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ABSTRACT: This research evaluated blends of six essential oils (EOs) to maximize their antimicrobial effect without compromising sensory acceptability, aiming to inhibit *Pseudomonas aeruginosa* (ATCC 33152), *Salmonella enteritidis* (ATCC 13076), *Escherichia coli* (ATCC 25922), *Staphylococcus aureus* (ATCC 25923), *Klebsiella aerogenes* (ATCC 35029), *Listeria monocytogenes* (ATCC 19114), *Aspergillus brasiliensis* (ATCC 16404), and *Aspergillus niger* (ATCC 6275). Thirty-one mixtures of six essential oils: eucalyptus (*Eucalyptus globulus* Labill.), basil (*Ocimum basilicum* L.), geranium (*Pelargonium graveolens* L'Hér.), rosemary (*Salvia rosmarinus* Spenn.), oregano (*Origanum vulgare* L.), and lemongrass (*Cymbopogon citratus* (DC.) Stapf) were proposed and evaluated by measuring the inhibition halo of microbial growth. Through optimization, four new mixtures were evaluated at 100% concentration and four at 5% concentration. Semi-trained panelists assessed these new blends for the most palatable combination by qualitatively selecting their organoleptic properties. Several blends showed desirability values above 0.90, reaching a maximum of 0.9499 for oregano. However, the blend containing rosemary (0.93%), eucalyptus (1.82%), oregano (94.95%), and lemongrass (2.3%) was selected for having the highest average sensory acceptance in terms of desirability ranking and taste preference. All tested samples inhibited fungal growth. The chosen blend showed total inhibition of *K. aerogenes* and inhibition halos greater than 30 mm for *S. typhimurium*, *E. coli*, *S. aureus*, and *L. monocytogenes*.

Keywords: essential oils; antimicrobial; sensory acceptability; microbial inhibition; optimization; desirability.

Desenvolvimento de filmes biodegradáveis de amido de milho reforçados com nanocelulose para embalagens de alimentos

RESUMO: Esta pesquisa avaliou misturas de seis óleos essenciais (OEs) para maximizar seu efeito antimicrobiano sem comprometer a aceitabilidade sensorial, visando inibir Pseudomonas aeruginosa (ATCC 33152), Salmonella enteritidis (ATCC 13076), Escherichia coli (ATCC 25922), Staphylococcus aureus (ATCC 25923), Klebsiella aerogenes (ATCC 35029), Listeria monocytogenes (ATCC 19114), Aspergillus brasiliensis (ATCC 16404) e Aspergillus niger (ATCC 6275). Trinta e uma misturas de seis óleos essenciais: eucalipto (Eucalyptus globulus Labill.), manjericão (Ocimum basilicum L.), gerânio (Pelargonium graveolens L'Hér.), alecrim (Salvia rosmarinus Spenn.), orégano (Origanum vulgare L.) e capim-limão (Cymbopogon vitratus (DC.) Stapf) foram propostas e avaliadas medindo o halo de inibição do crescimento microbiano. Por meio da otimização, quatro novas misturas foram avaliadas na concentração de 100% e quatro na concentração de 5%. Os painelistas semitreinados avaliaram essas novas misturas para a combinação mais palatável, selecionando qualitativamente suas propriedades organolépticas. Várias misturas apresentaram valores de desejabilidade acima de 0,90, atingindo um máximo de 0,9499 para orégano. Entretanto, a mistura contendo alecrim (0,93%), eucalipto (1,82%), orégano (94,95%) e capim-limão (2,3%) foi selecionada por ter a maior aceitação sensorial média em termos de classificação de desejabilidade e preferência de sabor. Todas as amostras testadas inibiram o crescimento fúngico. A mistura escolhida apresentou inibição total de K. aerogenes e halos de inibição maiores que 30 mm para S. typhimurium, E. coli, S. aureus e L. monocytogenes.

Palavras-chave: óleos essenciais; antimicrobiano; aceitabilidade sensorial; inibição microbiana; otimização; desejabilidade.

1. INTRODUCTION

Food preservation, especially for perishable foods, is a challenge for the food industry, as degradation and contamination by pathogenic microorganisms are significant problems. The Food and Agriculture Organization and the World Health Organization report approximately 200 monthly food safety incidents, often due to inadequate awareness, fraudulent practices, or regulatory weaknesses.

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Consequently, foodborne diseases have increased, with Salmonella spp., Escherichia coli, Campylobacter spp., and Listeria monocytogenes as common pathogens (RODRÍGUEZ; SAMANIEGO-PUERTAS, 2023; RODRÍGUEZ, 2024).

