

Kasdi Merbah University - Ouargla

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Faculty of Natural and Life Sciences

**Department of Biological Sciences** 

### **PROJECT DISSERTATION REPORT:**

In view of obtaining the degree of Academic Master in Biology Specialty: Quality control and safety of food products

### Theme:

Innovative application of waste frying oil for the recovery of bioactive antioxidants from kitchen food waste via supramolecular solvents

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Ouargla, 05/09/2022



### Acknowledgement

Above all, we thank God for giving us strength, courage, and patience, and for allowing us to study and follow the path of science.

This project would not have been possible without the support of many people. Many thanks to the supervising professor, Dr.KEDDAR Mohammed Nadir. We also thank the members of my committee, who provided guidance and support.

And finally, we thank all our families and friends who have endured this long process with us, always giving us

support and love.

Nithout forgetting to thank all the professors at the Department of Biology at the University of Kasdi Merbah, Ouargla.





To the owner of a fragrant biography and enlightened thought, for he had the first credit for my attaining higher education (my beloved father)

To the one who set me on the path of life, and cared for me until I became old

(my dear mother).

To my sisters, who have had a great impact on many obstacles and difficulties.

To whom I rely on in every big and small (my respected only brother Mohamed Ali).

To my grandfather and grandmother, from my father, may Allah bless them

and from my mother, may Allah prolong their life. To the one body, my dear family.

To my friends whom I witness that they are the best companions in all matters:

MOUNIRA, Aziza , RABAB , LINA, ROUMISSA, SAFA ,Aida , ANFAL, HIBA, ASSIA, FATIMA, RAYHANA,

DOHA, SOMIA, KHADIDJA, IBTIHAL and OMAIMA.

To everyone who had a simple reason to help me, without forgetting all my teachers from primary to higher education

To all my friends of quality control and analysis

2021/2022.

Maroua

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### Abbreviations

- **DPPH**: 2,2-dyphenyl-1-picrylhydrazyl
- **EqS:** equilibrium solution
- FOWEA: frying oil-water-Ethyl Acetate
- FOWT: frying oil-water- tetrahydrofuran
- FTIR : Fourier transform infrared spectroscopy
- **O/W**: Oil in Water
- **SD**: standard of deviation
- **SUPRAS:** Supramolecular solvents
- THF: tetrahydrofuran
- ABTS: 2, 2'-Azino-Bis-3-Ethylbenzothiazoline-6-Sulfonic Acid (biochemical reagent
- W/V: weight /volume
- W/W: Weight/ Weight
- WCO: used cooking oil

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### Abstract

### Abstract

In this research project, we have created two new SUPRASs ; FOWEA-SUPRAS and FOWT-SUPRAS using frying oil as an amphiphilic as it contains fatty acids in order to form a new SUPRAS capable of recovering and extracting bioactive molecules as antioxidants from vegetables (polyphenols, flavonoids and carotenoids) from kitchen waste and also to reduce pollution from the accumulation of waste Food, the antioxidant activity value of FOWT-SUPRAS-Extract was recorded  $75.56 \pm 1.50$  % This value is the highest two time according to FOWEA-SUPRAS-Extract 57.02  $\pm$  3.60 %. Through this research, we gained the skills to research and create supramolecular solvents using frying oil while at the same time finding new ways to reduce vegetable residues from cooking.

Keywords: Supramolecular Solvents, Frying Oil, Bioactive Molecules, Antioxidant Activity, Waste Food

### Résumé

Dans ce projet de recherche, nous avons créé deux nouveaux SUPRAS ; FOWEA-SUPRAS et FOWT-SUPRAS, utilisant l'huile de friture comme amphiphile car elle contient des acides gras afin de former un nouveau SUPRAS, capable de récupérer et d'extraire des molécules bioactives des végétaux comme des antioxydants (polyphénols , flavonoïdes et caroténoïdes) des déchets de cuisine et aussi pour réduire la pollution due à l'accumulation de déchets Alimentaire, la valeur d'activité antioxydant de l'Extrait FOWT-SUPRAS a été enregistrée à 75,56 ± 1,50 % Cette valeur est le plus élevé deux fois selon l'Extrait FOWEA-SUPRAS 57,02 ± 3,60 %. Grâce à cette recherche, nous avons acquis les compétences nécessaires pour rechercher et créer des solvants supramoléculaires à l'aide d'huile de friture tout en trouvant de nouvelles façons de réduire les résidus végétaux de la cuisson.

Mots clé : Solvants Supramoléculaires, Huile De Friture, Molécules Bioactives, Activité Antioxydant, Déchets Alimentaire

