Journée Portes Ouvertes et de la Célébration du 60e Anniversaire de l'Institut National de la Recherche Agronomique d'Algérie (INRAA)

PRELIMINARY MICROBIOLOGICAL VALORIZATION OF OLIVE MILL EFFLUENT THROUGH POLYPHENOLIC BIODEGRADATION

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INTRODUCTION

Olive mill wastewater (OMWW) represents an abundant source of organic residues. These effluents are rich in bioactive compounds, particularly polyphenols, and are attracting increasing interest for their potential use. However, despite their availability in Algeria, these by-products remain largely underutilized, especially in the field of microbial biotechnology. The present study was carried out under ***OLIVENET EU Project*** to enhance the utilization of different OMWW, to promote the proliferation of two (2) strains (Sar 1 and Sar 2) belonging to the *Bacillus* genus and their biosurfactant production.

MATERIAL & METHODS

OMWW Polyphenols Characterization

1- Oil Mills, OMWW Sampling & pH measurement

Traditional oil mill

Modern oil mill

tOMWW — deep beige, pH= 4.468

mOMWW — red-black, pH= 5.026

2- Phenolic compounds Liquid-liquid extraction with Ethyl acetate

Polyphenols extract tOMWW

Polyphenols extract mOMWW

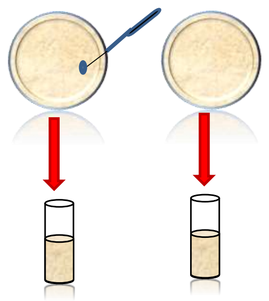
Dionex Ultimate 3000 UHPLC system (Thermo Scientific, USA) equipped with a Diode Array and Multiple-Wavelength Detectors / C18 Luna Omega polar column

3-Qualitative analysis by UHPLC-DAD

Funding

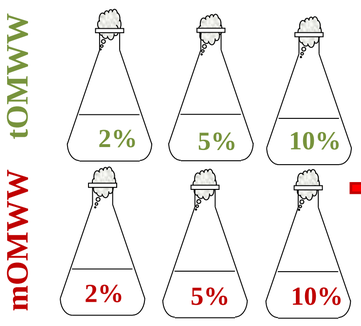
Bacterial Fermentation

1- Bacterial Culture



Bacterial Inoculum

2- Medium Preparation

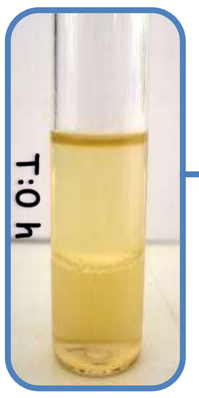


100 ml of mineral salt medium (MSM) with 3 different [OMWW]

3- Fermentation conditions

incubated for 120 hours at 30°C with shaking at 120 rpm

4- Fermentation & biosurfactant production monitoring



(Trouble + gasoil)

Agitation

Kinetic growth

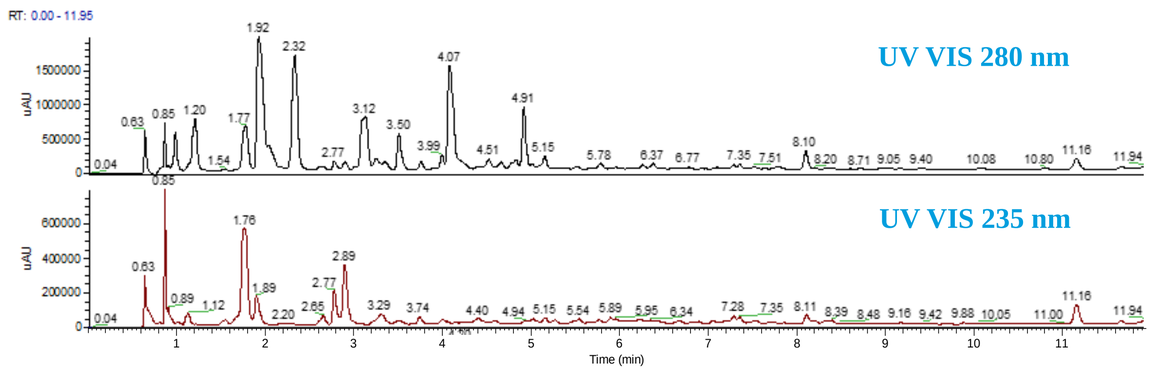
Emulsification Index (E24%)

RESULTS & DISCUSSION

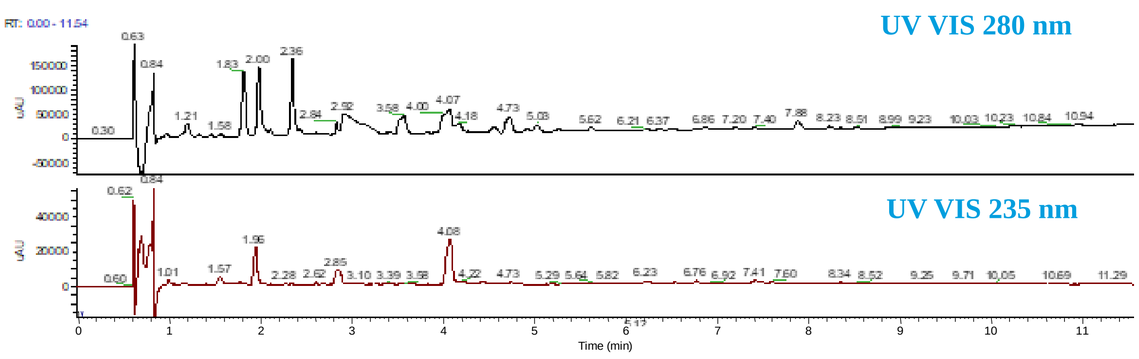
OMWW Polyphenols Characterization

Phenolic composition of **mOMWW** and **tOMWW** extracts analysed by HPLC-MS revealed the presence of several bioactive compounds such as: **phenolic alcohols** (**hydroxytyrosol** & **tyrosol**), **phenolic acids** (**vanillic acid**), **secoiridoids** (**oleuropein**, **3,4-DHPEA-EDA**, **hydroxy-de-carboxymethyl-oleuropein**, **ligstroside** & **elenolic acid**), **flavonoids** (**apigenin** & **luteolin**), and **lignans** (**1-acetoxypinoresinol**)