Faced with this problem, essential oils (EOs) have emerged as a natural alternative, as they possess antimicrobial and antioxidant properties capable of inhibiting the growth of bacteria and fungi (WANI et al., 2021; CHONGO, 2025). However, their application in food faces challenges related to the variability of their chemical composition (GUIMARÃES et al., 2019; ALMEIDA et al., 2023; KUMAR et al., 2023), the need for synergistic combinations to improve their effectiveness, and the impact on the organoleptic characteristics of the products. The chemical variability of EOs is influenced by various factors, including the extraction method (mechanical extraction, hydrodistillation, steam distillation, supercritical CO2 (Zhang et al., 2022), solvent distillation, microwave-assisted distillation, and ultrasound (BRAH et al., 2024), as well as their extraction from different plant parts (SHANKAR et al., 2021; SOLÍS-QUISPE et al., 2023). The chemical characteristics of EOs also vary depending on their origin, botanical source, geographic area, season, storage conditions, and (BEAUBRUN et al., 2018; GRANATA et al., 2018).

The antimicrobial and antifungal characteristics of EOs depend on their chemical composition, concentration, interaction between their major components, and the vulnerability of the microorganism (IMAEL et al., 2012). Thus, the level of antimicrobial activity is determined by the majority presence of first aldehydes (cinnamaldehyde and citral) and phenols (carvacrol, eugenol, thymol). By terpene alcohols (linalool, geraniol, menthol, α-terpineol), followed by ketones (β-myrcene, α-thujone) or esters (geranyl acetate) that have a weak antimicrobial activity, and finally, terpene hydrocarbons (α-pinene, β-pinene, limonene, caryophyllene) that are inactive but their presence influences biological responses (INOUYE et al., 2001; DORMAN; DEANS, 2008). Araújo et al. (2019) explained the antioxidant capacity of essential oils (EOs) by the presence of the main component and the synergy between the latter and the other minor components of the EO; that is, all components influence the responses of their properties.

Although adding EOs to foods improves antimicrobial and antioxidant activity, their aroma and flavor could influence the sensory properties of foods, so their inclusion should be controlled. Previous research, such as that conducted by Tsigarida et al. (2000), indicated that including 0.8% (v/m) oregano EO in beef fillets showed acceptable flavor after storage at 5°C and cooking. Mejlholm; Dalgaard (2002) showed that including 0.05% (v/m) oregano EO in cod produced a pleasant flavor that diminished during storage at 2 °C. Ouattara et al. (1997) indicated that 0.9% (v/m) thyme EO on shrimp had no adverse effects on flavor or appearance. However, 1.8% of this same EO on shrimp decreased the crustaceans' acceptability.

Therefore, the present study evaluated blends of six EOs: eucalyptus (Eucalyptus globulus Labill.), basil (Ocimum basilicum L.), geranium (Pelargonium graveolens L'Hér.), rosemary (Salvia rosmarinus Spenn.), oregano (Origanum vulgare L.) and lemongrass (Cymbopogon citratus (DC.) Stapf), which inhibit pathogenic microorganisms responsible for food spoilage, and are also organoleptically accepted. An optimal blend of EOs was determined to maximize their antimicrobial effect without compromising sensory acceptability, thus offering a

sustainable alternative to synthetic preservatives. This blend could have a wide range of practical applications, either as an ingredient in food product formulations or the development of packaging materials, thus contributing to preventing infections and increasing product shelf life.

2. MATERIALS AND METHODS

2.1. Reagents and EOs

The reagents used in this study, including ethanol, methanol, sodium chloride, barium chloride, polysorbate, and ascorbic acid, were of analytical grade (Merck, Germany). The culture media used were potato dextrose agar, blood agar, brain heart infusion agar, and Mueller-Hinton agar, all from Oxoid (Thermo Fisher Scientific Inc., USA).

The essential oils (EOs) used as raw materials were oregano, rosemary, eucalyptus, geranium, and lemongrass, acquired locally (Ecoesencias, Ecuador). According to the supplier, these oils were obtained by hydrodistillation, dried with sodium sulfate, and stored at 4 °C until commercialization.

2.2. Physical and chemical characterization of EOs

The refractive index of each essential oil (EO) was measured at 25 °C using an Abbe refractometer (Thermo Spectronic, USA), following the NTE INEN-ISO 6320 (2013) standard. The relative density was determined according to the NTE INEN 35-1 (2012) standard, which consists of comparing the mass of the EO to the mass of water contained in a pycnometer, both measured at 25 °C.

The volatile compounds of the EOs were analyzed using gas chromatography with a hydrogen flame ionization detector (GC-FID) and mass spectrometry (GC-MS) (QUIJANO-CELIS et al., 2010). A QP-2010 Ultra chromatograph (Shimadzu, Japan) was used, and their blends were analyzed by Fourier-transform infrared spectroscopy (FTIR) to identify the functional groups present. The readings were performed using a Jasco FT/IR 4200 spectrometer in ATR mode, within a spectral range of 400 to 4000 cm⁻¹.