### ملخص

FOWT- و FOWEA-SUPRAS; SUPRAS و FOWT- و FOWEA-SUPRAS; SUPRAS و SUPRAS و SUPRAS و SUPRAS و SUPRAS باستخدام زيت القلي كمادة برمائية، لأنه يحتوي على أحماض دهنية من أجل تكوين SUPRAS جديد قادر على استعادة واستخراج الجزيئات النشطة بيولوجيًا كمضادات للأكسدة من الخضروات, (البوليفينول، الفلافونويد والكاروتينات) من فضلات المطبخ وأيضًا لتقليل التلوث من تراكم النفايات الغذائية ، تم تسجيل قيمة الفلافونويد والكاروتينات) من فضلات المطبخ وأيضًا لتقليل التلوث من تراكم النفايات الغذائية ، تم تسجيل قيمة نشاط مضادات الأكسدة له من أجل تكوين على مرتين الفلافونويد والكاروتينات) من فضلات المطبخ وأيضًا لتقليل التلوث من تراكم النفايات الغذائية ، تم تسجيل قيمة نشاط مضادات الأكسدة لـ 5.50 FOWT-SUPRAS قوفيًا لـ 5.50 FOWT-SUPRAS و من تراكم التلوث من تراكم النوايات الغذائية ، تم تسجيل قيمة نشاط مضادات الأكسدة لـ 5.50 FOWT-SUPRAS قوفيًا لـ 5.50 FOWT-SUPRAS من تراكم النفايات الغذائية ، تم تسجيل قيمة الفلافونويد والكاروتينات) من فضلات المطبخ وأيضًا لتقليل التلوث من تراكم النفايات الغذائية ، تم تسجيل قيمة الفلافونويد والكاروتينات من فضلات المطبخ وأيضًا لتقليل التلوث من تراكم النفايات الغذائية ، تم تسجيل قيمة في أعلى مرتين الفلافونويد والت الأكسدة لـ 5.50 FOWT-SUPRAS قول القليل التلوث من تراكم النفايات الغذائية ، المام رات الفلافية في أحمان المطبخ ، اكتسبنا المهارات وفقًا لـ المعار و و الفلان و في نفس الوقت إيجاد طرق جديدة لتقليل بقايا الخضروات من الطهى.

الكلمات المفتاحية: المذيبات فوق الجزيئية، زيت القلي ، الجزيئات النشطة بيولوجيًا، نشاط مضادات الأكسدة، النفايات الغذائية

### Introduction

### **INTRODUCTION**

Approximately, 34.22 million liters of used cooking oil (WCO), growing at 2 % is produced worldwide annually (Nusrat et al., 2021), so that frying oil is the main ingredient of fast food. The choice of frying oil depends on cost, stability or shelf life, taste and nutritional content, which is largely determined by the amount of trans and saturated fats currently mandated on food labels (Serna-Saldivar et al., 2019).

Currently, a very small percentage of oils are used in soap production, and this is due to the poor quality of soap produced from frying oil, and most of the frying oil is wasted, creating environmental and economic problems (Trupti and Virendra, 2011).

Vegetable oils and other materials are used for known biological solvents or new through innovative synthesis. Used in manufacturing, pharmaceuticals, cosmetics, chemicals, energy, food, etc. To reduce the impact on the environment, vegetable food waste is a promising resource for bio-solvent production (Glossman-Mitnik and Maciejewska, 2020).So Used cooking oil can be explored ecologically, economically and commercially as an alternative green solvent and resource-rich waste management too (Nusrat et al., 2021).

There is great industry interest in their efficient extraction from plant biomass, but most current methods are neither environmentally friendly nor economical. Supramolecular solvents (SUPRAS) composed of self-assembled amphiphilic aggregates have shown great potential in extracting bioactive compounds from biomass and fulfilling many green chemistry criteria (Gonz'alez-Rubio et al., 2022).

Supramolecular solvents (SUPRASs) are a new term Nanostructured liquids produced from amphiphiles Sequential self-assembly processes at two scales, molecular and nano. This process initially results in three dimensions Aggregates aggregated, in the second stage and Immiscible liquids composed of large supramolecular aggregates disperse in the continuous phase, usually water. (Ballesteros-Gómez Et al., 2010), with multiple binding sites and microenvironments of different polarities for efficient extraction of multiple compounds (Dueñas-Mas et al., 2022).

### INTRODUCTION



(Dueñas-Mas et al., 2022)

### Figure 1 : Schematic picture of the SUPRAS formation and its microstructure

Large quantities of agricultural food by-products, inedible food and waste are produced throughout the supply chain production up to the final consumption stages, compounds with high added value from this biomass are the focus of extensive research over the past decade (Laura, 2020).

In another study, under the optimal conditions, the edge of active constituents uprooted by Octanoic Acid-Ethanol-Water supramolecular solvents were mainly advanced than conventional organic reagents and the OAEW- SUPRAS excerpts also showed a high antioxidant capacity (IC50 = 1.49 mg/ mL) (Zi-Hui et al., 2022).

Also according to (Keddar et al., 2020), the SUPRAS extract showed high antioxidant activity too, by ABTS and DPPH, respectively. SUPRAS is non-toxic and fast (5 min extraction) and cost-effective (room temperature, atmospheric pressure) for recovery of antioxidant compounds from algal biomass, and is a good alternative to using traditional solvents.

So, How Can we valorize frying oil as an amphiphil in order to form a new SUPRAS capable of recovering and extracting antioxidant bioactive molecules from vegetable kitchen waste, and also to reduce pollution due to the accumulation of food waste?

In this study we discuss the use of green chemistry for the valorization of agricultural foods, and it's ability to innovate new SUPRAS solvent replace traditional organic solvents.

The aim of the study is the use SUPRAS for the recovery of the bioactives particularly phenolics from food waste as this technique had already proved its efficiency for such purpose (Zi-Hui et al., 2022). As the formulation of SUPRAS requires an amphiphilic agent, the use of frying oil waste seems to be a good

### **INTRODUCTION**

alternative that meet the requirements without altering the process efficiency, along with ensuring potential valorization of this waste in a circular economy framework.

In the first part, is used to present a general introduction of this study, this is followed by the second part for materials and methods used for obtained our objectives. The third part includes the most important results obtained in this study with discussions. Finally a general conclusion.

#### 1. Materials and methods:

### 1.1. Materials and chimicals :

Waste frying oil, Eyhyl-acétate solvent and THF solvent as well as acidified water (ph = 2.8); 500 ml distilled water contain 0.4 ml HCL, was used to prepare supramolecular solvent.

