Study and characterization of essential oils from medicinal plants for applications in the food sector

Abstract: The general objective of the present work was to study the applicability of cornstarch containing natural agents as active packaging. We have selected five medicinal plants, such as rosemary, eucalyptus, bay leaf, chamomile, and cloves, to prepare active food packaging. First, the extraction yields were 0.871, 0.868, 0.861, 0.865, 1.001%, and 0.59% of the EOs of *Syzygium aromaticum L., Eucalyptus globulus, Laurus nobilis, Anacyclus pyrethrum, and Rosmarinus officinalis*, respectively, with the crude yields of rosemary and eucalyptus being the highest compared to other essential oils. Their pH values are acidic at 4.5, 5.17, 5.3, 5.1, and 4.89, respectively, values acceptable for the majority of oils with an acidic character. Looking at the antioxidant results, rosemary EO and eucalyptus have the highest capacity to inhibit free radicals. Rosemary and eucalyptus EOs were also remarkable against Escherichia coli and Staphylococcus aureus gilt, recording an inhibition diameter of 22 mm. The results of the chemical composition by CGSM showed that the essential oil extracts of the studied plants were mainly monoterpenic oxygenates, with eucalyptol, camphor, borneol, α-terpineol, linalool, eugenol, and carvacrol being the main compounds, present in large quantities in the essential oils of rosemary and eucalyptus. Currently, science is converging towards the development of essential oil extraction processes in order to limit the spread of diseases.

Key words: Medicinal plants, monoterpenes, antibacterial effect, CGSM.

1-Introduction

Passive packaging has four main functions: food safety, protection, convenience and communication, while active packaging offers active protection, continuously releasing active compounds such as antioxidants, antimicrobial agents, enzymes and flavors [1], [2]. Active packaging's innovative concept and interaction with packaging, food and the environment increase shelf life, preserve flavor, and reduce additives and preservatives in food formulations, while preserving product quality [3], [4]. Most of the active plastic packaging systems encountered in the literature allow the incorporation of synthetic and artificial active substances [4-7]. To ensure that the emerging changes in the food packaging industry strengthen our country'seconomy by improving food safety and quality, minimizing food losses, and protecting the environment from the damage caused by plastic packaging [8].

The aim of this work is the development of new plastic packaging based on the essential oils of ramarin, rosemary, eucalyptus, bay leaf, chamomile, and clove, traditionally known for their therapeutic effects and their antimicrobial, antibacterial, and nutritional properties.

2-Materiels and methods

Eucalyptus, laurel, rosemary, clove and chamomile (pyrethrum) plants were harvested in February in the Boumerdes region, Algeria. The aerial parts of the plants are dried in an airyplace in the shade until their weight is stabilized (about 10 days). The organoleptic parameters of the oils analyzed are in line with AFNOR standards. The EOs was extracted by hydrodistillation using a Clevenger-type apparatus. The following analyses were carried out: chromatography by UV-visible spectroscopy, gas chromatography coupled with CGSM mass spectroscopy. These analyses were carried out at theC.R.D.'s Organic Geochemistry and Environment Department laboratory and at the Chemistry Department teaching laboratory in Algeria. The antioxidant power of the essential oils was tested by the method using DPPH (2,2-Diphenyl Picryl-Hydrazyl) as a relatively stable free radical.

3. Results and discussions

3.1. Analysis of essential oils

3.1.1. Yield of essential oils

The yield values of essential oils of *Syzygium spiceum L., Eucalyptus globulus, Laurus nobilis, Anacyclus pyrethrum,* and *Rosmarinus officinalis* are reported in Table 1:

Table 1. Variation in HE extraction yields

HEs	Yield (%)
Eucalyptus	0.871
Laurel	0.868
Chamomile	0.861

Cloves	0.865
Rosemary	1.001

The recorded results show that the extraction yield peaks after two hours from the start of water boiling at percentages of 0.871, 0.868, 0.861, 0.865, 1.001% and 0.59% of the EOs of *Syzygium aromaticum L, Eucalyptus globulus, Laurus nobilis, Anacyclus pyrethrum and Rosmarinus officinalis*, respectively. Variability in EO content was observed, with rosemary and eucalyptus yielding the highest crude yields compared with the other plants. EO yields can vary from one region to another depending on pedoclimatic factors such as temperature. Le rendement en huile essentielle extraite de clous de girofle par hydrodistillation en laboratoire est de 0,865 %. Ce résultat coïncide presque avec ceux obtenus par d'autres auteurs utilisant la même technique d'hydrodistillation [1], [2]. Other authors have recorded a significantly higher yield using the same hydrodistillation technique [9]. This small difference in yield is probably due to a loss of oil in the aqueous phase of the distillate.

