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Assessment of olive mill wastewaters bioconversion potential into biotechnological and health interest microbial biomass

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ABSTRACT/RESUME

Abstract: *Olive mill by-products which are characterized by powerful antioxidant and recalcitrant compounds to biodegradation represent an important environmental problem. They are produced in huge quantities in short periods of time. Therefore, finding a suitable biovalorization strategy to exploit these by-products is a great interest. In this context, the current study aimed to olive mill wastewaters (OMWW) bioconversion into valuable microbial biomass, oleaginous yeasts, as well as healthy probiotic bacteria as enrichment substrat. This study also describes and discusses the antimicrobial potential of OMWW. Yeasts and bacterial growth was monitored by plating on malt extract agar (MEA) and Man Rogosa and Sharpe Agar (MRSA) agar respectively. The antimicrobial properties of ethyl acetate polyphenolic extracts from fermented and unfermented OMWW were assessed using the disc diffusion method. The experiments trials reveals that the raw OMWW exert a strong inhibition against oleaginous yeasts and probiotic bacteria while substrate dilution or enrichment led to cell growth promoting and polyphenol removal. Moreover, polyphenolic extracts provide a wide antimicrobial spectrum potential against pathogenic and food spoiling bacteria. Thus, OMWW could be considered as a beneficial and valuable nutritional source for microbial biomass production and mainly those are of biotechnological and health interest.*

I. Introduction

Because of the growing interest and demand in high dietetic and healthy nutritional value compounds, olive oil production is an expanding industrial sector worldwide which yields reached a value close to 2.89 million tons in 2017 [1]. Olive oil extraction process leads to two residues: a solid fraction, olive mill solid residu (OMSR), resulting from the pressed olive oil vegetation (olive cake or Amegruche in Kabylia region, Algeria) and a liquid fraction, commonly known as OMWW as well as Alpechin, Amurca [2] and Amuredj in Kabylia region. In pressure (traditional method) or three-phase centrifugation process effluents are olive pomace and OMWW, whereas in a two-phase centrifugation oil extraction system, the effluent is composed of wet olive pomace. The structural characterization of these two natural by-products showed that they are mainly composed of simple and complexes polyphenolic compounds, pectines and prebiotic polysaccharides, organic acids and mineral nutrients variably distributed depending on the employed olive oil extraction process and the agronomic practices [3, 4, 5]. Beside, OMWW are associated to a huge pollution issues due to their color darkness and high content of phenolic compounds. Unfortunately, OMWW are often disposed in evaporation ponds or various environmental receptors leading to strong odor nuisance, soil contamination, plants growth or germination inhibition [3, 5].

OMWW can be considred as serious threat to the aquatic flora and fauna, thus severe effects on

ecological status when directly discharged in stream water and rivers [6]. The darkly high concentrated polyphenols can color aquatic area and the high concentration of reduced sugars can lead to the lowering in the dissolved oxygen concentrations by stimulating the microbial respiration [7]. Due to their high polluting impact, an economical and ecological-friendly sustainable management method must be developed for OMWW. In this context, in recent decade the main challeng for many researchers focused toward the finding of protocols and biotechnological innovations strategies aiming to agro-industrial by-products bioconversion into high added-value products. Chemical composition of OMWW depends on many factors, such as the employed system for oil extraction, the degree of maturity, the time of olive storage and the olive trees varieties [3]. The high values of chemical oxygen demand (COD) and polyphenols concenrtations (e.g., phenolic acids, hydroxytyrosol, oleuropein, tannins, flavonols, anthocyanins, etc.), up to 12 g/L [8], makes the OMWW difficult to their managment. Some of these polyphenols were reported being responsible for several biological activities, including antibiosis against *Bacillus megaterium* [9] and phytotoxicity [10]. They also appear to be involved in the plants defense against invading pathogens, including bacteria, fungi and viruses [11].

OMWW could be exploited either as a nutritional source for microorganisms growth, bioproducts production and animal feed [3, 5], or as a natural source for polyphenols recovery as well as dietary fiber or prebiotic [4, 12, 13, 14, 15, 16]. Due to their several health benefits and richness in organic compounds, the worldwide researches on OMWW bioconversion and their own bioactive compounds is increasing rapidly. Oleaginous yeasts and probiotic bacteria are considred as intereting and eco-freindly microorganismes for agro-indutrial by-products biotechnological exploitation. It should be emphazised that the inhibitory effects of the by-products mainly OMWW must be taken into acount in order to improve the bioprocess or the food application if used as supplement. Thus, despite the OMWW beneficial effects, if subjucted to use as a food matrix for probiotics food products their effects on these bacteria should be well undrestanded. Probiotic bacteria are well known to provide a wide variety of benificial health effects, contribute to intestinal balance, play a role in maintaining health and can even improve food quality if incorporated with them [17].

Oleaginous yeasts mainly *Yarrowia lipolytica* and *Cryptococcus curvatus* were reported to have several application in biotechnological industry [18, 19, 20, 21, 22]. Among oleaginous yeasts [23], the most reported productive ones are *Lipomyces starkeyi*, *Rhodosporidium toruloides*, *Cryptococcus curvatus* and *Yarrowia lipolytica* [24, 25, 6, 21, 22].

Biodiesel production using microbial lipids is considred as an alternative to oils production for biodiesel generation. In this later field, the progress of research in lipids biosynthesis using fungi and bacteria is now known and controlled [21, 22]. Application of agro-industrial wastes and co-products in bioprocess provides an alternative way to replace the refined and costly raw materials. In addition, the bulk use of agro-industrial wastes residues will help to solve environmental issues. OMWW bioconversion as well as the antimicrobial and antioxidant potentials of phenolic compounds recovered from olive mill byproducts were reported in scientific literature. However, fewer data are available on OMWW bioconversion in Algeria. In the frame of the present work, the suitability of OMWW as nutiritional or enrichment substrates for oleaginous yeast production as well as probiotic biomass was assessed and in our knowlidge, no study have been focuced on this later field in Algeria.

II. Materials and methods

II.1. Substrate collection

OMWW samples, OMWW1 and OMWW2, were collected respectively, from olive oil extraction press traditional and three phase units located in Tizi-Ouzou, Algeria. The fresh samples were taken directly after oil extraction, from olive *Chamlal* variety, distributed in 500 ml bottles and immediately frozen at -20° C until further uses.

II.2. Microorganisms and cultivation media

All strains used in the current study are from ATCC culture collections (American Type Culture Collection, Rockville) and DSMZ (Deutsche Sammlungvon Mikroorganismen Zellkulturen, Braunschweig, Allemagne). Yeast and probiotic strains were taken from Pascal institut, Clermont Ferrand (France). OMWW physicochemical characteristics as well as the used yeast and bacterial strains are displayed in tables 1 and 2 respectively. Before each experimental trial, OMWW were defrozen.