2.3. Preparation of EO blends

The essential oils (EOs) and the proposed blends through the mixture design of the Minitab statistical software version 18 (Minitab LLC, USA) are presented in Table 1. The study defined 31 experimental runs, each with a specific composition of six essential oils. Each blend was formulated based on the inclusion percentages of the EOs, ensuring that the total sum in each run was 100%. The experimental design included combinations ranging from binary mixtures to more complex formulations with all oils in similar proportions. In particular, runs 1, 6, 10, 20, 21, and 23 evaluated the effect of each EO in isolation at 100%. In contrast, other runs combined two or more oils in different proportions to identify synergies that would enhance the antimicrobial activity. Additionally, runs 9, 26, and 29 were replicates of the design to validate the blend at different stages of the study.

For the design's numerical optimization, each EO's influence was determined based on its antimicrobial capacity, and the desirability function was applied to estimate the optimal blend. The process identified a formulation that maximized the desired effect. The antimicrobial activity of preselected mixtures was verified using the experimental design model. For this, strains of Gram-positive and Gramnegative bacteria were used.

Table 1. Experimental runs for essential oils based on a mixture design.

Tabela 1. Execuções experimentais para óleos essenciais baseadas

em um design de misturas.

	design de		LEO	OFO	DEC	OFO
Run	REO	EEO	LEO	OEO	BEO	GEO
1	0.00	0.00	0.00	100.00	0.00	0.00
2	50.00	0.00	0.00	0.00	0.00	50.00
3	0.00	50.00	0.00	0.00	0.00	50.00
4	0.00	50.00	50.00	0.00	0.00	0.00
5	50.00	0.00	0.00	0.00	0.00	50.00
6	0.00	0.00	0.00	0.00	100.00	0.00
7	0.00	0.00	0.00	50.00	0.00	50.00
8	50.00	0.00	0.00	0.00	50.00	0.00
9	8.33	18.33	18.33	18.33	18.33	18.33
10	100.00	0.00	0.00	0.00	0.00	0.00
11	0.00	50.00	0.00	0.00	50.00	0.00
12	0.00	0.00	50.00	0.00	50.00	0.00
13	0.00	0.00	50.00	0.00	0.00	50.00
14	0.00	0.00	33.33	33.33	33.33	0.00
15	0.00	50.00	0.00	50.00	0.00	0.00
16	50.00	0.00	0.00	50.00	0.00	0.00
17	0.00	0.00	0.00	50.00	50.00	0.00
18	0.00	0.00	0.00	33.33	33.33	33.33
19	50.00	0.00	0.00	50.00	0.00	0.00
20	0.00	100.00	0.00	0.00	0.00	0.00
21	0.00	0.00	0.00	0.00	0.00	100.00
22	50.00	50.00	0.00	0.00	0.00	0.00
23	0.00	0.00	100.00	0.00	0.00	0.00
24	50.00	0.00	0.00	0.00	50.00	0.00
25	50.00	0.00	50.00	0.00	0.00	0.00
26	8.33	18.33	18.33	18.33	18.33	18.33
27	0.00	0.00	33.33	0.00	33.33	33.33
28	0.00	0.00	33.33	33.33	0.00	33.33
29	8.33	18.33	18.33	18.33	18.33	18.33
30	0.00	0.00	50.00	50.00	0.00	0.00
31	0.00	0.00	0.00	0.00	50.00	50.00
DEC	т.	EO 1	· IEO I		OEO	DEO

REO, rosemary; EEO, eucalyptus; LEO, lemongrass; OEO, oregano; BEO, basil; GEO, geranium.

2.4. Determination of antimicrobial activity

The antimicrobial activity of each experimental unit was assessed using the Zone Inhibition Method (Kirby-Bauer method). Sterilized Muller-Hinton agar was solidified in 8 cm Petri dishes. Microorganism strains, prepared according to the McFarland scale, were spread over the solidified agar with the help of a sterile swab. Ten microliters of the essential oil mixture were placed onto a sterilized filter paper disc (ALBET LabScience, S.L., Spain) positioned on the contaminated agar. The inhibition zone was measured following the Rajendra et al. (2004) method, or the area surrounding the disc impregnated with essential oils, free from microbial growth. The analysis was performed in triplicate.

