduction compared to the control without statistically significant difference.

Yield straw Barley straw yield over the 3-years period decreased from the first to the last years in request to the control whatever the dose applied. This reduction is correlated with the assigned dose. Indeed, the reduction induced by the T1 dose (15 m^3 /ha) is significant. However, it's highly significant for the higher doses.

At the end of the third year, this reduction is by 62% and 58% respectively for the T2 ($30 \text{ m}^3/\text{ha}$) and T3 ($45 \text{ m}^3/\text{ha}$).

Grain yield For the first year, a reduction in grain yield by 18% compared to the control was recorded for the T1 dose (15 m³/ha). However, this decline is highly significant for T2 (30 m³/ha) and T3 (45 m³/ha) and is 61% and 54% respectively.

For the second year of trial, this reduction is much more important. Indeed, it is by 59% relative to the control respectively for the doses T1 and T2 and by 69% for the higher dose T3. Concerning the third year of experiment, T1 (15 m^3 /ha) induced a significant reduction in Barley grains yield by 37% in respect to the control. However, T2 (30 m^3 /ha) and T3 (45 m^3 /ha) were distinguished by catastrophic damage with a yield reduction by 62% and 81%, respectively.



Figure 1a. Effect of different doses of OMW on soil pH, electrical conductivity and Sodium during three successive years of studies.

DOI: 10.6092/issn.2281-4485/7295



Figure 1b. Effect of different doses of OMW on soil chlorides, total and exchangeable potassium during three successive years of studies.

Barley yield components, over the 3-year period decreased from the first to the last years, in particular the plots receiving 30 m³/ha and 45 m³/ha. This could be explained by a cumulative effect of chemical substances in the soil after 3 years of application of OMW. These results confirmed those obtained by Bonari and Ceccarini (1993) for wheat and barley. Also, Moreno et al. (1987) and (Mekki et al. 2006; Rusan et al., 2015; Dakhli et al., 2009, 2013a, 2013b) reported that application of OMW causes impose phytotoxic effects on Barley crop.

The use of OMW as a fertilizer has therefore not encouraging results with respect to straw and grain yields of barley. On the contrary, it has been a factor of stress and metabolic disturbance.

Conclusion

The results of the three-years of study led to the conclusion that the application of OMW as a fertilizer to a barley crop induced a highly significant decrease in all yield components inspit of the improvement of some soil chimical properties such as carbon, nitrogen and potassium.

The accumulation of phenolic substances and the excessive increase in sodium and chloride soil levels caused by the incorporation of OMW are the cause of an hydromineral imbalance, a disturbance of the mineralization process and an exogenous nutritional stress affecting the physiological behavior of barley and its productivity.

The results of this study seem to confirm that the impact of the OMW residues on soil properties is the result of opposite effects, depending on the relative amounts of beneficial and toxic organic and inorganic compounds present.

In conclusion, the contribution of OMW as an organic amendment have no agronomical interest.

On the contrary, it can be a source of toxicity especially for annual crops with a relatively short root system such as Barley (*Hordeum vulgare* L.).

OMW in sandy soils in arid conditions of southern Tunisia can be considered as an alternative to implement with caution for the rehabilitation of poor organic matter soils. However, to improve the use of these effluents in agriculture, further studies are needed to reach an affirmative answer; sustainable management of OMW irrigation and periodic monitoring of soil fertility and crop quality parameters are required to ensure safe and long-term reuse of OMW in agriculture.

Acknowledgements

We are thankful to the Institute of Arid Areas Medenine-Tunisia for all necessary assistance to carry out this research.

Competing interests The author declares that they have no competing interests.

Authors' contributions This work built on previous studies in the same Laboratory. Houcine Khatteli and Elhem mallak maalej contribute by discussion results. Raja Dakhli as Corresponding author contributes by the most part of this

work as the studies of effects of OMW on soil chemical properties and on crops growth. All authors read and approved the final manuscript.

References

ABICHOU M. (2011). L'épandage des Margines: une alternative pour améliorer la structure superficielle du sol, son activité biologique et pour lutter contre l'érosion éolienne dans le sud Tunisien. Thèse de doctorat. Arido-culture et lutte conte la désertification. INAT Tunis.

AGGELIS, G., ICONOMOU, D., CHRISTOU, M., BOKAS, D., KOTZAILIAS, S., CHRISTOU, G., TSAGOU, V., & PAPANIKOLAOU, S. (2003). Phenolic removal in a model olive oil mill wastewater using pleurotus ostreatus in bioreactor cultures and biological evaluation of the process. Water Research, 37(16):3897–3904.

ALBI ROMEROS M. A., ROS DE URSINOS J. A. (1960). Estudio del alpechin para su aprovechamiento industrial. Ensayos efectuados par su posible utilization come fertilisante. Grasa y Aceites, 11(3):123-124.