3-1.2. pH study

It's worth noting that pH plays a key role in chemical and biochemical reactions, and can influence an oil's stabilizing properties (antioxidant and antimicrobial effects) [10].

Table 1. Variation in HE extraction pH

EOs	pH Value
Eucalyptus	5.17
Laurel	5.30
Chamomile	5.10
Clove	4.50
Rosemary	4.89

The pH values obtained in our study for oils are acidic at around 4.5, 5.17, 5.30, 5.10 and 4.89 for Syzygium aromaticum L, Eucalyptus globulus, Laurus nobilis, Anacyclus pyrethrum and Rosmarinus officinalis respectively, which are acceptable values for the majority of oils with an acidic character [11].

3.1.3. Study of the organoleptic properties

Organoleptic properties are a means of verifying and controlling oil quality. The organoleptic

parameters of the oils analyzed are in line with AFNOR standards [12]. The results of organoleptic characterization are reported in the table below:

Table 3. Organoleptic characteristics of the essential oils of the plants studied

HE	Color	Odor	Aspect
Eucalyptus	Yellow-Pale yellow	Strong in 1,8-cineole	liquid
Laurel	Colorless or yellow	Cineole, mentholated and camphorated	liquid
Rosemary	Amber to greenish yellow	Rosy	Clear, mobile liquid
Chamomile	Colorless to pale yellow	Fresh and Cineolated	Clear liquid
Clove	Yellow to light yellow	Very spicy	More viscous liquid

The results of the organoleptic analysis show that the essential oils have a liquid appearance. The essential oils obtained have a liquid appearance and almost identical color.

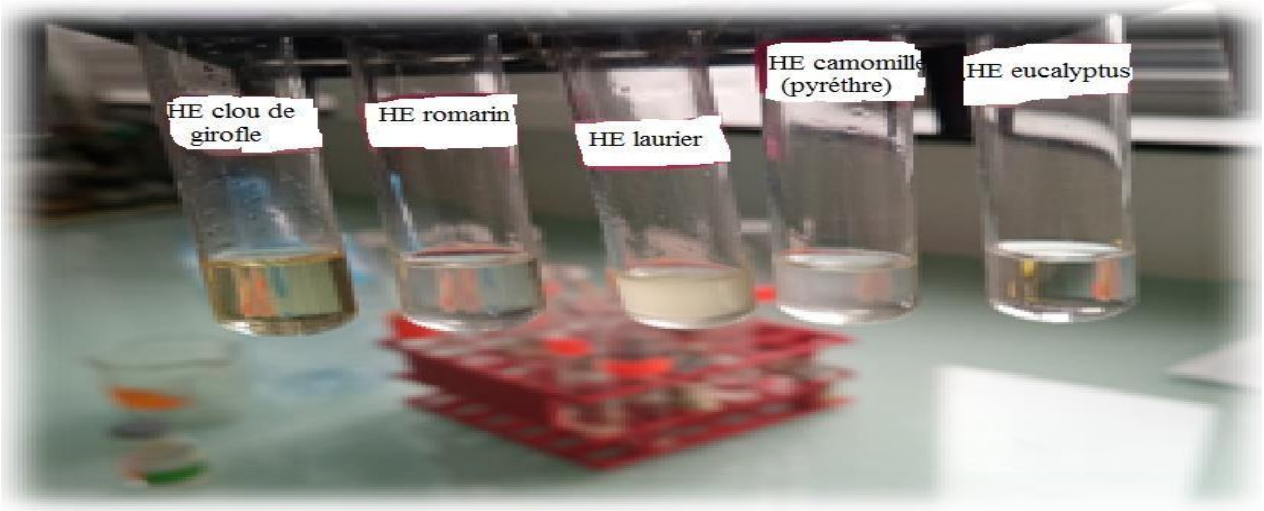


Figure 1: Essential oils extracted from medicinal plants

3.2. Biological activities of essential oils

3.2.1. Antioxidant activity

Comparison of the DPPH radical activity of the extracts obtained shows a concentration-dependent anti-radical activity. The inhibition percentages for rosemary, clove, laurel, eucalyptus, and

chamomile EOs were 86.92, 56.53, 64.45, 82.29, and 57.74%. We find that even at low concentrations, the extract shows a high percentage of inhibition, which allows us to deduce that the bioactive compounds contained in the oil extracts are very effective as antioxidants [13], [14]. The results obtained for the study of the antioxidant power of EOs by the DPPH method are shown in Table 4. These results show that rosemary and eucalyptus EOs have the highest capacity to inhibit free radicals. However, the inhibition capacity of clove, laurel, and chamomile EOs is low.