Specific culture conditions were used for each one to evaluate the microorganisms. *Pseudomonas aeruginosa* (ATCC 33152), *Salmonella enteritidis* (ATCC 13076), and *E. coli* (ATCC 25922) were cultured on blood agar and incubated at 37 °C for 24 hours. *Staphylococcus aureus* (ATCC 25923) and *Klebsiella aerogenes* (ATCC 35029) were cultured on Brain Heart Infusion (BHI) agar and incubated at 37 °C for 24 hours. *L. monocytogenes* (ATCC 19114) was also cultured on BHI agar, but its incubation time was extended to 48 hours at 37 °C due to its slower growth requirements. *Aspergillus brasiliensis* (ATCC 16404) and *Aspergillus niger* (ATCC 6275) were

cultured on potato dextrose agar and incubated at 28 °C for 48 hours to promote their proper growth.

2.5. Sensory evaluation of preselected essential oil blends

This evaluation was approached from a qualitative perspective. The selected essential oil blends were evaluated through sensory analysis. This analysis aims to detect the preference and taste order of the samples prepared with different essential oil concentrations. Five samples with varying compositions of EO blends were prepared according to the guidelines in the optimization trial. To effectively assess these EOs, their essence was masked by consuming papaya (Carica papaya L.) slices coated with the different EO blends. The coating was made by preparing an emulsion with gelatin, glycerol, and each preselected EO blend. This blend was homogenized at 5200 rpm to ensure a single phase. Minimally processed papaya pieces were coated following the methodology of Fon-Fay et al. (2018). The emulsion was left to dry, and a qualitative evaluation was conducted using the expert judgment method. A panel of semi-trained tasters assessed the papaya pieces based on taste and preference.

The panel performed separate evaluations, with a 30-minute interval between samples to ensure that the taste lingering on the taste buds would dissipate between each assessment. These measurements were repeated three times, with two-day intervals between each session. The five-point scale used for this evaluation was as follows: very little perceived (1), little perceived (2), moderately perceived (3), well perceived (4), and excellent perceived (5).

For statistical analysis of the sensory evaluation results, the appropriateness of a simple ANOVA was determined, and the necessary assumptions were verified, including independence of observations, data normality, and homogeneity of variance. Since the assumptions of normality and homogeneity were not met, the Kruskal-Wallis test was used as a nonparametric alternative. After obtaining a significant result in the Kruskal-Wallis test, post-hoc tests using Tukey's HSD test were performed to identify differences between the EO blends.

3. RESULTS

3.1. Characterization of EOs

The EOs exhibited different physical and chemical properties. Table 2 presents the refractive index and density. The GC-MS analysis revealed major, minor, and trace components. In the rosemary EO, the predominant components were myrcene (27%), 1,8-cineole (15.1%), and camphor (17.4%). In the case of basil EO, the main components were methyl chavicol (74.8%) and linalool (17.6%). On the other hand, the geranium EO was characterized by its content of linalool (11%), β-citronellol (30.3%), and geraniol (25.6%), along with other compounds in smaller proportions. The oregano EO analyzed contained mainly carvacrol (51.7%), β-citronellol (6.7%), carvone (5.0%), and geraniol (5.0%). The eucalyptus EO showed a high concentration of 1,8-cineole (58.8%), α-pinene (16.7%), and myrcene (3.3%). Finally, the lemongrass EO exhibited a composition that included geranial (32.7%), neral (27.7%), geraniol (17.3%), and β-citronellol (6.8%).

The FTIR analysis (Figure 1) of the EOs highlights the diversity of their chemical components, which influence their antimicrobial and antioxidant properties (GAVILANEZ, ROJAS, 2024). At 2959 cm⁻¹ in rosemary oil, a band attributed to the hydroxyl (-OH) group of borneol is

observed, according to various authors (OLOPADE, 2019; SATTARY et al., 2020; HOSSEINI et al., 2021), along with the stretching of aliphatic CH, CH₃, and CH₂ bands (KUMAR et al., 2012; PARTHENIADIS et al., 2017; MOKHTAR et al., 2023), suggesting a possible contribution to its antioxidant activity. The presence of ketones at 1745 cm⁻¹ confirms the camphor content, a key compound in its chemical profile (STRAMARKOU et al., 2020; HOSSEINI et al., 2021). Additionally, bands at 1375 cm⁻¹ corresponding to OH bending, at 1244 cm⁻¹ to C-O stretching, and C-O-C stretching (PARTHENIADIS et al., 2017) were detected, which are associated with terpenoids and flavonoids.

Table 2. Refractive index and density of essential oils Tabela 2. Índice de refração e densidade dos óleos essenciais

Essential oil	Refractive index*	Density (g mL-1)
Salvia rosmarinus	1.4720	0.8750
Eucalyptus globulus	1.5530	0.8990
Cymbopogon citratus	1.4970	0.9352
Origanum vulgare	1.5120	0.9557
Ocimum basilicum	1.5185	1.0094
Pelargonium graveolens	1.4645	0.8476

*At 25 °C.