The medium of OMWW was sterilized in an autoclave at 121°C for 20 minutes. The serial experiments units for yeasts were performed in 250 ml Erlenmeyer flasks containing 100 ml of the medium and plugged with cotton. Experiments trials were performed on raw OMWW (M), diluted OMWW (DM) and / or enriched with glucose (M-Glu, DM-Glu) or glycerol (M-Gly, DM-Gly) at 3% in both cases [27].

Regarding to probiotic bacteria, fermentations were conducted in MRS broth (MRSB) enriched with raw OMWW. It should be empphazised that in this section of experiments using probiotic bacteria, two different OMWW samples, OMWW1 and OMWW2 were used. Cell growth, for the six

employed probiotic bacteria, was monitored on OMWW2 over 72 h incubation time.

For inoculum production, the yeast and bacterial strains were subcultured respectively on MEA and MRSA media. After 24 h cultivation time, a loop of strain is transferred in their respective broth, MEB and MRSB and incubated for 24 h. The resulting culture broths were subjucted to centrifugation for cell harvesting and washing three times. The obtained cells suspensions were used for media inoculation.

II.3. Microbial growth assessment on olive mill wastewaters

Because of the darkness of the OMWW, microorganisms optical densities (OD) measurement was not possible. Consequently, the total colony forming units (CFU) counting of microorganisms was carried out by serial dilution spreading in plate count agar medium for the different inoculated culture media (initial and final count). The microorganisms cell count was carried out following a series of dilution (10^{-1} to 10^{-10}) in sterile physiological water for the different inoculated culture media.Then, 100 µl were taken from each dilution and spreaded on MEA or MRSA.

II.4. Substrates physico-chemical characteristics

…Substrates characterization (table1) was performed by measuring pH, moisture (MS), total solids (TS), volatil organic matter (VS). The moisture content was assessed by drying at 105°C for 24 h and organic matter (VS) by determining the loss-on ignition at 550°C for 4 h. Total phenolic compounds content of OMWW extracts were determined using Folin–Ciocalteau reagent [28]. The absorption at a wavelength of 750 nm was determined with a spectrophotometer. Data are expressed as gallic acid equivalents (mg gallic acid/ml OMWW).

Table 1. *Physico-chemical characteristics of the used OMWW samples (mean values ± SD).*

Parameters	**OMWW1 Press system**	**OMWW2 Three phase system**
TS (g/l)	88.47	95
MS (%)	92.24	89.32
VS (g/l)	83.35	84.9
Polyphenol (g/L)	3.8	4.2
pH	4.89	6.2

II.5. Polyphenol extraction

Polyphenol recovery was performed according to ethyl acetate solvent extraction. The extraction operation was repeated three times for a maximum phenolic compounds recovery. The organic phase rich in phenolic compounds was subjucted to evaporation under vacum in a rotary evaporator at 40°C. The dry extracts were then recovered in a minimum volume of Dimethyl sulfoxide (DMSO) (10%) for antimicrobial activity. As well known, ethyl acetate is more effective than other extraction solvents with a high extraction effiency. The polyphenol of each sample are extracted before and after fermentation by each strain.

Table 2. *Microbial strains used in this study.*
LB: Lactobacillus, CR: Cryptococcus curvatus, Y: Yarrowia lipolytica.

Probiotic Strains	**T°**	**Yeast strains**	**T°**	**Food born and pathogenic strains**	**T°**
Lactobacillus gasseri DSM 20243 (LB_2)	37	*Yarrowia lipolytica* MUCL 28849 (Y)	28	*Staphylococcus aureus* ATCC 25923	37
Lactobacillus paracasei subsp paracasei DSM 20312 (LB_3)	30			*Bacillus cereus* ATCC 14579	37
Lactobacillus rhamnosus GG ATCC 53103 (LB_4)	37			*Enterococcus faecalis* ATCC 49452	37
				Bacillus megaterium ATCC 9885	37
Lactobacillus casei DSM 20011 (LB_5)	30	*Cryptococcus curvatus* ATCC 20509 (CR)	28	*Klebsiella pneumoniae* ATCC 700603	37
Lactobacillus brevis DSM 20054 (LB_6)	30			*Pseudomonas aeruginosa* ATCC 27853	37
Lactobacillus acidophilus DSM 20079 (LB_7)	37			*Pseudomonas marginalis* DSM 13124	37
				Brochotrix thermosphacta CIP 103251	28

II.6. Antibacterial activity of polyphenolic extracts.

After strains incubation at their optimal temperature on nutrient agar medium, young colonies were subsequently used for inoculum preparation followed by well homogenization in sterile physiological water and ajustement to an optical density in range of 0.08 to 0.10 at 625 nm, which corresponds approximatively to a concentration of 10^6 to 10^7 CFU/ml.

Disc diffusion method was performed on Mueller-Hinton agar (MHA) medium. Discs of 6 mm in diameter were impregnated with 20 µl of the different phenolic extracts, previously reconstituted in 10% DMSO and they were gently placed on MHA. Discs impregnated with DMSO served as a negative control. Antimicrobial tests were incubated at the optimum temperature for 24 h, each test was repeated twice under the same conditions of experimentation. Conventional statistical methods were used. All data are presented as mean ± standard deviation to calculate averages and standard deviations.

III. Results and discussion

III.1. Olive mill wastewaters bioconversion into oleaginous yeasts biomass

OMWW contain a variety of assimilable carbon sources and dietary fibers, thus it could be considered as a valuable nutritional source for microbial growth and mainly those are of biotechnological and health interest. Unlike the result of Yousuf *et al*. [29], yeast strains were not able to grow in the presence of undiluted OMWW without enrichement with organic supplements (table 3, 4 and 5). The cells number decreased after fermentation, which is likely to be related to the lack in oxygen (asphyxiation) and the inhibition exerted by the high concentration of polyphenolic compounds [27]. Regarding to the diluted OMWW without enrichement, cells number for *Yarrowia lipolytica* and *Cryptococcus curvatus* increased to reach a biomass yield of 5.6×10^4 CFU/ml and 5.3×10^5 CFU/ml respectively. This achievement is a great interest for further target metabolits with biotechnological interest such us lipids. As well rewied, oleaginous yeasts mainly *Yarrowia lipolytica and Cryptococcus curvatus* may have several applications in biotechnological industry [18, 19, 20, 21, 22]. Moreover, around 30 yeasts strains were identified as oleaginous [23] and the most reported productive ones are *Lipomyces starkeyi*, *Rhodosporidium toruloides*, *Cryptococcus curvatus* and *Yarrowia lipolytica* [21, 22, 24, 25, 26]. When OMWW medium is enriched with inexpensive organic supplements, such as glycerol or glucose, yeast biomass increased. *Yarrowia lipolytica* have shown a remarkable growth compared to *Cryptococcus curvatus*. The cell growth yield of *Yarrowia lipolytica* and *Cryptococcus curvatus* on diluted OMWW and enriched with glucose were respectively 1.1×10^5 CFU/ml and 8.9×10^4 CFU/ml for the two strains, whereas the best values were recorded on the medium enriched with glycerol and were respectively 5.3×10^5 CFU/ml and 5.2×10^5 CFU/ml. The study conducted by Dourou *et al*. [27] on diluted OMWW enriched with glycerol and fermented by *Yarrowia lipolytica*, have shown that this strain consumes preferentially glycerol for OMWW bioconversion into mannitol and / or citric acid. It can also convert it into biomass rich in protein of high nutritional value. *Yarrowia lipolytica* is a nonpathogenic dimorphic aerobic yeast with ability to grow in hydrophobic environments thus allowing this yeast to metabolize triglycerides and fatty acids as carbon sources leading to biomass growth. From the tables 3 and 6, in the experiments undertaken using raw enriched OMWW and diluted one, the medium led to the decrease in cell number after incubation time, thus only cell survival was detected.