Elio's frying oil was collected from fast-food restaurants, and kept in a bottle, at room temperature, away from light.

The vegetables food wastes were collected from the house (potatoes, eggplant, oranges, garlic, onions, cabbage, lettuce, carrot, tomato and cauliflower), after which the process of freezing drying and grinding was carried out in the laboratory and kept in the refrigerator in an airtight container.

### **1.2.** Apparatus:

SUPRASs preparations were performed using Classic Advance Vortex Mixer for mixing ingredients and Sigma Centrifuge for accelerating phase separation. The lyophilizer was used to calculate the percentage of SUPRASs components and to dry plant residues by removing water under low temperature and pressure.

Chemical characterization of frying oil was performed using GCMS-TQ8040NX and The Agilent Cary 630 FTIR spectrometer is for determinate the composition of SUPRASs and frying oil. Also, the bioactive content (TPC, TFC and TCC) in SUPRASs extracts and the antioxidant activity were measured by Cary 100 UV-visible spectrophotometry. Light microscopy is used in order to study the microstructure of the synthesized SUPRASs.

### **1.3. Preparation of SUPRASs:**

Acidified water (pH = 2.8) was added to frying oil diluted in different proportions of Ethyl Acetate solvent and THF solvent to make FOWEA-SUPRAS and FOWT-SUPRAS respectively of different compositions while keeping the volume of the mixture tripled to 2 ml.

The ternary mixture was vortexed for 30 sec to homogenize, this mixture was separated into upper and lower two phases after centrifugation 4500 rpm for 10 min, of SUPRASs, and equilibration solution (EqS), respectively.

Finally, the two phases were stored separately in airtight containers at room temperature until extraction.



### Figure 2 : SUPRASs formation

(A): Acidified water + Frying oil + Solvent (B): After Vortex 30s (C): After centrifugation 4500 rpm for 10min)

Each synthetic condition was done in duplicate and the volume of the formed phase should be determined.

### 1.4. Chemical characterization:

### 1.4.1 Ternary phase diagrams of SUPRASs:

### 1.4.1.1 Volume of SUPRASs:

Production of SUPRAS under different synthetic conditions calculate volume by measuring its height in the cylindrical tube with a ruler and using the equation  $V(\mu l) = h.\pi$ . r<sup>2</sup> (Sánchez-Vallejo et al, 2022).



Figure 3: Calcule the Volume of SUPRAS

### 1.4.1.2 The percentages of Water, oil and solvents in SUPRASs:

The content of frying oil, Ethyl Acetate solvent and water in the FOWEA-SUPRAS and the content of frying oil, THF solvent and water in the FOWT-SUPRAS were determined as Weight percent (%w/w), taking into account the volatility of water and solvents. The frying oil content was determined after the evaporation of solvents and water after lyophilization (Zi-Hui et al., 2022).

### **1.4.2** The FTIR spectra :

The FTIR of frying oil, FOWEA-SUPRAS and FOWT-SUPRAS were recorded using Agilent Cary 630 FTIR spectrometer.

Infrared spectroscopy is a widely used method for qualitative and quantitative analysis. It is fast, easy to use, and environmentally friendly. It is frequently coupled with chemometrical methods that allow appropriate spectral handling and models development, and improves the extraction of relevant spectral information. Models were built using three separate spectra for each sample (Tan et al., 2022).

#### **1.4.3 Fatty acids composition:**

The Gas Chromatography (GC/MS) analysis was performed with GCMS-TQ8040NX for the identification of the major fatty acids constituents in fraying oil, FOWEA-SUPRAS and FOWT-SUPRAS in a very short time.

In order to simplify the identification procedure, esterified of the three compounds by an alcohol potassium hydroxide (KOH) titration procedure following the (Gaithersburg, 2002).

The sample was injected to the GC column for separation of the constituents. They were detected by an MS detector and the identification was carried out by reference compounds and MS spectral libraries. Concentrations were calculated as relative peak areas, and fatty acid contents are given in % (Tan et al., 2022).

#### **1.4.4 Extraction and dosage of bioactive molecules :**

### **1.4.4.1 Preparation of vegetables food waste sample : (Drying and Grinding)**

Cut the peels of vegetables and fruits into small pieces: orange, carrot, potato, tomato, eggplant, lettuce, onion, garlic, cabbage, and cauliflower. Then put it in the freezer for a whole day 24 hours.

The next day, put them in a freeze dryer at 80°C for 72 hours. After the peels have dried, they are ground with a grinder (high-performance ball with a circular cooler) at 39-40 ° C for 20 min until a fine powder is obtained.

Finally, sieving to remove remaining impurities and store in a box in the refrigerator until use.

### 1.4.4.2 Preparation of SUPRAs vegetables food waste extract :

The extraction capacity of the different SUPRAS was investigated by extracting phenolic, flavonoids and carotenoids metabolites from vegetables food waste peels that were collected from kitchen house, and blended until homogenized.

First SUPRAs (FOWEA–SUPRAS and FOWT-SUPRAS) were prepared as previously indicated. Then 100 mg of vegetables food waste peels powder was wetted

with 400  $\mu$ L of equilibrium solution and then 400  $\mu$ L for previously THF and Ethyl acetate SUPRAs was added. The mixture was vortex shaken 15 min to favour the extraction and centrifuged (15,000 rpm, 15 min) to accelerate the separation of the SUPRAS phase.

The SUPRAS extract was collected and diluted with propanol, then directly was analysis by UV spectrophotometry for the determination of the content of bioactive molecules.