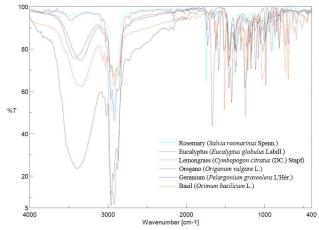


Figure 1. IR spectrum of essential oils. Figura 1. Espectro IR dos óleos essenciais.

In the eucalyptus EO, bands were observed at 1214 and 986 cm⁻¹ (BARANSKA, 2005; STRAMARKOU et al., 2020), related to the ether function of the epoxy ring in 1,8-cineole, the main compound with expectorant and antimicrobial properties. The lemongrass EO exhibited bands in the 1669 cm⁻¹ region, attributed to aldehydes and ketones, characteristic of citral, a key compound in its aromatic profile and antimicrobial activity. In the basil EO, bands were found at 1375-1383 cm⁻¹, associated with phenols such as eugenol, responsible for its antifungal action.

In the oregano and lemongrass EOs, a strong band was identified at 3393-3426 cm⁻¹ due to the stretching of the -OH group (OLOPADE, 2019; SATTARY et al., 2020; HOSSEINI et al., 2021). The oregano EO showed intense bands at 1580-1430 cm⁻¹, associated with the presence of aromatic rings (C=C), according to Hosseini et al. (2021). This indicates a high concentration of phenols such as carvacrol and thymol, responsible for its potent antimicrobial effect.

In the basil EO, peaks were found at 1375-1383 cm⁻¹, associated with phenols such as eugenol, responsible for its antifungal action. Finally, the geranium EO exhibited a

strong signal at 1361 cm⁻¹, related to bending bands of the -OH group, highlighting its composition rich in geraniol and citronellol, known for their antibacterial and insect-repellent activity.

3.2. Antimicrobial capacity of the EO blends

Table 3 presents the inhibition halos generated by different EO blends on the growth of various microorganisms. Overall, the blends with oregano EO showed the most significant inhibition halos. This is evident in the results for *P. aeruginosa*, *L. monocytogenes*, *K. aerogenes*, and *S. enteritidis*, where the highest inhibition halo values exceeded 35 mm, indicating significant bacterial growth inhibition. *P. aeruginosa* and *S. enteritidis* showed potent inhibition with some blends, reaching up to 39.4 mm halos. This is notable and suggests that oregano is particularly effective against these bacteria. On the other hand, *S. aureus* seems more resistant to the EO blends, with generally lower values than other microorganisms.

Table 3. Inhibition halo (mm) of microbial growth induced by essential oil blends

Tabela 3. Halo de inibição (mm) do crescimento microbiano induzido pelas misturas de óleos essenciais

induzido pelas misturas de óleos essenciais							
Run	Α	В	С	D	Е	F	
1	35.20	35.30	33.40	31.36	33.40	35.00	
2	8.53	15.26	16.60	7.35	8.50	16.47	
3	25.48	13.10	11.30	8.00	35.20	23.70	
4	29.40	21.20	18.80	9.00	35.40	15.60	
5	8.52	15.18	16.13	7.31	7.30	16.10	
6	14.23	16.90	13.40	8.63	33.40	13.30	
7	31.21	36.69	22.20	9.91	10.80	33.70	
8	7.50	17.80	10.50	7.70	12.23	10.03	
9	19.78	21.82	16.80	7.06	18.90	19.10	
10	14.40	21.70	21.29	6.33	21.00	8.70	
11	4.70	14.60	15.20	11.10	22.83	12.80	
12	18.17	17.30	15.20	10.29	15.47	10.70	
13	18.28	12.00	9.70	7.90	19.20	14.86	
14	25.00	25.64	25.48	10.27	13.12	19.50	
15	24.50	34.40	21.80	6.50	35.70	27.60	
16	29.76	27.30	28.50	10.43	11.80	22.50	
17	29.75	31.03	32.53	11.27	3.86	24.50	
18	24.40	31.42	23.90	7.02	2.50	25.40	
19	29.53	27.30	28.30	10.70	11.00	22.30	
20	16.41	16.50	13.86	9.70	12.30	17.10	
21	17.28	21.90	24.30	13.46	33.30	27.40	
22	6.00	15.50	11.24	9.53	25.20	11.10	
23	39.40	23.50	21.50	8.54	29.50	8.80	
24	7.90	17.50	10.90	8.00	11.80	9.65	
25	33.60	19.20	15.54	12.01	29.30	8.30	
26	19.73	21.90	16.53	7.13	17.70	19.20	
27	12.79	15.90	11.90	8.11	7.90	14.30	
28	25.30	25.20	17.15	8.12	15.70	24.15	
29	19.83	21.94	16.81	7.06	15.40	19.50	
30	31.20	28.92	25.94	13.26	35.00	22.50	
31	10.02	22.20	16.09	7.40	10.80	18.20	
A D	7	· / \ / T	CC 221F0	D I		ATCC	