Table 4. Anti-DPPH potency of essential oil extracts from the five plants at different doses.

Plante / Solution	Rosemary		clove		Laurel		Eucalyptus		chamomile	
	D O optical densite	PI (%) power inhibition	DO optical densite	PI (%) power inhibition	DO optical densite	PI (%) power inhibtion	DO optical densite	PI (%) power inhibition	DO optical densite	PI (%) power inhibition
V_1(HE)=10µl	0792	20.32	0.797	19.93	0.718	27.76	0.512	48.49	0.620	37.625
V_2(HE)=20µl	0,636	36	0.787	20.82	0.701	29.64	0.489	50.80	0.589	40.74
V_3(HE)=30µl	0.230	76.86	0.694	30.18	0.613	38.32	0.410	58.75	0.579	41.75
V_4(HE)=40 µl	0.151	84.80	0.522	47.48	0.505	49.189	0.289	71.35	0.552	44.46
V_5(HE)=50µl	0.130	86.92	0.432	56.53	0.352	64.45	0.179	82.29	0.420	57.74

The IC50s of rosemary and eucalyptus are of the order of 23.31 mg/L and 18.58 mg/L, respectively, so they have greater anti-free radical powerIf we compare the phenolic compound content of the extracts, we see that extracts with a high composition of bioactive molecules are the most active. This finding confirms the hypothesis of [15] , [16] that the high antioxidant power of medicinal plant extracts is often attributed to the presence of active molecules such as phenolic compounds, among other groups of polar compounds [17].