### 1.4.4.3 Quantification of bioactive molecules in vegetables food waste:

### **1.4.4.3.1** Total Phenolic content (TPC) :

TPC is quantified colorimetrically by the Folin-Ciocalteu assay. Briefly, 2.40 ml of distilled water, 0.1 ml of SUPRAS extract and 0.25 ml of Folin-Ciocalteu reagent are placed into 10 ml-test tubes. After mixing and addition of 0.5 ml of 20 % (W/V) sodium carbonate solution. Distilled water 1.75 ml was added to reach a final volume of 5 ml. All the solutions were mixed and allowed to stand at room temperature in the dark for 1h. The concentration of total polyphenols was determined by absorbance measurements at 725 nm. Gallic acid is used as standard with distinct concentrations ranging from 0.008 to 1 mg ml<sup>-1</sup>; and results are expressed as milligram of gallic acid equivalent per gram dw (mgGAEgDB<sup>-1</sup>).

### **1.4.4.3.2** Total Flavonoïd content (TFC) :

TFC is quantified colorimetrically. Briefly ; 1.25 ml of deionised water is mixed with 0.25 ml of diluted extract (dilute with propanol) and 0.075 ml of 5 % sodium nitrite solution , and allowed to react for 5 min .Then , 0.15 ml of 10 % aluminium chloride ; and after 6 min, 0.5 ml of 1.0 M sodium hydroxide are added to the mixture. Distilled water 0.775 ml is added to reach a final volume of 3.0 ml. Absorbance was read at 510 nm. A calibration curve is made with standard solutions of catechin with concentration in the rang 0.01-0.50 mg ml<sup>-1</sup>. TFC is expressed in milligrams of catechin equivalent per gram of dry biomass (mgCEgDB<sup>-1</sup>).

#### **1.4.4.3.3** Total carotenoids content (TCC) :

The absorbance of acetone diluted SUPRAS extract was read at 470 nm (A 470), 661.6 nm (A 661.6) and 664.8 nm (A 664.8) on a UV-visible spectrophotometer.

The concentrations of carotenoids were calculated according to the following formulas of (Hartmut, 1987):

Acetone 100 % (pure solvent):

 $C_a = 11.24 \ A_{661.6} - 2.04 \ A_{644.8}$ 

C  $_{b} = 20.13 A_{664.8} - 4.19 A_{661.6}$ 

 $C_{x+c} = 1000 \ A_{470} - 1.90 \ C_a - 63.14 \ C_b \ / \ 214$ 

– C a : Chlorophyll a

– C b : Chlorophyll b

- C x + c: Total of carotenoids (x : xanthophylls and c: carotene).

### 1.5. Physical characterization:

Microstructure of FOWT- SUPRAS and FOWEA- SUPRAS was analyzed by optical microscopy (A. KRUSS-OPTRONIC, GERMANY).

### 1.6. Antioxidant Activity: (Test DPPH)

The antioxidant activity of FOWT-SUPRAS and FOWEA- SUPRAS extracts was determined by DPPH radical scavenging activity (Li, 2008).

In Brief, firstly 1 mg of DPPH was dissolving in 50 ml of methanol then agitated for 30 sec. Then 0.1 mL of the FOWT -SUPRAS extracts and FOWEA-SUPRAS extract was mixed with 1.9 mL of DPPH methanoique solution, vortex all the mixture to homogenized then the mixture was left to react in darkness for 1 hour. Subsequently, the absorbance of the samples was determined at 517 nm utilizing an UV Spectrophotometer (Cary 100 UV-visible spectrophotometry).

Results were calculated as mean  $\pm$  SD by performing all measurements in triplicate (Zi-Hui et al., 2022).

## RESULTS



### DISCUSSION

### 2. **RESULTS AND DISCUSSION:**

### 2.1 Chemical characterization:

### 2.1.1 Ternary phase diagrams of SUPRASs:

The ternary phase diagrams for the mixtures Frying Oil–Water–THF and Frying Oil–Water–Ethyl acetate is depicted in Figure 4 and 5 respectively show that both ternary plots are exhibited From the figure, notice that there is a direct relationship between the volume of the supra and the concentration of the solvent.

The higher ternary plots of the solution, the greater the volume of the supra. We notice that the highest value of the volume of the supra formed when the oil is dissolved in THF solvent is 1099  $\mu$ l in proportion to 70.44 %, compared to the volume of the SUPRAS formed when the oil is dissolved in Ethyl acetate solvent, where the value exceeded 1099  $\mu$ l and rose to 2355  $\mu$ l and this is at a ratio of 81.38 %.

Comparing the results obtained by the results of phase diagrams of octanoic acid- ethanol- water ternary system (Zi-Hui et al., 2022), we can say that they are the same proportional relationship between the SUPRAs formed and the concentration of solvents.

From it, it can be said that THF is a good solvent, as it dissolved all the oil compared to Ethyl acetate solvent, and this was explained in the synthetic condition with the highest % of THF.



Figure 4 : Phase diagram of FOWEA after ternary system



Figure 5 : Phase diagrams of FOWT after ternary system

### 2.1.1 The FTIR spectra :



Figure 6 : The FTIR spectra of Frying oil, FOWEA-SUPRAS and FOWT-SUPRAS

**Figure 6** presents the FTIR spectra of Frying oil, FOWEA-SUPRAS and FOWT-SUPRAS. It is clear that, there were no big different between the three FTIR spectre, generally we can notice that there are seven peak perfectly clear so the **C-H** stretching vibration to **2923** cm<sup>-1</sup> in fraying oil and FOWEA-SUPRAS had the same transmittance **55** % and stretching vibration to **2926** cm<sup>-1</sup> of FOWT-SUPRAS had a short transmittance **69** % due to our less concentration of the THF solvent.