ARIENZO M, CHRISTEN EW, QUAYLE W, KUMAR A (2009) A review of the fate of potassium in the soil-plant system after land application of wastewaters. J Hazard Mater., 164:415–422.

AYOUB S, AL-ABSI K, AL-SHDIEFAT S, AL-MAJALI D, HIJAZEAN D (2014) Effect of olive mill wastewater land-spreading on soil properties, olive tree performance and oil quality. Sci Hortic 175:160–166.

BEN ROUINA B., AMMAR E. (1999). L'utilisation des margines comme fertilisant biologique pour les cultures des plantes maraichères. Séminaire national sur l'huile d'olive et ses dérivés. Société chimique de Tunisie. Hammamet 26-28 Nov. 1999.

BEN ROUINA B., TAÂMALLAH H. (2000). L'utilisation des margines comme fertilisant en agriculture. Collaboration Institut des Régions Arides de Medenine – Institut de l'Olivier de Sfax. Rapport technique.

BALICE V., CERA O. (1984) Acidic phenolic of the oil vegetation water determined by a gas chromatographic method. Grasas y Aceites. 178 p.

BALICE V., BOARI G., CERA O., RINDONE B. (1988) The fate of tanninlikecompounds from olive mill effluent in biological treatment. Proceeding of the fifth international symposium on anaerobic digestion. Bologna, Italy. Eds Hall Erand Hobson NP. pp 279- 280.

BONARI, E., CECCARINI, L. (1993) Sugli effetti dello spargimento delle acque di vegetazione sul terreno agrario: risultati di una ricerca sperimentale. Genio Rurale, 5:60–67.

BOX J.D. (1983) Investigation of the Folin-Ciocalteau phenol reagent for the determination of polyphenolic substances in natural waters, Water Research, 17:511-522.

BRICCOLI-BATI C., GRANATA R., LOMBARDO N. (1991) Valutazione del pericolo di inquinamento delle falde acquifere in seguito allo spandimento di acque di vegetazione su terreno agrario. In: Senesi, N., Miano, T.M. (Eds.), Riciclo di biomasse di rifiuto e di scarto e fertilizzazione organica del suolo. Realtà e prospettive, vantaggi e rischi per l'agricoltura e l'ambiente, ed. Patron, pp. 303–308.

CABRERA F., LOPEZ R., MARTINEZ-BOURDIU E., DUPUY DE LOME E., MURILLO J.M. (1996) Land treatment of olive oil mill wastewater. International Biodegradation and Biodeterioration, 38,:215-225.

CATALANO M., GOMEZ T., DE FELICE M., DE LEONARDIS T. (1985) Disposal of wastewaters from oil mills. What alternatives to treatment? Inquinamento, 27(2):87-90.

DAKHLI.R. (2009). Valorisation des margines en agriculture: effet sur les proprietés chimiques du sol et sur le rendement d'une culture d'Orge. DEA. IRA-INAT.

CASA R, D'ANNIBALE A, PIERUCCETTI F (2003) Reduction of the phenolic components in olive-mill wastewater by enzymatic treatment and its impact on durum wheat (Triticum durum Desf.) germinability. Chemosphere, 50:959–966.

CERETI C.F., ROSSINI F., FEDERICI F., QUARATINO D., VASSILEV N., FENICE M. (2004) Reuse of microbially treated olive mill wastewater as fertilizer for wheat (Triticum durum Desf). Biores Technol., 91:135–140.

CHAARI L, ELLOUMI N, MSEDDI S, GARGOURI K, BOUROUINA B, MECHICHI T, KALLEL M (2014) Effects of olive mill wastewater on soil nutrients availability. Int J Interdiscip Multidiscip Stud., 2:175–183.

CHAARI L., ELLOUMI N., MSEDDI S., GARGOURI K., ROUINA B.B., MECHICHI T., KALLEL M. (2015) Changes in soil macronutrients after a long-term application of olive mill wastewater. J Agric Chem Environ, 4:1–13

CHARTZOULAKIS K., PSARRAS G., MOUTSOPOULOU M., STEFANOUDAKI E. (2010). Application of olive mill wastewater to a Cretan olive orchard: effects on soil properties, plant performance and the environment. Agriculture, Ecosystems and Environment, 138:293:298.

DAKHLI R, LAMOURI R. (2013a) Effet de l'epandage des margines sur les proprietes chimiques du sol et sur le comportement phenologique et le rendement d'une culture d'orge. European Journal of Scientific Research, 112(1):94-109.

DAKHLI R, LAMOURI R, TAAMALLAH H, OUESSAR M. (2013b) Short term effects of olive mill waste water on soil chemical properties under semi arid mediterranean conditions. Journal of Life Sciences, 7(11):1209-1218.