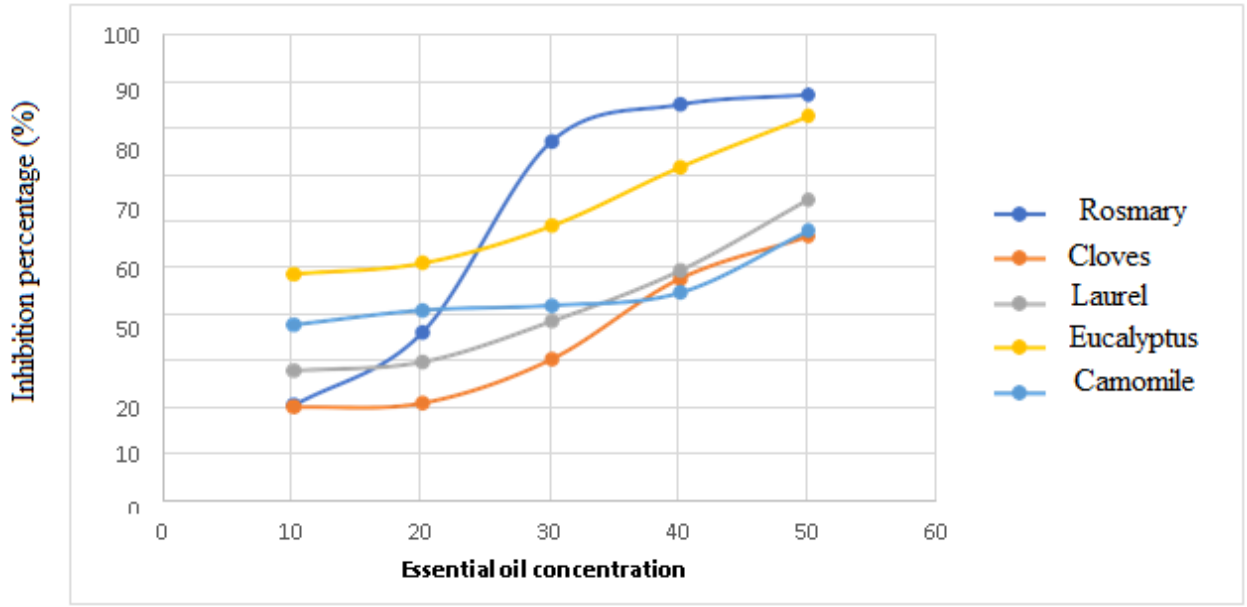


Figure 1. Inhibition percentage graph of Essenseil Oils

3.2.2. Antibacterial activity

The results of the antibiogram test show a variation in the effectiveness of rosemary, bay, chamomile, eucalyptus, and clove essential oils, as well as that of the two antibiotics tested, chloramphenicol and gentamicin, against the bacterial strains tested: Escherichia coli (E.COLI) and Staphylococcus aureus aureus (SASM.). The table below shows that both bacterial strains tested were sensitive to chloramphenicol. Escherichia coli and Klebsiella pneumoniae showed an inhibition diameter of up to 22 mm. The antibiotic gentamicin was equally effective against the other bacterial strains tested. The results of the tests carried out showed that the antibacterial effects of the five essential oils tested varied from one bacterial strain to another and from one oil to another.

Clove essential oil showed extremely sensitive inhibition (22 mm) on *Staphylococcus aureus aureus*, but its action was relatively weak on *Escherichia coli* (10 mm). Laurel essential oil showed sensitive inhibition (16 and 15mm) on *Escherichia coli and staphylococcus aureus*,

respectively. Eucalyptus essential oil showed very sensitive inhibition (22, 22 mm) on all strains on both bacterial strains. Chamomile essential oil showed no antibacterial effect. Rosemary essential oil showed extremely sensitive inhibition for strains of : *Staphylococcus aureus doré and Escherichia.* The effect of rosemary EO was also remarkable against *Escherichia coli* and *Staphylococcus aureus aureus,* recording an inhibition diameter of 22 mm. Studies carried out by [18-22] support our own results..

Table 2: Results of antibacterial activity evaluation of essential oils studied by aromatogram.

Bacterial strains	Diameters (mm)				
	EO laurel	Clove EO	Eucalyptus EO	Chamomile EO	Rosemary EO
Escherichia coli	16 ++	10 +	22 +++	00 -	22 +++
Golden Staphylococcus aureus	15 ++	22 +++	22 +++	00 -	22 +++

Results show that rosemary essential oil, eucalyptus and clove were the most active, with a broad spectrum of action against all the strains tested.

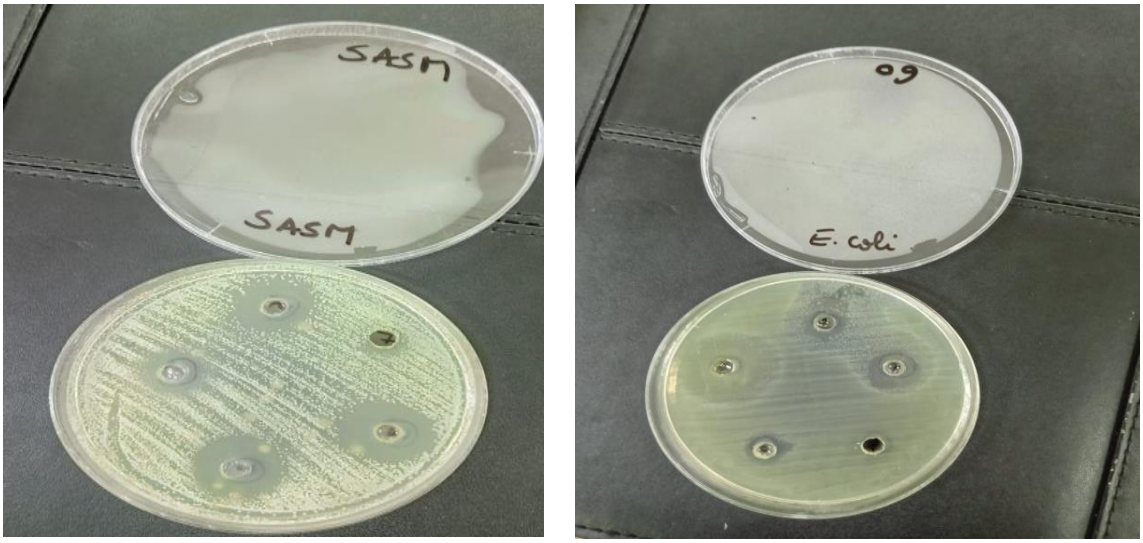


Figure 2: Result of the aromatogram method on: (a) Staphylococcus aureus doré, (b) Escherichia coli