The C=O peak to 1743 cm<sup>-1</sup> in fraying oil and FOWEA–SUPRAS are the same, but in FOWT–SUPRAS are increased which indirectly proved polar groups of THF solvent were restricted by the hydrophilic inner core due to the hydrogen bonding interactions and the THF solvent formed anti-micelles in the mixture.

The stretching vibration to  $1460 \text{ cm}^{-1}$  are also the same in fraying oil and FOWEA-SUPRAS but decreased in FOWT-SUPRAS due to our low concentration.

The stretching vibration to  $1159 \text{ cm}^{-1}$  is the same in fraying oil and FOWEA-SUPRAS but increased in FOWT-SUPRAS C- O peak at  $1163 \text{ cm}^{-1}$ .

There is tow new significant peak in FOWT-SUPRAS stretching vibration to  $1068 \text{ cm}^{-1}$  and stretching vibration to  $908.9 \text{ cm}^{-1}$ .

The stretching vibration to **720.8**  $cm^{-1}$  in fraying oil are increased in both the two SUPRAS and are more in the FOWT–SUPRAS.

Comparing this result with the result of (Zi-Hui Cai et al, 2022) for octanoic acid (OAEW-SUPRAS) the intensity of v-OH and v C=O peak in OAEWSUPRAS decreased and the intensity of v-CH and v C=O peak in our SUPRAS increased, so all the results above improve that FOWT-SUPRAS formed were medium hydrophobic partly oil-in water-in-oil (O/W/O) and mostly water-in-oil (W/O) nano aggregates.

### 2.1.2 Fatty acid composition:

**Table 1** present the major fatty acids content in fraying oil, FOWT-SUPRAS and FOWEA-SUPRAS respectively. After GC/MS analysis, we noticed that they are result four major unsaturated fatty acids (Palmitic acid, Stearic acid, Oleic acid and Linoleic acid).

Carbon Number and Unsaturation	Composition (% total fatty acids in fraying oil)	Composition (% total fatty acids in FOWT - SUPRAS)	Composition (% total fatty acids in FOWEA - SUPRAS)	Remarks
C16:0	11.88	9.95	8.65	Palmitic acid
C18:0	8.2	6.65	5.21	Stearic acid
C18:1	32.01	35.75	36.81	Oleic acid
C18:2	42.1	44.02	47.21	Linoleic acid

 Table 1 : Major fatty acid composition

The linoleic acid C18:2 present the highest content 47.21 % in FOWEA– SUPRAS comparing with our content in fraying oil and FOWT - SUPRAS and the stearic acid present the lowest content with is 5.21 % also in FOWEA- SUPRAS.

The result of C18:1 ranged from 32.01 % to 36.81 %; and the result of C16:0 are near the three compounds between (8.65 %-11.88 %). We notice that the linoleic acid C18:2 as well as C18:1 are the abundant fatty acid; due to the fatty acids of long chain C20, C22 and C24 that the double bonds being broken and this acid losing two, four or six carbon owing to polymerization, cyclization, and oxidation reactions, as well as the incorporation of saturated fats from the fried foods into the frying oil.

### 2.1.3 Results of extraction and dosage of bioactive molecules:

The TPC expressed as (mg GAE/ g DB) of two SUPRAs extract FOWEA-SUPRAS and FOWT-SUPRAS are present in figure 10, we seen that the maximum content of polyphenols are in FOWT SUPRAS 23.86  $\pm$  3.63 mg GAE/g DB, this value was twice as high as before FOWEA- SUPRAS. According to the result of (Suleri et al, 2020) characterization of phenolic compounds and their antioxidant capacity in different fruit peels the TPC are 27.51  $\pm$  0.63 mg GAE/g, we can say that this value obtained 23.86  $\pm$  3.63 mg GAE are slightly low but this means improve that SUPRAS was able to improve the yield for polyphenols obtained with propanol, so the solvent play a vital role in the extraction of the plant constituents. Specially, THF is high polar among the Ethyl acetate. (Figure 7)



Figure 7: TPC of FOWEA-SUPRAS and FOWT-SUPRAS

**Figure 8** present the TFC expressed as (mg CE/g DB) of two SUPRAs we have seen that the best values of flavonoids content were obtained with FOWT-SUPRAs is  $15.99 \pm 1.65$  mg CE/g DB; this value is almost three times of the value obtained by utilization the FOWEA-SUPRAS  $8.73 \pm 0.88$  mg CE/g DB.

According to the result of (Suleri et al,2020) from mango peels, this results obtained  $15.99 \pm 1.65$  mg CE/g DB are highest than the result of mango peels ( $1.75 \pm 0.08$  mg QE/g). These results highlight the importance of fruit peels as a potential source of flavonoids.



Figure 8 : TFC of FOWEA-SUPRAS and FOWT-SUPRAS

Figure 9 present two diagrams of TCC expressed as (mg/g) for two SUPRAs extract (FOWEA-SUPRAs and FOWT-SUPRAS); we notice that the optimal values were obtained with FOWT-SUPRAS that provided a TCC of  $0.23 \pm 0.01$  mg/g. This value was almost two times higher than the one obtained with FOWEA-SUPRAS 0.14  $\pm 0.03$  mg/g SUPRAS.

According to the result of (Jayesree et al, 2021) from carrot waste peel the TCC are (1.17 mg/100g), we can say that the content of carotenoids obtained are low, due to the utilizing a mixture sample of vegetables waste peels in our experiment not only carrot peel wish is rich on carotenoids as well as the content of carotenoids is concentrated in the fruit and vegetables themselves not in its peels.