OMWW-Modern Chromatogram

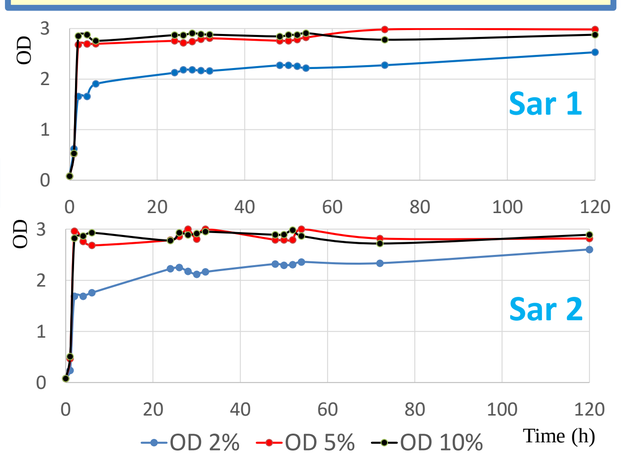


OMWW-Traditional Chromatogram

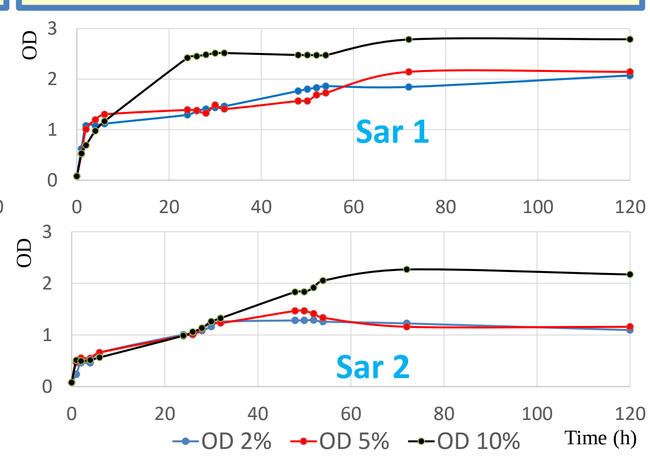


Kinetic growth

OMWW-Modern

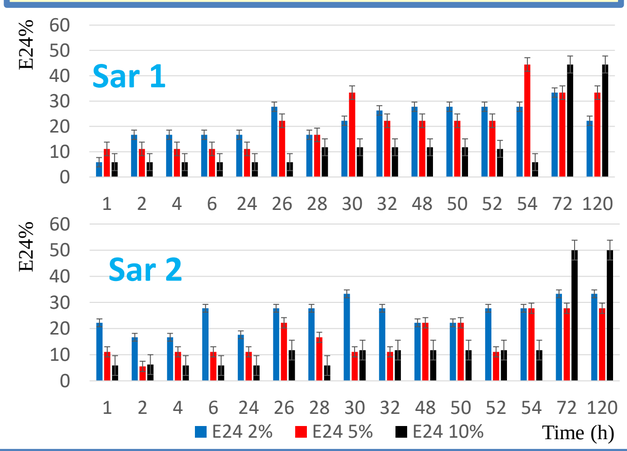


OMWW-Traditional

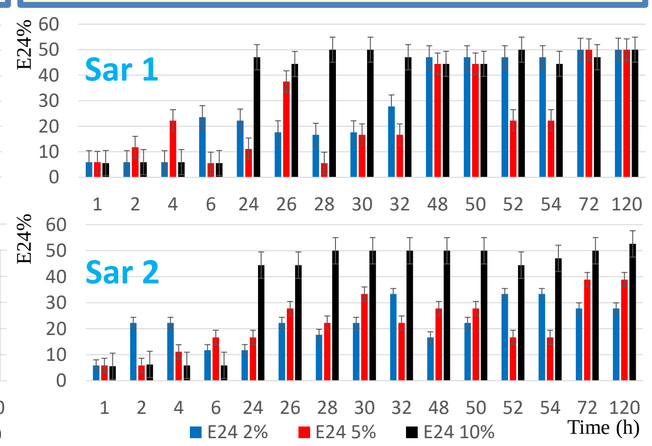


Biosurfactant production

OMWW-Modern



OMWW-Traditional



CONCLUSION

The amount of crude extract of the OMWW samples varies according to the processing technique. Otherwise, the HPLC biophenols shown the presence of numerous bioactive compounds with a higher concentration in **mOMWW** than in **tOMWW**. These findings indicate that the processing technique influences the phenolic composition of OMWW. Moreover, the bacterial fermentation revealed a difference in *Bacillus* strain tolerance on a culture medium with **mOMWW-based medium** promoting bacterial growth as more effectively. Paradoxically, the 10% **tOMWW-based medium** had the strongest emulsifying capacity, with an E_{24}% index close to **50%**, whilst the 2% **mOMWW-based medium** had the best biosurfactant production performance with E_{24}% index of **34%**. This study validates the strains' adaptability and provides encouraging data on their capacity to degrade polyphenolic chemicals found in OMWW.