3.3 .Analysis of HEs by gas chromatography-mass spectroscopy (GC-MS)

GC-MS analysis of rosemary, eucalyptus, laurel, eucalyptus, chamomile and clove EOs identified 91.42, 98.89, 61.44, 80.39 and 88.98% monoterpene content in these active agents, respectively, which are the most important compounds in EOs that provide antimicrobial and antibacterial properties. The tables and figures present the structure of the main terpene

compounds present in the EOs studied. The chemical composition of the EOs obtained in this study is summarized in the tables below. The EOs of rosemary, eucalyptus, bay leaf, chamomile, and clove are characterized by a predominance of oxygenated monoterpenes. Eucalyptol, camphor, borneol, α-terpineol, linalool, eugenol, and carvacrol are the main constituents of this fraction. Hydrocarbon monoterpenes dominated by α-pinene, camphene, β-pinene, p-cymene, limonene, and α-phenyllandrene are less present. As for the sesquiterpene fraction characterized by the presence of copaene, α-caryophylene and β-panasinsene in essential oils were the lowest.

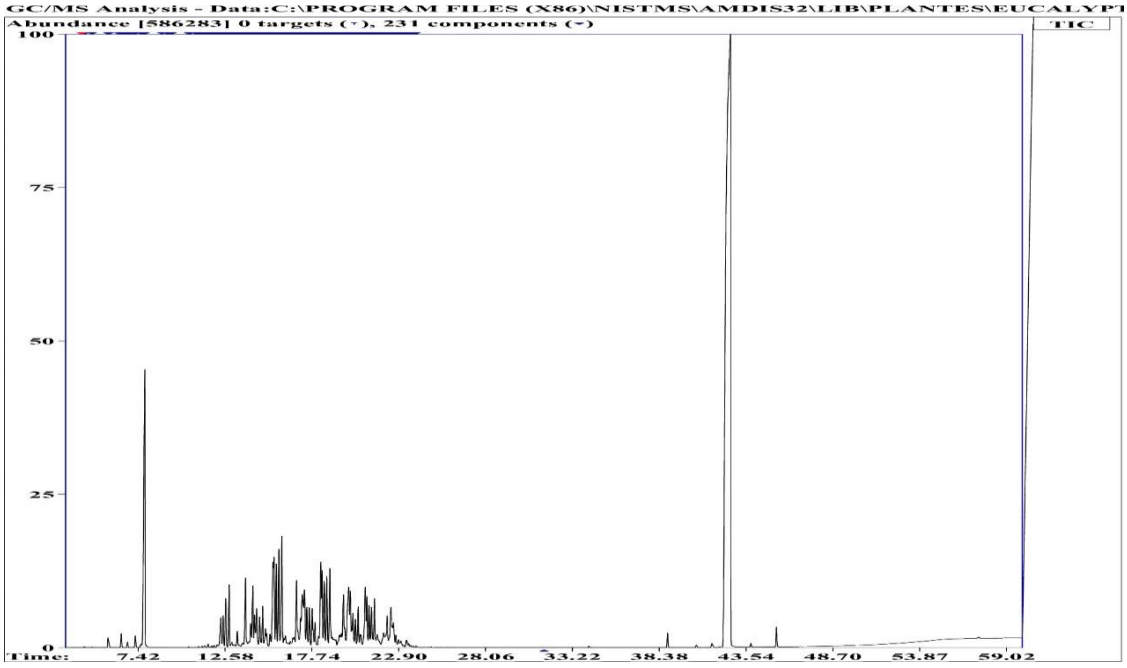


Figure 7: Chromatogram of bioactive compounds in eucalyptus EO

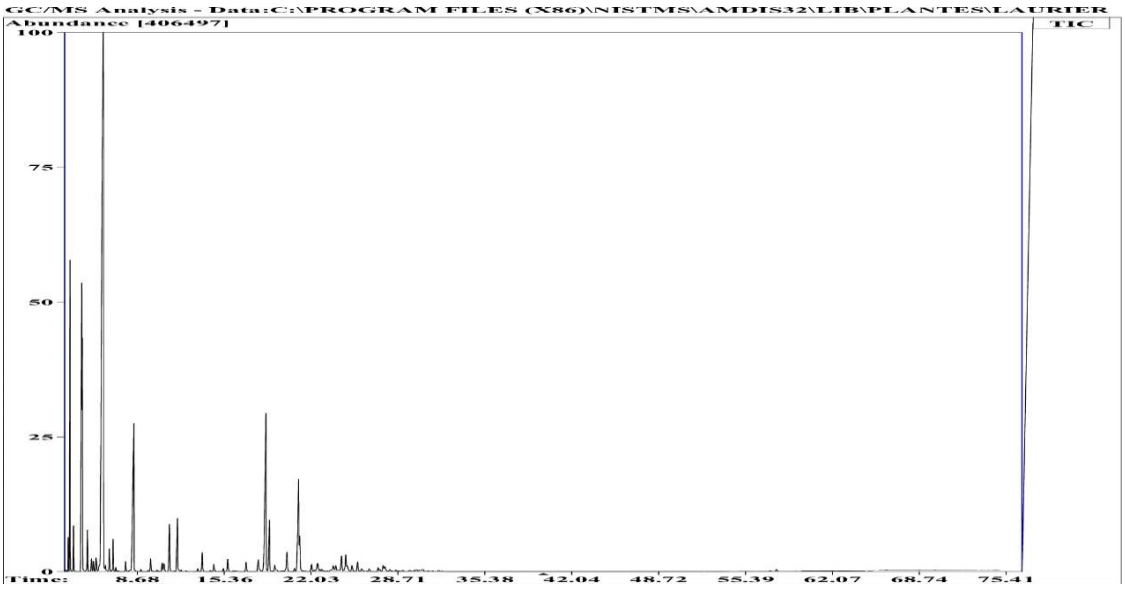
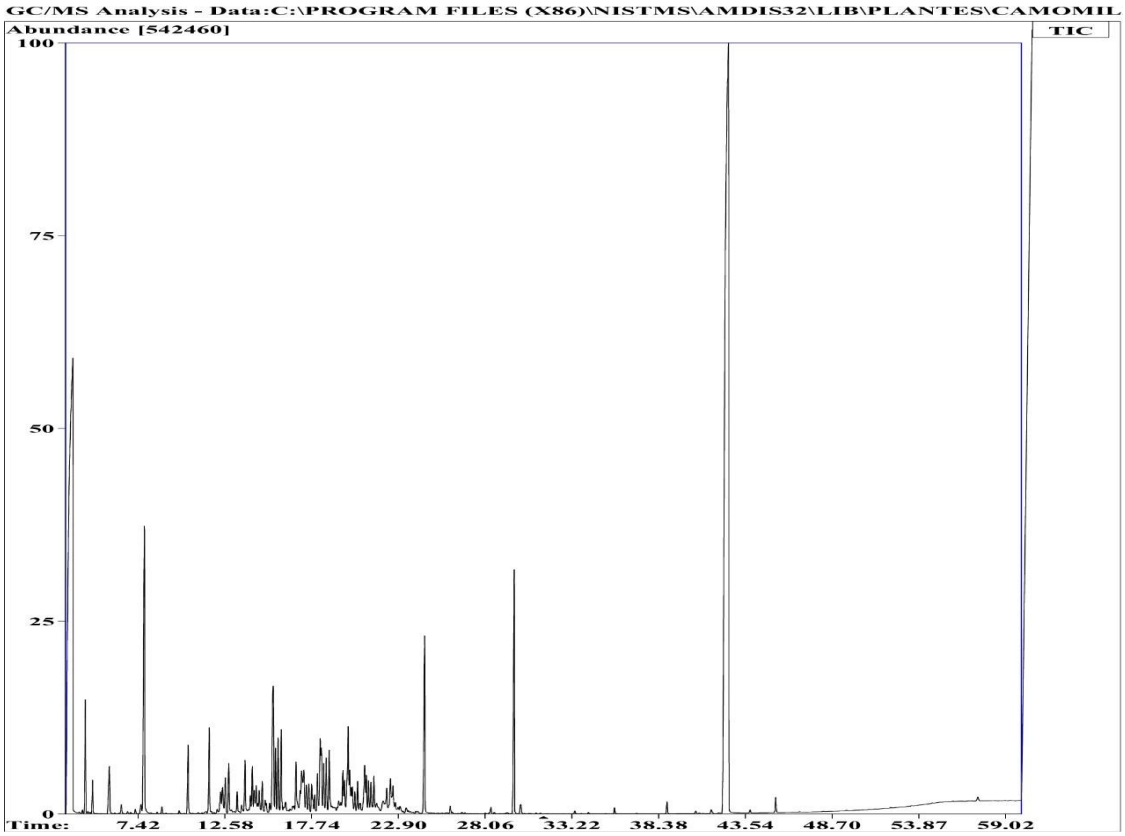