Figure 9 : TCC of FOWEA-SUPRAS and FOWT-SUPRAS

The different behaviour of polyphenols and carotenoids and flavonoids as a function of SUPRAS composition could be a consequence of both the relative of: the nature of solvents so we founded it in the solvents more polar such as polyphenol and flavonoids, but carotenoids are apolar compound due to our lowest content, and the specific environment where they are expected to be solubilised (Keddar et al., 2020).

### 2.2 Physical characterization:

### 2.2.1 Microstructure observation of SUPRAS :

We observed under the optical microscopy a spherical droplets in both SUPRAs, so the micelle of THF SUPRAS are biggest then the micelle of Ethyl acetate SUPRAS. (Figure 10 and Figure 11)

Comparing with the result of optical microscope image of Octanoic acidethanol-water supramolecular solvents (OAEW-SUPRAS) (Zi-Hui et al., 2022) we show the series of spherical droplets were clearly seen as previously reported (Francisco-Javier et al., 2007).

So the THF solvent is more polair than Ethyl acetate solvent according to the capacity total of dissolving the oil. So it is probable that THF is incorporated at the SUPRAS hydrophobic region while water flows through the pores. This assumption is in good agreement with the nearly constant percentage of water into the SUPRASs (Gonz'alez-Rubio et al., 2022).



Figure 10 : Micrograph of FOWT -SUPRAS using optical microscopy at 10X



Figure 11 : Micrograph of FOWEA -SUPRAS using optical microscopy at 40X

### 2.3 Antioxydant Activity:

The DPPH radical scavenging activities of two SUPRAs extract are presented in **figure 12**. All the compound extracts showed concentration dependent increases in radical scavenging capacity. The greatest DPPH radical scavenging potency value was recorded for FOWT-SUPRAS extract  $75.56 \pm 1.50$  % this value is highest tow time according of the FOWEA-SUPRAS extract  $57.02 \pm 3.60$  %.

Comparing this results obtained with the result of (Sadef et al, 2022) from the different fruits and vegetables peels (15.02 % to 75.95 %),we can say that our SUPRAS had better antioxidant activity ranged from 57.02 % to 75.56 % and both SUPRAS extract had an antioxidant activity depend of the nature of solvents; so the THF solvent had good antioxidant activity then ethyl acetate solvent due to our contain it of bioactive compounds (polyphenols, flavonoids and carotenoids) with are remarkable antioxidant.



Figure 12 : Antioxidant activity of SUPRASs Extract

## CONCLUSION

### Conclusion

We started this research project basing on the following question, How Can we valorize frying oil as an amphiphil in order to form a new SUPRAS capable of recovering and extracting antioxidant bioactive molecules from vegetable kitchen waste, and also to reduce pollution due to the accumulation of food waste? The objective is based on produce under different synthetic conditions new supramolecular solvents; Frying Oil-Water-Ethyl Acetat-SUPRASs and Frying Oil-Water-Tetrahydrofuran-SUPRAS using a new amphiphilic frying oil, in order to save more environmentally friendly and sustainable operations and extraction of antioxidant compounds from vegetable food waste (polyphenols flavonoids and carotenoids).

To reach this project, we firstly prepare FOWEA-SUPRAS and FOWT-SUPRAS, from this results, it can be said that THF is a good solvent, as it dissolved all the oil compared to ethyl acetate solvent. Also, from the FTIR spectra of frying oil, FOWEA-SUPRAS and FOWT-SUPRAS, it is clear that, there were no big differences between the three FTIR spectra mostly are the same; the formation of a large number of hydrogen bonds facilitated the synthesis of FOWT-SUPRAS and FOWEA-SUPRAS. Using GC/AAS analysis we performed the oil Characterization We notice that the linoleic acid C18:2 is the abundant fatty acid; so C18:2 and C18:1 of FOWEA-SUPRAS FOWT-SUPRAS and fraying oil was of a high percentage. As well as we observed under the optical microscopy a spherical droplets in both SUPRAs, so the micelle of FOWT-SUPAS are biggest then the micelle of FOWEA-SUPRAS, Finally, the extraction capacity of different SUPRASs species was examined by extraction of bioactive molecules; the maximum content of polyphenols are in FOWT SUPRAs 23.86 ± 3.63 mg GAE/gDB, The total flavonoids content (mgCE/gDB) of two SUPRAs mark that the best values of flavonoids content were obtained with FOWT-SUPRAs is  $15.99 \pm 1.65$  mgCE/g DB, and for the total of carotenoid content so we saw than the optimal values were obtained with FOWT-SUPRAS that provided a total of carotenoids content is  $0.23 \pm 0.01$  mg/g, and for the greatest DPPH radical scavenging potency value was recorded for FOWT-SUPRAS-Extract is  $75.56 \pm 1.50$  % this value is highest tow time according of the FOWEA-SUPRAS-Extract is  $57,02 \pm 3.60$  %.

Finally, through this research, we have acquired the skills to research and create organic solvents using frying oil while at the same time finding new ways to reduce vegetable residues from cooking.