A, Pseudomonas aeruginosa (ATCC 33152); B, Listeria monocytogenes (ATCC 19114); C, Escherichia coli (ATCC 25922); D, Staphylococcus aureus (ATCC 25923); E, Klebsiella aerogenes (ATCC 35029); F, Salmonella enteritidis (ATCC 13076).

The combinations of EOs produced different results in terms of microbial inhibition. This variability suggests that the synergy between the essential oils or their concentrations may influence antimicrobial effectiveness, indicating that not all EOs work the same way when combined. Therefore, selecting EOs and their combinations is crucial for optimizing their antimicrobial action. These results could help develop new antimicrobial products based on EOs, such as natural preservatives for the food industry or cosmetic products.

The EO blends showed inhibition halos greater than 32 mm against A. brasiliensis and A. niger, indicating their potential as antifungal agents. The ANOVA revealed significant differences in the size of inhibition halos for the various bacteria (Table 4). It presented the results of various statistical models, including regression, linear, and quadratic analyses, and interactions between different EOs. For the majority of the microorganisms (P. aeruginosa, L. monocytogenes, E. coli, S. aurens, K. aerogenes, and S. enteritidis), the p-values for the regression, linear, and quadratic models were 0.000, indicating a significant effect of the essential oils on microbial growth. This suggests that their concentrations and combinations highly influence the EOs' antimicrobial properties.

In the case of interaction terms, such as REO*LEO (rosemary and lemongrass) or REO*OEO (rosemary and oregano), most interactions showed significant p-values (below 0.05) for certain microorganisms, suggesting that the combination of specific oils enhances or modifies the antimicrobial effect. For instance, the interaction between rosemary and oregano (REO*OEO) significantly affected S.

aureus and S. enteritidis. In contrast, combinations like EEO*LEO (eucalyptus and lemongrass) and EEO*OEO (eucalyptus and oregano) were significant for some microorganisms, while others, such as EEO*BEO (eucalyptus and basil), showed less significance.

These results suggest that the effectiveness of EOs in inhibiting microbial growth depends not only on the individual oil but also on the specific combinations and interactions between them. Some interactions, like REO*LEO, seem to have more significant antimicrobial effects than others, and carefully selecting EO combinations is crucial for optimizing antimicrobial properties.

Table 5 presents the coefficients of determination (R^2) of the inhibition models as a function of the microorganism, including adjusted R^2 , the predictive residual sum of squares (PRESS), and the predictive R^2 coefficient. These indicators allow for assessing the accuracy and predictive capability of the models used in the study.

Overall, the models exhibit a high explanatory capacity, with R² and adjusted R² values exceeding 98.73% for all evaluated microorganisms. *L. monocytogenes* stands out with the highest coefficients (R²=99.98% and adjusted R²=99.94%), indicating an excellent fit of the model to the experimental data. Other microorganisms, such as *E. coli* and *S. enteritidis*, also show high determination coefficients, reflecting accurate behavior modeling.

Table 4. Data and response variables.

Tabela 4. Dados e variáveis de resposta

Source	A	В	C	D	E	F
Regression	0.000	0.000	0.000	0.000	0.000	0.000
Linear	0.000	0.000	0.000	0.000	0.000	0.000
Quadratic	0.000	0.000	0.000	0.000	0.000	0.000
REO*EEO	0.000	0.000	0.000	0.000	0.000	0.000
REO*LEO	0.000	0.000	0.000	0.000	0.015	0.258
REO*OEO	0.000	0.000	0.001	0.000	0.000	0.043
REO*BEO	0.000	0.000	0.000	0.028	0.000	0.002
REO*GEO	0.000	0.000	0.000	0.000	0.000	0.000
EEO*LEO	0.065	0.000	0.002	0.925	0.000	0.000
EEO*OEO	0.197	0.000	0.000	0.000	0.000	0.000
EEO*BEO	0.000	0.000	0.000	0.000	0.721	0.000
EEO*GEO	0.000	0.000	0.000	0.000	0.000	0.001
LEO*OEO	0.000	0.002	0.000	0.000	0.045	0.019
LEO*BEO	0.000	0.000	0.000	0.000	0.000	0.509
LEO*GEO	0.000	0.000	0.000	0.000	0.000	0.000
OEO*BEO	0.000	0.000	0.000	0.000	0.000	0.233
OEO*GEO	0.000	0.000	0.000	0.000	0.000	0.000
BEO*GEO	0.000	0.000	0.000	0.000	0.000	0.000

A, Pseudomonas aeruginosa (ATCC 33152); B, Listeria monocytogenes (ATCC 19114); C, Escherichia coli (ATCC 25922); D, Staphylococcus aureus (ATCC 25923); E, Klebsiella aerogenes (ATCC 35029); F, Salmonella enteritidis (ATCC 13076). REO, rosemary; EEO, eucalyptus; LEO, lemongrass; OEO, oregano; BEO, basil; GEO, geranium.