DAKHLI R.(2015) .Effet des margines sur les proprieté chimiques et sur le rendement d'une culture d'orge. IRA/INAT.pp.39-112.

DE MONTPEZAT G. L. (1994). Présentation et discussion des méthodes d'épuration et de transformation des margines en vue de leur utilisation agricole. Etude d'épuration des eaux résiduaires des huileries d'olives (Géoépuration et épandage agricole des margines avec et sans transformation). Atelier d'écotechnologie organisé par le ministère de l'environnement et l'aménagement du territoire de Tunisie avec la collaboration de la GTZ. Tunis 13 décembre 1994. Eds. Deutshe Gesellschaft für Tesnishe Zusammenaarbit (GTZ) Gmbh Imprimerie Réunie de Tunisie. 380 p.

DI SERIO M.G., LANZA B., MUCCIARELLA M.R., RUSSI F., IANNUCCI E., MARFISI P., MADEO A. (2008) Effects of olive mill wastewater spreading on the physico-chemical and microbiological character.

DOULA M.K., TINIVELLA F., MORENO J.L., KAVVADIAS V., SARRIS A., THEO-CHAROPOULOS S., SANCHEZ-MONEDERO M., ELAIOPOULOS K. (2012) Good practices for the agronomic use of olive mill wastes-Application guide. (Edited by M.K. Doula), LIFE07 ENV/GR/280 PROSODOL, 2012, pp. 1-61..

ERGÜL F.E., SARGIN S., ÖNGEN G., SUKAN F.V. (2009) Dephenolisation of olive mill wastewater using adapted Trametes versicolor. International, Biodeteroration and Biodegradation, 63:1–6.

FEDELI E., CAMURATI F. (1981) Valorisation des margines et des grignons épuises par récupération de quelques composants. Séminaire International sur la valorisation des sousproduits de l'olivier PNUD/FAO/COI. Monastir, Tunisia, December 1981.

FIESTAS ROS DE URSINOS J.A. (1981). Différentes utilisations des margines. Actes du Séminaire Internatinal sur la valorisation des sous produits de l'olivier. Organisation des Nations Unies pour l'Alimentation et l'Agriculture (FAO), 93-110. Tunisie.

DOI: 10.6092/issn.2281-4485/7295

KJELDAHL J. (1883) A new method for the determination of nitrogen in organic matter, Z. Analytical Chemistry, 22:366.

KACHOURI S., HALAOULI S., LOMASCOLO A., ASTHER M., HAMDI M. (2005) Decolourization of black oxidized olive mill wastewater by a new tannase-producing Aspergillus flavus strain isolated in soil. World Journal of Microbiology and Biotechnology, 21:1465–1470.

KAVVADIAS V., DOULA M, THEOCHAROPOULOS S (2014) Long-term effects on soil of the disposal of olive mill waste waters (OMW). Environ Forensics, 15:37–51.

KISSI, M., MOUNTADAR, M., ASSOBHEI, O., GARGIULO, E., PALMIERI, G., GIARDINA, P., SANNIA, G., 2001. Roles of two white-rot basidiomycete fungi in decolourization and detoxification of olive mill waste water. Applied Microbiology and Biotechnology, 57:221–226.

KOMILIS DP, KARATZAS E, HALVADAKIS CP (2005) The effect of olive mill wastewater on seed germination after various pretreatment techniques. J Environ Manage, 74:339–348.

KOTSOU, M., MARI, I., LASARIDI, K., CHATZIPAVLIDIS, I., BALIS, C., KYRIACOU, A., 2004. The effect of olive oil mill wastewater (OMW) on soil microbial communities and suppressiveness against Rhizoctonia solani. Applied Soil Ecology, 26: 113–121.

LEVI-MENZI R, SAVIOZZI A, RIFFALDI R, FALZO L (1992) L'épandage aux champs des eaux de vegetation: effet sur les propriétés des sols. Olivae, 40:20–25.

LESAGE-MESSEN L., NAVARRO D., MAUNIER S., SIGOILLOT J.C., LORQUIN J., DELATTRE M., SIMON J.L., ASTHER M., LABAT M. (2001). Simple phenolic content in oil residues as a function of extraction systems. Food Chemistry, 75:501–507.

LOPEZ-PIÑEIRO A., SILVA C., NUNES J.M., ROZAS M.A., GARCIA A. (2002) Effects of solid olive mill waste application on soil properties of irrigated olive grove, in : J.L. Rubio, R.P.C. Morgan, S. Asins, V. Andreu (Eds.), Man and Soil at the Third Millennium - Geoforma, 2:1117-1123.

LÓPEZ-PIÑEIRO A., FERNÁNDEZ J., ALBARRÁN A., RATO NUNES J.M., BARRETO C. (2008) Effects of de-oiled two-phase olive mill waste on Mediterranean agricultural soils and the wheat crop. Soil Sci Soc Am J., 72:424–430.