Figure 8: Chromatogram of bioactive compounds present in laurel EO



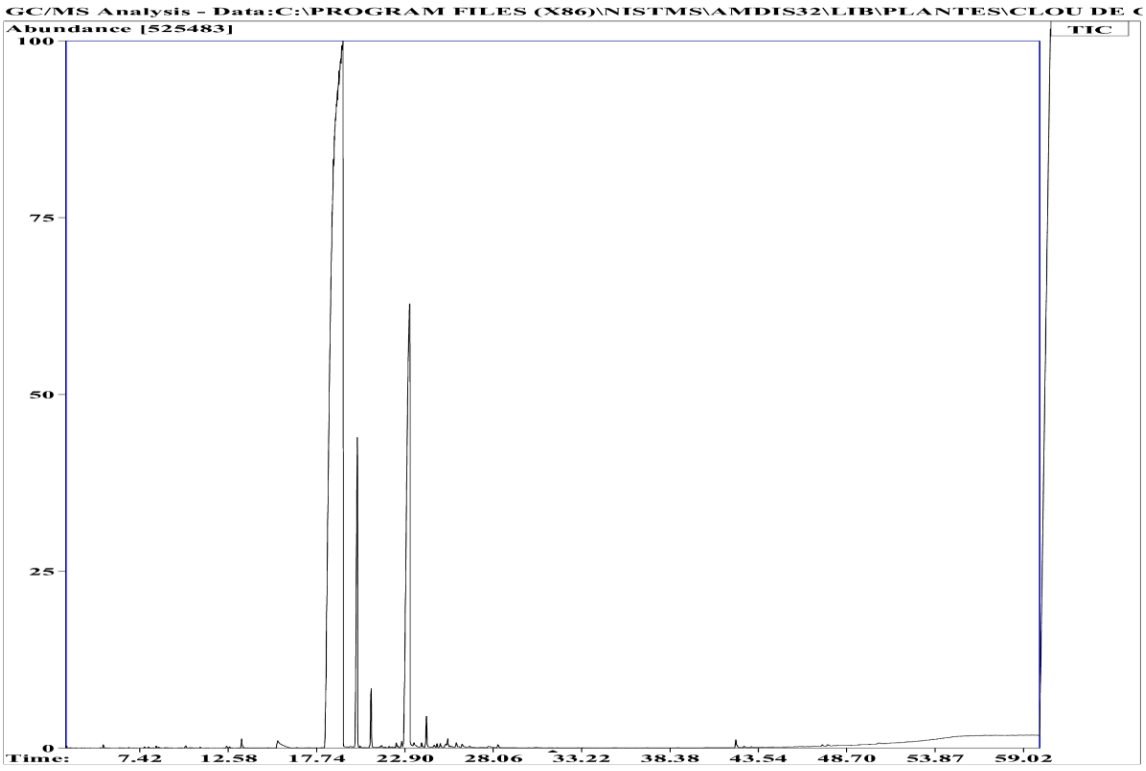


Figure 9: Chromatogram of bioactive compounds in chamomile EO.