Table 5. Coefficients of determinação dos modelos de inibição em função do microrganism Tabela 5. Coeficientes de determinação dos modelos de inibição em função do microrganismo

Bacterium	R ² (%)	R ² (Adjusted) (%)	PRESS	R2 (Predictive) (%)
P. aeruginosa	99.85	99.55	451.07	83.68
L. monocytogenes	99.98	99.94	23.86	98.27
E. coli	99.96	99.88	31.99	97.49
S. aureus	99.94	99.83	75.62	87.29
K. aerogenes	99.58	98.73	1090.3	66.91
S. enteriditis	99.95	99.86	29.52	98.07

PRESS, predictive residual sum of squares.

Regarding predictive capability, assessed through predictive R², L. monocytogenes, E. coli, and S. enteritidis have the highest values (>97%), suggesting that the models can reliably predict new data for these microorganisms. In contrast, K. aerogenes has the lowest predictive R² value (66.91%), indicating a reduced ability of the model to predict data outside the sample used for fitting.

The PRESS supports these findings. L. monocytogenes has the lowest PRESS value (23.86), suggesting the model achieves high precision in predicting its data. On the other hand, K. aerogenes shows the highest PRESS value (1090.3), which explains its lower observed predictive capacity.

The inhibition models demonstrate an adequate fit, with R² and adjusted R² values close to 100% for most microorganisms. However, predictive capability varies, being more reliable for *L. monocytogenes*, *E. coli*, and *S. enteritidis* while less effective for *K. aerogenes*. This suggests the need to optimize the model or include additional variables to improve accuracy in some instances.

3.3 Optimizing essential oil blends

The results in Tables 4 and 5 are prerequisites for the mathematical optimization of blends with antimicrobial activity according to their desirability (Table 6). Four possible blends, identified as B1, B2, B3, and B4, were prepared as indicated in Table 6. Sensory analysis also tested these to determine the most widely accepted mixture.

Table 6. Numerical optimization solutions for essential oil blends. Tabela 6. Soluções de otimização numérica para misturas de óleos essenciais.

Blend	REO (%)	EEO (%)	LEO (%)	OEO (%)	BEO (%)	GEO (%)	Desirability
B1	0.93	1.82	2.30	94.95	0.0	0.0	0.94
B2	0.0	0.0	0.0	100.0	0.0	0.0	0.95
В3	1.01	1.01	3.03	93.61	0.0	1.34	0.93
B4	3.15	4.03	7.07	85.75	0.0	0.0	0.90

REO, rosemary; EEO, eucalyptus; LEO, lemongrass; OEO, oregano; BEO, basil; GEO, geranium.

According to the data in Table 6, the most frequently used essential oils are oregano, lemongrass, eucalyptus, and rosemary. Furthermore, oregano EO stands out for its high concentration. This oregano EO displays notable antimicrobial and antioxidant activity even without other compounds. However, given consumer appeal, cost reduction, and availability considerations, there is a need to explore complementarity with other EOs that may offer additional benefits.

3.4 Antimicrobial activity of preselected blends

The antimicrobial activity of EO blends, preselected by numerical optimization, and their 5% dilutions were evaluated against Gram-positive and Gram-negative bacteria (Table 7). The 5% dilutions represent the minimum concentration that should be added to achieve inhibition. This value of 50,000 μg mL⁻¹ is similar to the value presented by Argote-Vega et al. (2017), who indicate that for rosemary and *S. aureus*, the minimum inhibitory concentration was 28.48 mg mL⁻¹, while wild oregano required 37.2 mg mL⁻¹.

The EO blends tested showed adequate antimicrobial and antifungal responses. Blend B1 had the best response against *E. coli* and *K. aerogenes*, while at 5% concentration, its response was better against each bacterium tested. Blend B4,

both at 100 and 5% concentrations, had the smallest inhibition zones.