MADEJON E., BURGOS P., LOPEZ R., CABRERA F. (2003) Agricultural use of three organic residues: effect on orange production on properties of a soil of the Comarca Costa de Huelva (SW Spain), Nutr. Cycl. Agroecosys., 65:281-288.

MECHRI B., BEN MARIEM F., BAHAM M., BEN ELHADJ S., HAMMAMI M. (2008) Change in soil properties and the soil microbiological community following land spreading of olive mill wastewater affects olive trees key physiological parameters and the abundance of arbuscular mycorrhizal fungi. Soil Biology and Biochemistry, 40:152–161.

MEKKI A., DHOUIB A., SAYADI S. (2006) Changes in microbial and soil properties following amendment with treated and untreated olive mill wastewater. Microbiological Research 161, 93–101.

MEKKI A, DHOUIB A, SAYADI S (2007) Polyphenols dynamics and phytotoxicity in a soil amended by olive mill wastewaters. J Environ Manage 84:134–140.

MORAETIS D., STAMATI F.E., NIKOLAIDIS N.P., KALOGERAKIS N. (2011). Olive mill wastewater irrigation of maize: Impacts on soil and groundwater. Agric. Water Manage. 98 (7):1125-1132.

MORENO E., PEREZ J., RAMOS-CORMENZANA A., MARTINEZ J. (1987) Antimicrobial effect of waste water from olive oil extraction plants selecting soil bacteria after incubation with diluted waste. Microbios, 51:169–174. OBIED H.K., ALLEN M.S., BEDGOOD D.R., PRENZLER P.D., ROBARDS K., STOCKMANN R. (2005) Bioactivity and analysis of biophenols recovered from olive mill waste. Journal of Agricultural and Food Chemistry, 53(4):823–837.

PAGLIAI M., PELLEGRINI S., VIGNOZZI N., PAPINI R., MIRABELLA A., PIOVANELLI C., GAMBA C., MICLAUS N., CASTALDINI M., DE SIMONE C., PINI R., PEZZAROSSA B., SPARVOLI E. (2001) Influenza dei reflui oleari sulla qualità del suolo. L'informatore Agrario, 50(1):13–18.

PAREDES C., CEGARRA J., BERNAL M.P., ROIG A. (2005) Influence of olive mill wastewater in composting and impact of the compost on a Swiss chard crop and soil properties. Environ Int., 31:305–312.

RINALDI, M., RANA, G., & INTRONA, M. (2003) Olive-mill wastewater spreading in southern Italy: effects on a durum wheat crop. Field Crops Research, 84(3):319–326.

ROS DE URSINOS J. A. (1981). Différentes utilisations des margines. Obtention de protéines unicellulaires. FAO. Séminaire international sur la valorisation des sous-produits de l'olivier. pp 101 -103.

ROS DE URSINOS F., MORISOT M. (1983). Différentes utilisations des margines: Recherches en cours, résultats obtenus et applications. Séminaire international sur la valorisation des sous produits de l'olivier. Monastir – Tunisie. 15 – 17 décembre 1981. Eds PNUD/FAO, Madrid Espagne. 1983. pp 93 -110.

ROS DE URSINOS F., BERNDT L., GEISSEN K., KACHOURI M., KLIMM E. (1996) Les expériences méditerranéennes dans le traitement et l'élimination des eaux résiduaires des huileries d'olives. Coopération Tunisie-Allemagne. 380 p.

RUSAN M.J.M., ALBALASMEH A.A., ZURAIQI S., BASHABSHEH, M. (2015) Evaluation of phytotoxicity effect of olive mill wastewater treated by different technologies on seed germination of barley (hordeum vulgare l.) Environmental Science and Pollution Research, 22(12):9127–9135.

RUSAN M.J.M., ALBALASMEH A.A., MALKAWI H.I. (2016) Treated Olive Mill Wastewater Effects on Soil Properties and Plant Growth. Water Air & Soil Pollut., 227:135. SAADI, I., LAOR, Y., RAVIV, M., MEDINA, S., 2007. Land spreading of olive mill wastewater: effects on soil microbal activity and potential phytotoxicity. Chemosphere 66, 75–83.

SIERRA J., MARTI E., GARAU M., GRUANAS R. (2007) Effects of the agronomic use of olive mill wastewater: field experiment, Sci. Total Environ., 378:90-94.

TAAMALLAH. H (2007) L'épandage des margines en vergers d'oliviers une alternative pour la valorisation de cet effluent. Thèse de Doctorat.pp.107

ZENJARI A., NEJMEDDINE A. (2001) Impact of spreading olive mill wastewater on soil characteristics: laboratory experiments. Agronomie, 21:749–755.