- Ballesteros-Gómez Ana, María Dolores Sicilia, Soledad Rubio.
   "Supramolecular solvents in the extraction of organic compounds. A review."
   Analytica Chimica Acta 677 (2010): 108-130.
- Dueñas-Mas María Jesús, Ana Ballesteros-G'omez, Soledad Rubio.
   "Supramolecular solvent-based microextraction probe for fast detection of bisphenols by ambient mass spectrometry." Chemosphere 294 (2022): 1-7.
- Francisco-Javier Ruiz, Soledad Rubio, Dolores Pérez-Bendito. "Waterinduced coacervation of alkyl carboxylic acid reverse micelles: phenomenon description and potential for the extraction of organic compounds." Anal. Chem, 2007: 7473–7484.
- Gaithersburg, MD, USA. "International AOAC Guidelines for Single Laboratory Validation of Chemical Methods for Dietary Supplements and Botanicals." Association of Official Analytical Chemists, 2002: 38.
- Glossman-Mitnik D., Maciejewska M. Solvents, Ionic Liquids and Solvent Effects. Eds, 2020.
- Gonz'alez-Rubio S., A. Ballesteros-G'omez, D. García-G'omez, S. Rubio.
   "Double-headed amphiphile-based sponge droplets: synthesis, characterization and potential for the extraction of compounds over a wide polarity range." Talanta 239 (2022): 1-11.
- Hartmut, K. Lichtenthaler. "Chlorophylls and Carotenoids: Pigments of Photosynthetic Biomembranes." METHODS IN ENZYMOLOGY 148 (1987): 350-382.
- Jayesree Nagarajan, Pui Kay Hang, Arumugam Priyangaa, Nagendra Prasad Krishnamurthy, Ramakrishnan Nagasundara Ramanan, M.S. Aldawoud Turki, M. Galanakis Charis, Chien Wei Oo. "Valorisation of carrot peel waste by water-induced hydrocolloidal complexation for extraction of carotene and pectin." Chemosphere, 2021: 1-9.

- Keddar M.N., b, A. Ballesteros-Gómezb, M. Amialia, J.A. Silesc, D. Zerroukid, M.A. Martínc,. "Efficient extraction of hydrophilic and lipophilic antioxidants from microalgae with supramolecular solvents." Separation and purification technology 251 (11 2020): 1-8.
- Laura Sofía Torres-Valenzuela, Ana Ballesteros-Gómez, Soledad Rubio2.
   "Green Solvents for the Extraction of High Added-Value Compounds from Agri-food Waste." Food Eng Rev, 2020: 83–100.
- Li, Y., Jiang, B., Zhang, T., Mu, W., Liu, J. "Antioxidant and free radicalscavenging activities of chickpea protein hydrolysate (CPH)." Food Chem, 2008: 444–450.
- Nusrat Iqbal, Ranju Sharma, Dipak Kumar Hazra, Saurabh Dubey, Natish Kumar. "Successful utilization of waste cooking oil in Neem oil based fungicide formulation as an economic and eco-friendly green solvent for sustainable waste management." Journal of Cleaner Production, 2021: 1-11.
- Sadef Yumna, Javed Tayyaba, Javed Rimsh, Mahmood Adeel, Alwahibi Mona S Elshikh, Mohamed Ragab AbdelGawwa, Jawaher Haji Alhaji,Rabab Ahmed Rasheed. "Nutritional status, antioxidant activity and total phenolic content of different fruits and vegetables peel." PLOS ONE, 2022: 1-9.
- Sánchez-Vallejo Celia, Ana Ballesteros-Gómez, Soledad Rubio. "Tailoring composition and nanostructures in supramolecular solvents: Impact on the extraction efficiency of polyphenols from vegetal biomass." Separation and Purification Technology, 2022.
- Serna-Saldivar .O, Cristina Chuck-Hernandez, Sergio. "Chapter 17 Food Uses of Lime-Cooked Corn With Emphasis in Tortillas and Snacks." Corn (Third Edition) Chemistry and Technology, 2019: 469-500.
- Suleri Hafiz A. R., Colin J. Barrow and Frank R. Dunshea. "Screening and characterization of phenolic compounds and their antioxidant capacity in different fruit peels." Foods (Foods), 2020: 1-26.

- Tan Sook Ling, Syazwan Hanani Meriam Suhaimy, Nur Azimah Abd Samad.
   "Evaluation of fresh palm oil adulteration with recycled cooking oil using GC-MS and ATR-FTIR spectroscopy: A review." Czech Journal of Food Sciences, 2022: 1-14.
- Trupti W. Charpe, Virendra K. Rathod. "Biodiesel production using waste frying oil." Waste Management 31, no. 1 (2011): 85-90.
- Zi-Hui Cai, Jian-Dong Wang, Li-Tao Wang, Su Zhang, Xin-Yu Yan, Yan-Qiu Wang. "Green efficient octanoic acid based supramolecular solvents for extracting active ingredients from Zanthoxylum bungeanum Maxim. peels." Journal of Cleaner Production 331 (2022): 1-57.



## Appendices

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### APPENDIX

Synth condit	etic ions	Tube diameter (mm)	r <sup>2</sup>	SUPRAS phase height (mm)	SUPRAS volume (µL)
Δ	A1	10	25	5	392,5
А	Al	10	25	5	392,5
P	B1	10	25	7	549,5
D	B2	10	25	7	549,5
C	C1	10	25	10	785
C	C2	10	25	10	785
D	D1	10	25	15	1177,5
D	D2	10	25	15	1177,5
Б	E1	10	25	20	1570
E	E2	10	25	20	1570
Б	F1	10	25	22	1727
Г	F2	10	25	22	1727
G	Gl	10	25	26	2041
U	G2	10	25	26	2041
н	H1	10	25	30	2355
11	H2	10	25	30	2355