Table 3. *Cell growth (CFU/ml) assessment on OMWW1 and OMWW2 (*) for lactobacilli.*

Culture medium	Initial pH	Strain	Produced biomass yield	Final pH
M-Glu	4.91	Y	0	4.1
		CR	0	4.2
M-Gly	4.85	Y	0	4.11
		CR	0	4.2
DM	4.9	Y	5.6×10^4	4.79
		CR	5.3×10^5	4.77
DM-Glu	4.77	Y	1.1×10^5	4.01
		CR	8.9×10^4	4.3
DM-Gly	5.77	Y	5.3×10^5	4.05
		CR	5.2×10^5	4.09
M-MRS	5.22	LB_4	6.5×10^7 $8.6 \times 10^{8*}$	4.02
		LB_5	2.6×10^6 $6.7 \times 10^{6*}$	4
DM	5.17	LB_4	0	5.1
		LB_5	0	5.01

Table 4. *Produced biomass (CFU/ml) of Yarrowia lipolytica on OMWW1.*

Cultivation medium	Final biomass	Produced biomass yield
M-Glu-Y	$2.31 \times 10^4 \pm 31$*	0
M-Gly-Y	$2 \times 10^3 \pm 0$*	0
DM-Y	$1.47 \times 10^5 \pm 32$	0.56×10^5
DM-Glu-Y	$2.05 \times 10^5 \pm 10$	1.1×10^5
DM-Gly-Y	$6.5 \times 10^5 \pm 7$	5.3×10^5

(*) Cell survival

Table 5. *Produced biomass (CFU/ml) of Cryptococcus curvatus on OMWW1.*

.

Medium	Final biomass	Produced biomass yield
M-Glu	$1.4\times10^4 \pm13$*	0
M-Gly	$2.4\times10^3 \pm5$*	0
DM	$6.15\times10^5 \pm6$	5.3×10^5
DM-Glu	$2.02\times10^5 \pm 3$	8.9×10^4
DM-Gly	$6.3\times10^5 \pm14$	5.2×10^5

(*) Cell survival

Yarrowia lipolytica cultivation on raw enriched OMWW led to the decrease in biomass but reduced the polyphenol concentration from 252.6 to 62.6 and 213 to 49.5 mg/g TS, respectively for M-Glu and M-Gly media. Biomass increased in DM and diluted enriched OMWW, DM-Glu and DM-Gly, for both yeast strains.

A decrease in pH values are also recorded in all cases (table 3), this is probably associated to the cell growth and a subsequent acidification. Medium acidification and polyphenol removal where no growth was recorded or under stress condition such as raw OMWW, certainly indicate the secretion of enzymatic activities for polymeric organic compounds and polyphenol degradation or glucosidases production which are specialised in deglycosylation of the conjugated polyphenols to polysaccharides. This allow to suggest that this enzymatic activities likely occured to overcome the inhibitory effetct of the complexes toxic substrates. A remarkable growth was noticed for *Yarrowia lipolytica* in addition to polyphenol removal on DM from 207.6 to 43.5. Polyphenol reduction is mainly pronouced for the same strain in raw (M) and diluted OMWW without enrichment (DM). Similar results were reported by Dourou *et al*. [27] with a remarkable phenolic removal by strains of *Yarrowia lipolytica* and other yeasts strains like *Candida tropicalis* and *Saccharomyces cerevisiae*.

Cryptococcus curvatus also led to the decrease in polyphenol concentration when cultivated in M-Gly and DM media. Beside, enrichment in diluted OMWW led to biomass production with only slight polyphenol reduction In should be conluded that a significant growth were recorded for yeast strains as well as probiotic bacteria (table 3, 4, 5, 6, 7). The growth of yeasts produced a significant reduction in polyphenolic content. When a feed stock was diluted, a significant increase in growth was noticed leading to a lower level in phenolic compounds content. *Yarrowia lipolytica* seem to be interesting for further investigation.

Table 6. *Polyphenol concentrations expressed in mg of gallic acid equivalent/g TS*

Sample	Polyphenol (mg EAG/g TS)	Sample	Polyphenol (mg EAG/g TS)
M	274±6	**DM-Glu-Y**	102.6±8.0
M-Glu	252.6±1	**DM-Glu-CR**	120.4±3.2
M-Glu-Y	62.6±3.6	**M-MRS**	205.5±1.5
M-Glu-CR	195.7±1.9	**M-MRS-LB$_5$**	102.8±1
M-Gly	213.1±2.3	**M-MRS-LB$_4$**	105.5±8.3
M-Gly-CR	134.9±3.1	**DM**	199.9±15.3
M-Gly-Y	49.5±2.5	**DM-LB$_5$**	150.9±1.5
DM-Gly	165.5±5.5	**DM-LB$_4$**	180.6±2.4
DM-Gly-CR	136.5±3.5	**DM**	207.6±3.6
DM-Gly-Y	145.3±4.7	**DM-Y**	43±5.6
DM-Glu	136.7±2.5	**DM-CR**	155.8±0.6

III.2. Olive mill wastewaters bioconversion into probiotic biomass

III.2.1 Experiment trials using OMWW1

This experiment trials describe the effect of OMWW on probiotic bacterial growth and survival over 72 h cultivation time (figure1). As well known, lactic acid bacteria grow well on MRS medium and biomass or metabolites production requires less expensive media. Therefore, the use of low cost substrates or growth media like food processing wastes is a great of interest. In their experiments, Yin et al. [30], the results indicate that kitchen waste could be used for the economic production of probiotics. Beside, several studies focused on the optimization of the growth medium of probiotic strains.The biomass of lactobacilli increased considerably in OMWW enriched with MRS medium compared to MRS without OMWW addition (table 3, 7, 8 and figure 1). In fact, biomass production and polyphenol removal, around 50%, were recorded in M-MRS medium. Biomass yield recorded for *Lactobacillus rhamnosus* was 6.5×10^7

CFU/ml and 2.6 × 10^6 CFU/ml for *Lactobacillus casei*.