Figure 10: Chromatogram of bioactive compounds present in Clove EO

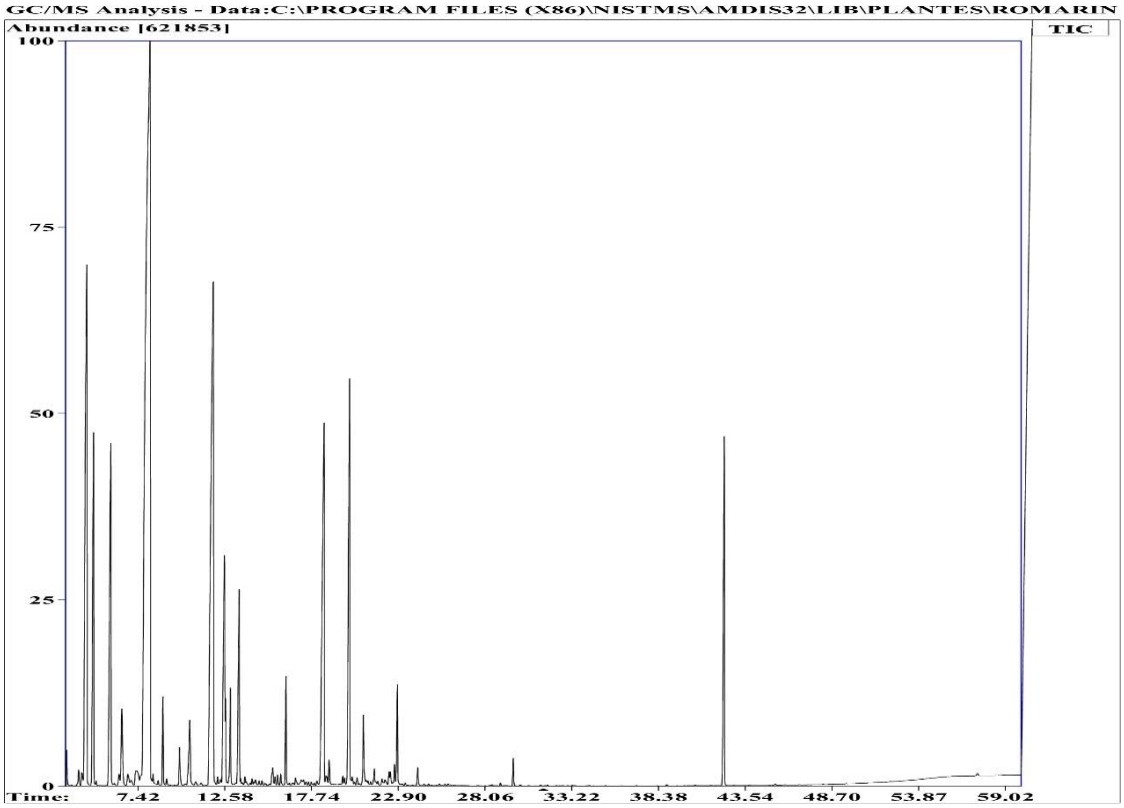


Figure 11: Chromatogram of bioactive compounds present in Rosemary EO.

Comparison with the chemical composition of the EOs studied in our study reveals that the essential oil extracts of the plants studied present a predominance of oxygenated monoterpenes, of which Eucalyptol, Camphor, Borneol, α-terpineol and Linalool, Eugenol and Carvacrol are the main compounds and are found in large quantities in the essential oils of rosemary and eucalyptus, justifying the results of the antioxidant analyses, which are similar to those identified in several works of [23].

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4. Conclusion

The objective of this study is, on the one hand, to enhance the value of essential oils, which are active molecules, with a view to their incorporation from a chemical and biological point of view. On the other hand, to deepen our knowledge on the mode of incorporation of the latter in the starchy matrix, to characterize these systems from a multi-scale point of view, and, finally, to identify the phenomena of quantification of essential oils in the film and their migration in a food simulant in order to predict the kinetics of controlled release. Variability in essential oil content was observed, with rosemary and eucalyptus showing the highest crude yields compared to other plants. pH values are acceptable for the majority of essential oils with acidic character. The results of the organoleptic analysis show that the essential oils have a liquid appearance. The results of the DPPH study of the antioxidant capacity of EOs show that Rosemary EO and Eucalyptus have the highest capacity to inhibit free radicals. However, the inhibitory power of Clove, Laurel and Chamomile EOs is low. The IC50s of rosemary and eucalyptus are of the order of 23.31 mg/L and 18.58 mg/L, respectively, so they have better anti-free radical power. The results of the tests carried out showed that the antibacterial effects of the five essential oils tested varied from one bacterial strain to another. These results show that rosemary, eucalyptus, and clove essential oils were the most active, with a broad spectrum of action against all the strains tested. A comparison with the chemical composition of the EOs studied in our study reveals that the essential oil extracts from our plants show a predominance of oxygenated monoterpenes, including Eucalyptol, Camphor, Borneol, α-terpineol, Linalool, Eugenol and Carvacrol are the main compounds and are found in high quantities in rosemary and eucalyptus essential oils, justifying the results of antioxidant analyses, which are similar to those identified in several studies.

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