### Appendix 01: Volume of FOWEA-SUPRAS

### Appendix 02: Volume of FOWT-SUPRAS

Synth condit	etic ions	Tube diameter (mm)	r <sup>2</sup>	SUPRAS phase height (mm)	SUPRAS volume (µL)
Δ	Al	10	25	4	314
	A1	10	25	4	314
в	B1	10	25	5	392,5
	B2	10	25	5	392,5
C	C1	10	25	6	471
	C2	10	25	6	471
D	D1	10	25	7	549,5
	D2	10	25	7	549,5
F	E1	10	25	9	706,5
	E2	10	25	9	706,5
F	F1	10	25	11	863,5
	F2	10	25	11	863,5
G	G1	10	25	14	1099
	G2	10	25	14	1099

### APPENDIX

Appendix 03: The content of frying oil, Ethyl Acetate solvent and water in the	e
FOWEA-SUPRAS	

Synthetic conditions		Weight Ethyl	Weight Water	Weight oil
		Acetat		
		(g)	(g)	(g)
Δ	Al	0,0238	0,0023	0,101
	Al	0,0292	0,0032	0,1019
В	B1	0,0452	0,0046	0,0865
	B2	0,0509	0,0051	0,0817
С	C1	0,067	0,005	0,0586
	C2	0,0659	0,0062	0,057
D	D1	0,0763	0,013	0,0443
	D2	0,0762	0,0108	0,0441
E	E1	0,0893	0,0084	0,0378
	E2	0,0845	0,0117	0,0357
F	F1	0,0963	0,0049	0,033
	F2	0,0974	0,0084	0,0295
G	Gl	0,1015	0,0088	0,0264
	G2	0,101	0,0124	0,0241
и	H1	0,1159	0,0008	0,0237
	H2	0,1107	0,0019	0,0254

### Appendix 04: The content of frying oil, THF solvent and water in the FOWT-SUPRAS

Synthetic conditions		Weight	Wieght	Weight oil
		THF(g)	Water(g)	(g)
Δ.	Al	0,0094	0,0036	0,1061
	A1	0,009	0,0031	0,1003
в	B1	0,0134	0,0046	0,0981
	B2	0,0165	0,0034	0,0886
C	C1	0,038	0,0043	0,0761
C	C2	0,0337	0,0084	0,0626
D	D1	0,0528	0,0041	0,0617
	D2	0,0514	0,0049	0,0577
F	E1	0,0623	0,0032	0,0476
	E2	0,0627	0,0048	0,0488
F	F1	0,0705	0,0027	0,04
Г	F2	0,0736	0,0041	0,0421
G	G1	0,0782	0,0014	0,0331
	G2	0,0808	0,0022	0,03

### APPENDIX

Synthetic conditions		Ethyl Acetate %	Water %	Oil %	
Α	A1	18,725413	1,8095987	79,464988	
	Al	21,742368	2,3827252	75,874907	
В	B1	33,162142	3,3749083	63,462949	
	B2	36,964415	3,7037037	59,331881	
С	C1	51,301685	3,8284839	44,869832	
	C2	51,045701	4,8024787	44,15182	
D	D1	57,110778	9,7305389	33,158683	
	D2	58,12357	8,2379863	33,638444	
E	E1	65,904059	6,199262	27,896679	
	E2	64,063685	8,8703563	27,065959	
F	F1	71,758569	3,6512668	24,590164	
	F2	71,988174	6,2084257	21,8034	
G	G1	74,250183	6,4374543	19,312363	
	G2	73,454545	9,0181818	17,527273	
Н	H1	82,549858	0,5698006	16,880342	
11	H2	80,217391	1,3768116	18,405797	

### Appendix 05: The percentages of frying oil, Ethyl Acetate solvent and water in the FOWEA-SUPRAS:

### Appendix 06: The percentages of frying oil, THF solvent and water in the FOWT-SUPRAS

Synthetic conditions		THF %	Water %	Oil %	
А	Al	7,89252729	3,02267003	89,0848027	
	Al	8,00711744	2,75800712	89,2348754	
В	B1	11,5417743	3,96210164	84,496124	
	B2	15,2073733	3,13364055	81,6589862	
C	C1	32,0945946	3,63175676	64,2736486	
C	C2	32,1872015	8,02292264	59,7898758	
D	D1	44,5193929	3,45699831	52,0236088	
	D2	45,0877193	4,29824561	50,6140351	
E	E1	55,0839965	2,82935455	42,086649	
	E2	53,9122958	4,12725709	41,9604471	
F	F1	62,2791519	2,38515901	35,335689	
	F2	61,4357262	3,42237062	35,1419032	
G	G1	69,3877551	1,24223602	29,3700089	
	G2	71,5044248	1,94690265	26,5486726	

### APPENDIX

### Appendix 07: Synthetic condition of FOWEA-SUPRAS and synthetic condition of FOWT-SUPRAS

Synthetic conditions	Oil (mL)	Water (mL)	THF (mL)	Synthetic conditions	Oil (mL)	Water (mL)	Ethyl Acétat
							(mL)
A	0.3	1.5	0.2	Α	0.3	1.5	0.2
			(12%)				(12%)
В	0.3	1.4	0.3	В	0.3	1.4	0.3
			(18%)				(18%)
С	0.3	1.2	0.5	С	0.3	1.2	0.5
			(30%)				(30%)
D	0.3	1	0.7	D	0.3	1	0.7
			(41%)				(41%)
Е	0.3	0.8	0.9	Е	0.3	0.8	0.9
			(53%)				(53%)
F	0.3	0.6	1.1	F	0.3	0.6	1.1
			(65%)				(65%)
G	0.3	0.4	1.4	G	0.3	0.4	1.4
			(82%)				(82%)