Table 7. *Produced biomass (CFU/ml) of Lactobacillus rhamnosus on OMWW1.*

Medium	Final biomass	Produced biomass
M-MRS	$6.7\times10^7 \pm 2$	6.5×10^7
DM	ND	0

Table 8*. Produced biomass (CFU/ml) of Lactobacillus casei on OMWW1.*

Medium	Final biomass	Produced biomass
M-MRS	$2.75\times10^6 \pm 7$	2.6×10^6
DM	$1.55\times10^2 \pm 49.5$*	0

(*) Cell survival

III.2.2. Experiment trials using OMWW2

As displayed in figure 1, probiotic bacteria were able to survive and proliferate in the presence of OMWW which is well documented to be highly resistant to biodegradation, thus difficult to use for biological conversion because of the antimicrobial effect of their phenolic compounds [31, 32].

OMWW exhibit a growth promoting effect on probiotic bacteria in both sapmles, press modern an three phase system OMWW, thus suggesting that diluted OMWW incorporation in food might be proposed for it's benificial Human health effects in a regard to probiotic growth stimulating in addition to antioxidant activities and antidiabetic effect recently reported by Senani-Oularbi *et al*. [33]. In addition, Ramos *et al*. [34] demonstrated the breast cancer antiproliferative effect of olive oil by-products hydroxytyrosol-rich extracts. The current findings are in agreament to the study of Giavasis *et al*. [35]. These researchers demonstrated that polyphenol from OMWW in liquid culture media exerted a clear stimulation for certain lactobacilli. Moreover, as described below, bioconversion process decreased polyphenolic content, this could be due to the change in polyphenolic profil, cell wall adsorption or depolymerisation by probiotic bacteria leading to benificial nutritional value if used for health [36, 37, 38, 39]. In fact, additional advantages were recently reported on OMWW, hypoglycemic and prebiotic effects have been stated, respectively for diluted OMWW, polyphenol and their purified polysaccharide extracts [4, 33]. Nadour *et al*. [4] reported the antioxidant and prebiotic activities of polysaccharidic fractions from OMWW as well as their ability to be fermented by lactobacilis while Senani-Oularbi *et al*. [33] evidenced that OMWW could reduce blood glucose through intestinal glucose transport inhibition after glucose oral administration in mice. Moreover, in their investigation, Gerasopoulos *et al*. [40] used a membrane retentate of OMWW as antioxidant ingredient for piglet feed. A higher total antioxidant capacity, glutathione and catalase activity were recorded both in its blood and tissues, with lower oxidative and stress-induced damage to lipids and proteins, than the control group.

The increase in antioxidant activity after *lactobacillus* fermentation has been already reported by Kachouri *et al*. [41] where a new aromatic structures compounds appeared. From the achieved results, as described above, enriched OMWW1 and OMWW2 didn't exhibit any significant inhibition against the growth of probiotic bacteria mainly *Lactobacillus paracasei Lactobacillus ramnosus*, *Lactobacillus casei*. It should be noticed that the microbial growth of the strains of *Lactobacillus paracasei*, *Lactobacillus ramnosus*, *Lactobacillus casei* is more pronounced in M-MRS medium, compared to the control, it reached respectively a values of 2.69 × 10^8 CFU/ml 3.77×10^{10} CFU/ml and 2,47 ×10^8 CFU/ml in stationary phase after 48 h of incubation time (figure1). This is probably related to a beneficial effect of OMWW due to the presence of metabolisable components as well as the prebiotic polymers which have favorable effect to strains development thus stimulating the growth as well studied by our laboratory colleagues [4]. In agreement to the current results, some studies reported that a combined action of dietary fiber and phenolic compounds stimulated the growth of probiotic cells and inhibited the pathogenic strains. In addition, Lopez *et al*. [42] confirmed the positive effect of tea infusions on probiotic bacteria (*Lactobacillus acidophilus* L10 and *Lactobacillus paracasei* L26) survivial. This is likely related to their need and ability to take its energy from polyphenols bioconversion using β-glucosidase, β-galactosidase and /or rhamnosidase activities in order to survive. These enzymes are involved in the metabolism of glycosylated flavonols [42, 62]. However and according to Lara-villoslada *et al*. [43] high phenolic acid concentrations (1000 mg/ml) might limit the in vivo viability of Lactobacilli as noticed in the current investigation when using raw or diluted OMWW for probiotic strains cultivation. Moreover, Nadour *et al*. [4] stated that the strains of *Lactobacillus gasseri, Lactobacillus acidophilus*, *Lactobacillus brevis* and *paracasei* have shown a difficulty in growth on dietary fiber, alcohol-insoluble residue and soluble fraction extracted from OMWW which was propably related to the bactericidal effect of the residual phenolic compounds in the polysaccharidic extracts. *Lactobacillus plantarum* and *Lactobacillus paracasei* have been also assessed for OMWW treatment and led to polyphenols removal in range of 46% and 50% respectively [44, 45].

Beside, as reported by Avila *et al*. [46] Lactobacilli strains like *Lactobacillus plantarum* and *Lactobacillus casei*, as well known, were able to release and express β-glucosidase activity, which

could play an important role in diethary fiber hydrolysis as well as polyphenols liberation from those are conjugated to polysacharides and fats. Regarding to *Lactobacilus Bevis*, cells number increased to reach a value of 8.1×10^7 CFU/ml in the stationary phase after 48 h in M-MRS medium against a maximum value of 2.41×10^8 CFU/ml recorded after 24h in MRS which decrease over time to a final value of $1.62.10^7$ CFU/ml. This behaviour could be related to microbiocidic effect of the released phenolic compounds from their inactive conjugated form to the active one. In fact, Cueva *et al*. [47] investigated the antimicrobial activity of phenolic acids against a panel of bacteria, lactobacilli (*Lactobacillus paraplantarum* LCH7, *Lactobacillus plantarum* LCH17, *Lactobacillus fermentum* LPH1, *Lactobacillus brevis* LCH23, *Lactobacillus coryniformis* CECT 5711, *Lactobacillus coryniformis* CECT 5711) and pathogens (*Staphylococcus aureus* EP167 and *Candida albicans* MY1055). These authors noticed that all the strains studied were sensitivities towards the phenolic compounds and only slight differences were observed between their established growth curves. It should be emphazised that even the polyphenolic concentrations decreased their bioactivities, antibacterial and antioxidant activites (the later not reported here), could remain intersting as it is described below. Thus, it can be concluted that these bioactivities are not only related to polyphenol concentration in the extracts but also to their synergetic effect and qualitatif profils which might remain important after fermentation.

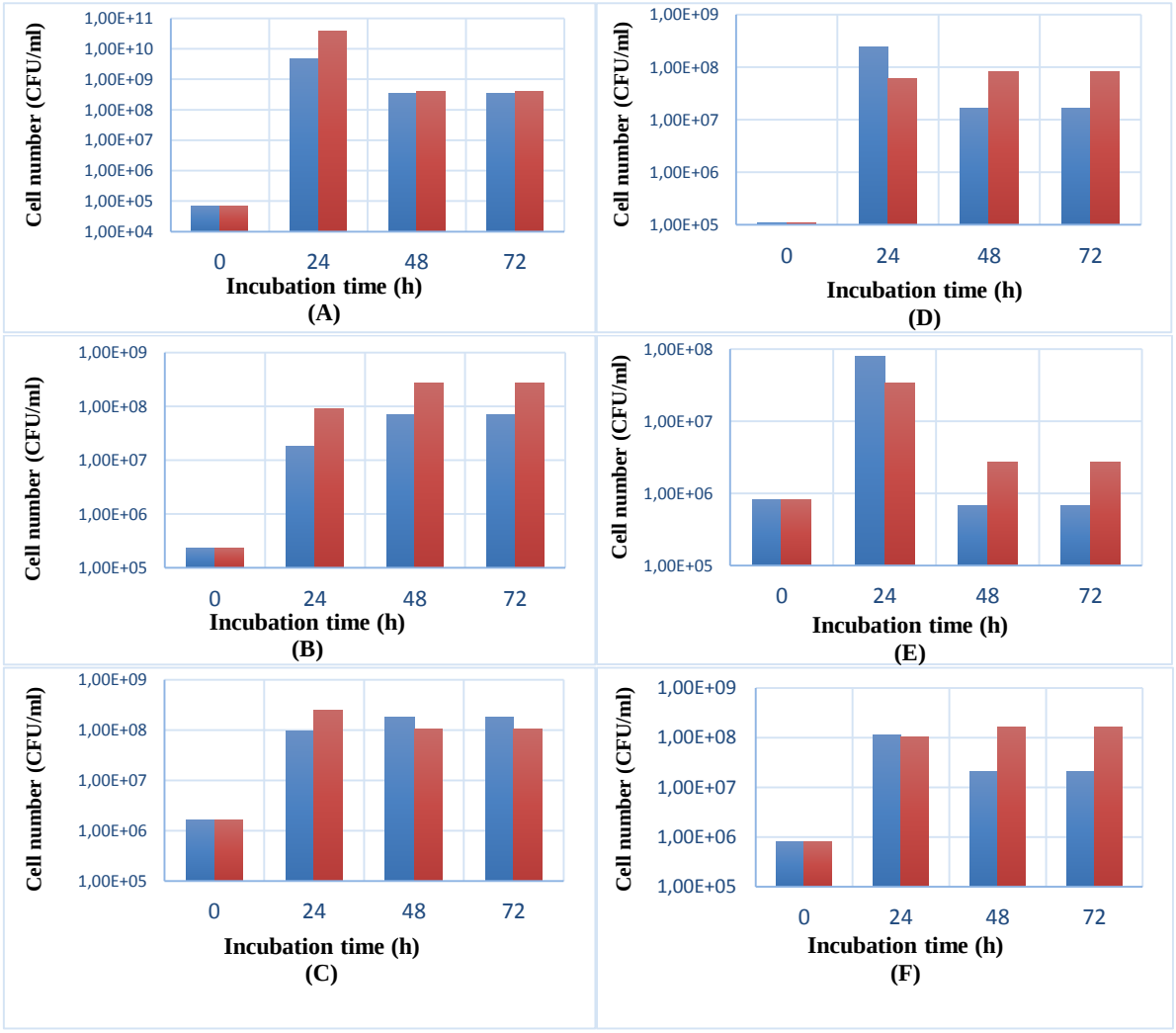

Figure 1. *Probiotic growth on MRS enriched with OMWW2. MRS control (blue). MRS-OMWW (red). (A) Lactobacillus rhamnosus ; (B) Lactobacillus paracasei ; (C) Lactobacillus casei (D) ; Lactobacillus brevis ; (E) Lactobacillus acidophilus ; (F) Lactobacillus gasseri.*

III.3. Antibacterial activities

Antimicrobial effect of polyphenolic extracts before and after press traditional OMWW fermentation was observed for the overall polyphenolic extracts against a panel of bacterial strains. The *in vitro* tests reveals an interesting antimicroial effect of OMWW1 extracts. Several studies reported the antimicrobial effect of olive polyphenols [2, 48, 49, 51, 52] as well as antifungal and antimycotoxines activities [16, 49, 50, 52, 53]. In fact, the stronger inhibitory effect of polyphenols could be attributed to their acidic side chain, which makes easy their own transport trough the cell membrane due to the low polarity. Polyphenols could affect membrane lipids by a neutralization of the membrane's electric potential following penetration of the molecule [32]. Antibacterial activities are related to the phenolic acid especially caffeic acid, vanillic acid, p-coumaric acid, and 4-hydroxybenzoic acid [54]. Tafesh *et al*. [55] suggested that the use of a combination of polyphenols extracted from OMWW is effective against several human pathogens. The inhibition zones displayed in tables 9 a and 9 b, show that the antibacterial activities of the extracts were slightly different against the used bacterial strains.

As it can be seen, OMWW1 polyphenolic extracts are effective against Gram-positive bacteria with diameter zones between 8.5 and 13.5 mm. The maximal inhibition zones were recorded against *Staphylococcus aureus* ATCC 25923 (12 mm) and *Bacillus cereus* ATCC 14579 (10 mm) while the minimal inhibition zones were noticed against *Enterococcus faecalis* ATCC 49452 (9.5 mm) and *Bacillus megaterium* ATCC 9885 (10 mm). It must be emphazised that the poor results performance of disc method could be attributed to low diffusion of the extract on the agar surface. In a regard to Gram-negative bacteria, the extracts exerted an inhibition zone of 10 mm against *Pseudomonas aeruginosa* ATCC 27853 whereas it didn't exceed 9 mm against *Klebsiella pneumoniae* ATCC 700603. Abu-lafi *et al*. [52] recorded diamater zones in range of 23-25 against *Staphylococcus aureus* and *Echerichia coli*. The extract from fermented M-MRS with *Lactobacillus casei* gave a remarkable inhibition against *Brochotrix termosphacta* with a value of 13.5 mm. This is certainly due to the different chemical agents present in the extracts as well as the combined action (synergy) of different phenolic compounds.

Raw OMWW were reported being highly toxic to both phytopathogenic *Pseudomonas syringae* pv. *Savastanoi* (Gram-negative), *Corynebacterium michiganense* (Gram-positive), *Clavibacter Michiganensis subsp*. and showed growth inhibition and bactericidal activity [56, 57].

Other investigations stated that *Staphylococcus aureus* and *Lysteria monocytogenes* have a higher sensitivity to phenols derived from several olive matrices (e.g olive oil, olive leaf and purified compounds [58, 59]. Esmail *et al*. [60] stated the antibacterial effects of raw concentrated OMW against reference and uropathogenic strains and Belaqziz *et al*. [61] observed inhibitory effect of ethyl acetate extract against *Staphylococcus aureus* (DPC5246).

Table 9(a). *Inhibition zones diameters exhibited by OMWW1 polyphenolic extracts against the food spoiling and pathogenic bacterial strains (means ± SD).*

	Klebsiella pneumoniae	***Pseudomonas aeruginosa***	***Pseudomonas marginalis***	***Brochotrix thermosphacta***
M	11.5±0	8.5±1.5	10±0	12.5±0
M-Glu	7±0.56	10±0	-	7±0
M-Glu-Y	7±0.56	10±0	7±0	8±0
M-Glu-CR	6.5±0.49	10±0	-	8±0
M-Gly	8.5±0.5	9.5±0.5	-	7.5±0.5
M-Gly-CR	8.5±0.5	8.5±0.5	-	-
M-Gly-Y	9±0	9.5±0.5	-	7±0
DM-Gly	-	8.5±0.5	-	-
DM-Gly-CR	-	7.5±0.5	7±0	-
DM-Gly-Y	-	7.5±0.5	-	-
DM-Glu	6.5±0.49	9±0	7.5±0	7.5±0.5
DM-Glu-Y	6.5±0.49	7.5±0.63	7±0.56	7.5±0.5
DM-Glu-CR	7±0.56	9±0	7.5±0.63	9±1
M-MRS-LB$_4$	7±0.56	7.5±0.5	7.5±0.63	7.5±0.63
M-MRS-LB$_5$	7.5±0.63	7.5±0.5	7.25±0.60	13.5±0
DM-LB	7.5±0.63	10±0	7.5±0.63	8±0
DM-LB$_5$	7.5±0.63	9±0	7±0.56	8±0
DM-LB$_4$	7±0.56	9.5±0.5	6.75±0.53	8.5±0.5
DM	7±1	10±0	7.5±0.5	8.5±0.5
DM-Y	8.5±0.5	10±0	7.75±2.5	8±1
DM-CR	6.5±0.5	9.5±0.5	6.75±0.25	8±1

Table 9(b). *Inhibition zones diameters exhibited by OMWW1 polyhenolic extracts against the food spoiling and pathogenic bacterial strains (means ± SD)*

	Staphylococcus aureus	***Bacillus cereus***	***Enterococcus faecalis***	***Bacillus megaterium***
M	12±0	10±0	9±0.5	9±0.5
M-Glu	8.5±0.5	9.5±0.5	9±0	-
M-Glu-Y	10±0	10±0	9.5±0.5	6.5±0.5
M-Glu-CR	11±1	9.5±0.5	8.5±0.6	-
M-Gly	8.5±0.5	7±0.5	9±0	7.5±0.5
M-Gly-CR	8.5±0.5	8.5±0.5	7±0	7±0.5
M-Gly-Y	11±1	9.5±0.5	9±1	8.25±0.75
DM-Gly	7.5±0.5	8±0	-	-
DM-Gly-CR	7.5±0.5	9±1	-	-
DM-Gly-Y	7.5±0.5	8±0	-	-
DM-Glu	9.5±0.5	8.5±0.5	-	7.75±0.25
DM-Glu-Y	8±0	8.5±0.5	-	7±0
DM-Glu-CR	9±0	9±0	9±0	7.75±0.25
M-MRS-LB$_4$	7±0.5	9.5±0.5	8±0	9±0
M-MRS-LB$_5$	8.5±0.5	8.5±0.5	8±0	10±0.5
DM-LB	9.5±0.5	9.5±0.5	8±0	6.5±0.5
DM-LB$_5$	9±0	9±1	8±0	7±0.5
DM-LB$_4$	8.5±0.5	8.5±1.5	7.5±0.5	6.75±0.5
DM	7.5±0.6	9±0	7±0	8.5±0.5
DM-Y	10±0	9.5±0.5	7.5±0.5	8.25±0.75
DM-CR	9.5±0.5	9±1	7.5±0.5	7.5±0.5

IV. Conclusion

Olive mill wastes are sources with a wide array of biological exploitation. Diluted and enriched OMWW allowed the growth and survival of oleaginous yeasts and probiotic bacteria. Thus OMWW could be proposed as feedstock for oleaginous yeasts as well as a food matrix enrichment for probiotics food products in addition to their benificial health polyphenols. Bioconverion technology of diluted or enriched OMWW by low cost substrates, glucose or glycerol, could lead to microbial biomass or biomolecules production with biotechnological interest. Moreover, when a feed stock was diluted or enriched, a significant growth was noticed leading to phenolic compounds removal. However, the current investigation is a proof of concept that need further detailed study to ensure sufficient and fast growth in a larg scal process. In addition, the obtained results revealed that OMWW could be considred as a benificial source of bioactive compounds, especially polyphenols. The use of natural phenolic compounds rather than the toxic synthetic antimicrobial and antioxidants biopreservatifs in food control, medical device is in great of interest. Other pertinent considerations need to be considered when these products will added to the food such as organoleptic traits and incorporation method. The extraction of valuable compounds from yeasts grown on OMWW, such as lipids, including phenolic compounds, could represent an interesting biovalorization route for these agro-industrial by-products, leading to high added value products. In addition, it would be advantageous to characterize qualitatively the residual phenolic compounds present in these by-products by chromatographic coupled to mass spectrometry method.

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V. References

1. COI (Conseil oléicole international). Huiles d'olive, Production. Newsletter – marché oléicole 121 (2017) 1-5.
2. Janakat, S.; Al-nabulsi, A.A.R.; Allehdan, S.; Olaimat, A.N.; Alan, H.R. Antimicrobial activity of amurca (olive oil lees) extract against selected foodborne pathogens. *Food Science and Technology* 35 (2015) 259-265.

3. Dermeche, S.; Nadour, M.; Larroche, C.; Moulti-Mati, F.; Michaud, P. Olive mill wastes biochemical characterizations and valorization strategies. *Process Biochemistry* 48 (2013)1532-1552.
4. Nadour, M.; Laroche, C.; Pierre, G.; Delattre, C.; Mati, F.; Michaud, P. Structural characterization and biological activities of polysaccharides from olive mill wastewater. *Applied Biochemistry and Biotechnology* 177 (2015) 431-45.
5. Galanakis, C.M.; Kotsiou, K. Recovery of Bioactive Compounds from Olive Mill Waste. Book chapter in Olive Mill Waste Recent Advances for Sustainable Management (2017) 205-229.
6. Komnitsas, K.; Modis, K.; Doula, M.; Kavvadias, V.; Sideri, D.; Zaharaki, D. Geostatistical estimation of risk for soil and water in the vicinity of olive mill wastewater disposal sites. *Desalination and Water Treatment* 57 (2016) 2982-2995.
7. McNamara, C.J.; Anastasiou C.C.; O'Flaherty, V.; Mitchell, R. Bioremediation of olive mill wastewater. *International Journal of Biodeterioration and Biodegradation* 61 (2008) 127-134.
8. Daassi, D.; Lozano-Sanchez, J.; Borras, I.; Lassaad, B.; Woodward, S.; Mechichi, T. Olive oil mill wastewaters: phenolic content characterization during degradation by *Coriolopsis gallica*. *Chemosphere* 113 (2013) 62-70.
9. Rodriguez, M. M.; Pérez, J.; Ramos-Cormenzana, A.; Martinez, J. Effect of extracts obtained from olive oil mill waste on *Bacillus megaterium* ATCC 33085. *Journal of Applied Bacteriology* 64 (1988) 219-222.
10. Capasso, R.; Cristinzio, G.; Evidente, A.; Scognamiglio, F. Isolation, spectroscopy and selective phytotoxic effects of polyphenols from vegetable wastewaters. *Phytochemistry* 31 (1992) 4125-4128
11. Marsilio, V.; Campestre C.; Lanza, B. Phenolic compounds change during California-style ripe olive processing. *Food Chemistry* 74 (2001) 55-60.
12. Galanakis, C.M.; Tornberg, E.; Gekas, V.A study of the recovery of the dietary fibres from olive mill wastewater and the gelling ability of the soluble fibre fraction. *LWT-Food Science and Technology* 43 (2010) 1009-1017.
13. Galanakis, C. M.; Tornberg, E.; Gekas, V. Recovery and preservation of phenols from olive waste in ethanolic extracts. *Journal of Chemical Technology and Biotechnolo*gy 85 (2010) 1148-1155.
14. Galanakis, C.M.; Tornberg, E.; Gekas, V. Clarification of high-added value products from olive mill wastewater. *Journal of Food Engineering* 99 (2010) 190-197.
15. Galanakis, C.M. Olive fruit and dietary fibers: components, recovery and applications. *Trends in Food Science and Technology* 22 (2011) 175-184.
16. Senani-Oularbi, N.; Riba, A.; Moulti-Mati, F. Inhibition of *Aspergillus flavus* growth and aflatoxin B1 production by olive mill wastewater. *Bioscience Research* 15 (2018) 369-380.
17. Soccol, C.R.; de Souza Vandenberghe, L.P.; Spier, M. R.; Pedroni Medeiros, A.B.; Yamaguishi, C.T.; Lindner, J. D. D.; Pandey, A.; Thomaz-Soccol, V. The Potential of Probiotics: A Review. *Food Technology and Biotechnolo*gy 48 (2010) 413-434.
18. Fontanille, P.; Kumar, V.; Christophe, G.; Nouaille, R.; Larroche, C. Bioconversion of volatile fatty acids into lipids by the oleaginous yeast *Yarrowia lipolytica*. *Bioresource Technology* 114 (2012) 443-449.
19. Ledesma-Amaro, R.; Nicaud, J.M. Metabolic Engineering for Expanding the Substrate Range of *Yarrowia lipolytica*. *Trends in Biotechnology* 34 (2016) 798-809.
20. Gonçalves, F.A.G.; Colen, G.; Takahashi, J.A. *Yarrowia lipolytica* and Its Multiple Applications in the Biotechnological Industry. Review Article. *The Scientific World Journal* 2014 (2014) 1-14.
21. Béligon, V.; Poughon, L.; Christophe, G.; Lebert, A.; Larroche, C.; Fontanille, P. Improvement and modeling of culture parameters to enhance biomass and lipid production by the oleaginous yeast *Cryptococcus curvatus* grown on acetate. *Bioresource Technology* 192 (2015) 582-591.
22. Beligon, V. Valorisation d'acides gras volatils issus de fermentation anaerobie par la production de lipides microbiens, precurseurs de biodiesel. Thèse : Alimentation et Nutrition. Universite Blaise Pascal - Clermont-Ferrand II, (2016) France.
23. Ratledge, C. Regulation of lipid accumulation in oleaginous micro-organisms. *Biochemical Society Transaction* 30 (2002) 1047–1050.
24. Papanikolaou, S.; Aggelis, G. Lipid production by *Yarrowia lipolytica* growing on industrial glycerol in a single-stage continuous culture. *Bioresource Technology* 82 (2002) 43-49.
25. Beopoulos, A.; Cescut, J.; Haddouche, R.; Uribelarrea, J. L.; Molina-Jouve, C.; Nicaud, J.M. *Yarrowia lipolytica* as a model for bio-oil production. *Progress in Lipid Research* 48 (2009) 375-387.
26. Beopoulos, A.; Chardot, T.; Nicaud, J.M. *Yarrowia lipolytica*: A model and a tool to understand the mechanisms implicated in lipid accumulation. Biochimie, Lipids for the future from agro-resources to human health. *Biochimie* 91 (2009) 692-696.
27. Dourou, M.; Kancelista, A.; Juszczyk, P.; Sarris, D.; Bellou, S.; Triantaphyllidou, I.; Rywinska, A.; Papanikolaou, S.; Agglis, G. Bioconversion of olive millwastewater into high-added value products. *Journal of Cleaner Production* 139 (2016) 957-969.
28. Singleton, V.L.; Orthofer, R.; Lamuela-Raventos, R. M. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteau reagent. *Methods in Enzymology* 299 (1999) 152-178.
29. Yousuf, A.; Sannino, F.; Addorisio, V.; Pirozzi, D.J. Microbial conversion of olive oil mill wastewaters into lipids suitable for Biodiesel Production. *Journal of Agricultural and Food Chemistry* 58 (2010) 8630-8635.
30. Yin, H.C.; Dong, X.L.L.; Wang, Z.G.; Xu Q.Q.; Liu X.L.; Yan, H. Economic production of probiotics from kitchen waste. *Food Science and Biotechnology* 22 (2013), 59-63.
31. Zirehpour, A.; Rahimpour, A.; Jahanshahi, M.; Peyravi, M. Mixed matrix membrane application for olive oil wastewater treatment: process optimization based on Taguchi design method. *Journal of Environmental Management* 132 (2014) 113-120.
32. Larif, M.; Ouhssine, M.; Soulaymani, A.E.; Elmidaoui, A. Potential effluent oil mills and antibacterial activity polyphenols against some pathogenic strains. *Research on Chemical Intermediates* 41 (2013) 1213-1225.
33. Senani-Oularbi, N.; Moulti-Mati, F.; Letteron, P.; Ducroc, R.; El benna, J.; Marie, J.C. Hypoglycemic effect of olive mill wastewater in mice involves the sglt-1 transporter. *International Journal of Pharmacy and Pharmaceutical Sciences* 9 (2016) 73-78.
34. Ramos, P.; Santosc, S.A.O.; Guerra, Â.R.; Guerreiro, O.; Felício, L.; Jerónimo, E.; Silvestre, A.J.D.; Netoc, C.P.; Duarte, M. Valorization of olive mill residues: Antioxidant and breast cancer antiproliferative activities of hydroxytyrosol-rich extracts derived from olive oil by-products. *Industrial Crops Products* 46 (2013) 359-368.
35. Giavasis, I.; Tsante, E.; Goutsidis, P.; Papatheodorou, K.; Petrotos, K. Stimulatory effect of novel polyphenol-based supplements from olive mill waste on the growth and acid production of lactic acid bacteria. *In Microbes in Applied Research: Current Advances and Challenges* (2012) 308-3012.

36. Tsangalis, D.; Ashton, J. F.; McGill, A.E.J.; Shah, N.P. Enzymic transformation of isoflavone phytoestrogens in soymilk by b-glucosidase-producing *Bifidobacteria*. *Journal of Food Science* 67 (2002) 3104-3113.
37. Aura, A. M. Microbial metabolism of dietary phenolic compounds in the colon. *Phytochemistry Reviews* 7 (2008) 407-429.
38. Donkor, O.N.; and Shah, N.P. Production of b-glucosidase and hydrolysis of isoflavone phytoestrogens by *Lactobacillus acidophilus*, *Bifidobacterium lactis*, and *Lactobacillus casei* in soymilk. *Journal of Food Science* 73 (2008) 15-20.
39. Uskova, M.A.; Kravchenko, L.V.; Avrenjeva, L.I.; Tutelyan, V.A. Effect of *Lactobacillus casei* 114001 probiotic on bioactivity of rutin. *Bulletin of Experimental Biology and Medicine* 149 (2010) 578-582.
40. Gerasopoulos, K.; Stagos, D.; Petrotos, K.; Kokkas, S.; Kantas, D.; Goulas, P.; Kouretas, D. Feed supplemented with polyphenolic byproduct from olive mill wastewater processing improves the redox status in blood and tissues of piglets. *Food and Chemical Toxicology* 86 (2015) 319-327.
41. Kachouri, F.; Ksontini, H.; Kraiem, M.; Setti, K.; Mechmeche, M.; Hamdi, M. Involvement of antioxidant activity of *Lactobacillus plantarum* on functional properties of olive phenolic compounds. *Journal of Food Science and Technology* 52 (2016) 7924-7933.
42. Lopez De Lacey, A.M.; Perez-Santin, E.; Lopez-Caballero, M.E.; Montero, P. Survival and metabolic activity of probiotic bacteria in green tea. *LWT- Food Science and Technology* 3 (2013) 1-9.
43. Lara-villoslada, F.; Olivares, M.; Sierra, S.; Rodriguez, J.M.; Boza, J.; Xaus, J. Beneficial effects of probiotic bacteria isolated from breast milk. *British Journal of Nutrition* 98 (2007) 96-100.
44. Ayed, L. and Hamdi, M. Fermentative decolorization of olive mill wastewater by *Lactobacillus Plantarum*. *Process Biochemistry* 39 (2003) 59-65.
45. Aouidi, F.; Gannoun, H.; Ben Othman, N.; Ayed, L.; Hamdi, M. Improvement of fermentative decolorization of olive mill wastewater by *Lactobacillus paracasei* by cheese whey's addition. *Process Biochemistry* 44 (2009) 597-601.
46. Avila, M.; Hidalgo, M.; Sánchez-moreno, C.; Pelaez, C.; Requena, T.; Pascual-Teresa, S.D. Bioconversion of anthocyanin glycosides by *Bifidobacteria* and *Lactobacillus*. *Food Research International* 42 (2009) 1453-1461.
47. Cueva, C.; Moreno-arribas, M.V.; Martín-Álvarez, P. J.; Bills, G.; Vicente, M.F.; Basilio, A.; López, C.R.; Requena, T.; Rodríguez J.M.; Bartolom, B. Antimicrobial activity of phenolic acids against commensal, probiotic and pathogenic bacteria. *Research in Microbiology* 161 (2010) 372-382.
48. Boskou, D. Sources of natural phenolic antioxidants. *Trends in Food Science and Technology* 17 (2006) 505-512.
49. Obied, H.K.; Bedgood, D.R.J.; Prenzler, P.D.; Robards, K. Bioscreening of Australian olive mill waste extracts: biophenol content, antioxidant, antimicrobial and molluscicidal activities. *Food and Chemical Toxicology* 45 (2007) 1238-1248.
50. Yangui, T.; Sayadi, S.; Gargoubi, A.; Dhouib, A. Fungicidal effect of hydroxytyrosol rich preparations from olive mill wastewater against *Verticillium dahliae*. *Crop Protection* 29 (2010) 1208-1213.
51. Carraro, L.; Fasolato, L.; Montemurro, F.; Martino, M. E.; Balzan, S.; Servili, M.; Novelli, E.; Cardazzo, B. Polyphenols from olive mill waste affect biofilm formation and motility in *Escherichia coli* K-12. *Microbial Biotechnology* 7 (2014) 265-275.
52. Abu-Lafi, S.; Al-Natsheh M. S.; Yaghmoor, R.; Al-Rimawi, F. Enrichment of Phenolic Compounds from Olive Mill Wastewater and *In Vitro* Evaluation of Their Antimicrobial Activities. *Evidence-Based Complementary and Alternative medicine* 2017 (2017) 1-9.
53. Lagrouh, F.; Dakka, N.; Bakri, Y. The antifungal activity of Moroccan plants and the mechanism of action of secondary metabolites from plants. *Journal de Mycology Medical* 27 (2017) 303-311.
54. Aziz, N.H.; Farag, S.E.; Mousa, L.A.; Abo, M.A. Comparative antibacterial and antifungal effects of some phenolic compounds. Microbios. 93(1998) 43-54.
55. Tafesh, A.; Najami, N.; Jadoun, J.; Halahlih, F.; Riepl, H.; Azaizeh, H. Synergistic antibacterial effects of polyphenolic compounds from olive mill wastewater. *Evidence-Based Complementary Alternative Medicine* 2011 (2011) 1-9.
56. Capasso, R.; Evidente, A.; Schivo, L.; Orru, G.; Marcialis, M.A.; Cristinzio, G. Antibacterial polyphenols from olive oil mill waste waters. *Journal of Applied Bacteriololgy* 79 (1995) 393-398.
57. Özdemir, Z. Growth inhibition of *Clavibacter Michiganensis subsp. Michiganensis* and *Pseudomonas syringae* pv. tomato by olive mill wastewaters and citric acid. *Journal of Plant Pathology* 91 (2009) 221-224.
58. Medina, E.; Antonio, C.D.; Concepcion, R.; Manuel B. Comparison of the concentrations of phenolic compounds in olive oils and other plant oils: correlation with antimicrobial activity. *Journal of Agricultural and Food Chemistry* 54 (2006) 4954-61.
59. Pereira, A. P.; Ferreira, I.C.; Marcelino, F.; Valentão, P.; Andrade, P.B.; Seabra, R.; Estevinho, L.; Bento, A.; Pereira, J. A. Phenolic compounds and antimicrobial activity of olive (*Olea europaea L. Cv. Cobrançosa*) Leaves. *Molecules* 12 (2007) 1153-62.
60. Esmail, A.; Chahboun, N.; Mennane, Z.; Amiyare, R.; Abed, H.; Barrahi, M.; Qebibo, A.; Ouhssine, M.; Berny, E. H. Étude de l'activité antimicrobienne des margines issues de Fès Boulman vis à-vis de souches pathogènes. *Journal of Material and Environmental Science* 6 (2015) 869-876.
61. Belaqziz, M.; El-Abbassi, A.; Lakhal, E.K.; Agrafioti, E.; Galanakis, C.M. Agronomic application of olive mill wastewater: Effects on maize production and soil properties. *Journal of environmental Management* 171 (2016) 158-165.
62. Schneider, H.; Schwiertz, A.; Collins, M.D.; Blaut, M. Anaerobic transformation of quercetin-3-glucoside by bacteria from the human intestinal tract. *Archives of Microbiology* 171 (1999) 81-91.

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