Table 7. Diameter of inhibition halos (mm) of bacteria exposed to blends of essential oils and their 5% dilutions

Tabela 7. Diâmetro dos halos de inibição (mm) de bactérias expostas a misturas de óleos essenciais e suas diluições de 5%

Blend	В	С	D	Е	F
B1	38.0 (5.3)	41.0 (3.6)	31.0 (1.0)	Total	30.7 (1.2)
B2	41.3 (3.1)	36.3 (3.2)	38.3 (3.2)	40.0 (7.2)	36.3 (1.5)
В3	40.7 (3.1)	31.7 (2.9)	34.7 (6.4)	34.7 (2.3)	40.3 (0.6)
B4	33.3 (3.1)	31.0 (3.6)	30.7 (1.2)	36.7 (6.1)	30.0 (2.0)
B1/5	20.7 (2.5)	20.0 (2.0)	16.0 (1.0)	23.8 (2.3)	19.0 (2.7)
B2/5	14.5 (1.0)	19.7 (2.1)	11.3 (0.6)	22.3 (0.8)	17.0 (1.0)
B3/5	15.7 (1.3)	20.3 (0.6)	13.3 (1.2)	21.5 (3.5)	15.7 (1.5)
B4/5	16.0 (2.2)	16.3 (4.7)	12.0 (1.0)	19.0 (1.8)	16.7 (2.1)

B1/5 ... B4/5, corresponds to blends B1 ... B4 diluted to 5%.

B, Listeria monocytogenes (ATCC 19114); C, Escherichia coli (ATCC 25922); D, Staphylococcus aureus (ATCC 25923); E, Klebsiella aerogenes (ATCC 35029); F, Salmonella enteritidis (ATCC 13076).

Mean (Standard deviation); n=5.

In their composition, the EO blends present more than 90% oregano EO with a majority content of phenols, alcohols, ethers, aldehydes, hydrocarbons, and other components. Blend B4, with 85% EO, presented slightly lower inhibition halo values; however, these are still significant values demonstrating the inhibitory capacity against the studied microorganisms. The proposed blends showed inhibitory capacity for both Gram-positive bacteria and Gram-negative bacteria.

All EO blends tested demonstrated significant antifungal activity against *A. niger* and *A. brasilensis*, reaching inhibition halo diameters greater than 40 mm. These results indicate a strong inhibitory capacity of the tested EO blends, suggesting their potential as natural antifungal agents. The significant inhibition against both fungi highlights the blends' efficacy, possibly attributed to the synergy between the active components of the EOs, which affect cell membrane integrity and spore viability.

3.5 Sensory analysis of preselected blends

The results of the normality tests using the Shapiro-Wilk test indicated that B1, B2, and B3 did not meet the normality assumption, as their $p \le 0.05$. However, B4 had a $p \approx 1.0$, suggesting that this blend follows a normal distribution. Regarding the homogeneity of variances, the Levene test yielded a p-value of ≈ 0.28 , meaning the variances between groups were equal, fulfilling one of the requirements for conducting an ANOVA.

Since not all mixtures met the normality assumption, a nonparametric alternative, such as the Kruskal-Wallis test, was considered. This test yielded a value of 25.09 and p \leq 0.05, indicating a statistically significant difference between groups, i.e., at least one of the EO mixtures was significantly different from the others. The results of post-hoc comparisons using Tukev's test are shown in Table 7.

The preselected EO blends, evaluated by semi-trained tasters, showed a preference for B1, which had the highest average acceptance, followed by B3, B4, and B2, which showed statistically significant differences between the samples (p \leq 0.05). The small amounts of EOs other than oregano influenced the flavor responses of the proposed mixtures.

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Table 7. Sensory hedonic analysis of essential oil blends (n= 9). Tabela 7. Análise sensorial hedônica de misturas de óleos essenciais (n= 9).

()		
Blend	Mean (Standard deviation)	
B1	4.7 (0.5) a	
B2	2.0 (0.8) b	
B3	4.3 (0.5) ac	
B4	4.1 (0.3) ac	

Different letters indicate significant differences (p \leq 0.05).

3.6 Analysis of the selected EO blend

Table 9 presents the components of the selected EO blend (B1). This blend, B1, showed a large number of components, the majority being carvacrol (49%), β -citronellol (6.5%), p-cymene (5.5%), geraniol (5.1%), carvone (4.7%), linalool (3.8%), γ -terpinene (3%), 1,8-cineole (2.3%), (E)-caryophyllene (1.8%), myrcene (1.7%), α -terpineol (1.5%), thymol (1.6%), geranial (1.2%) and α -pinene (1%).

Figure 3 shows its FTIR spectrum. The EOs of oregano, rosemary, lemongrass, and eucalyptus improved antimicrobial responses in the studied blend. In contrast, lower responses were observed when combined with the EOs of basil and geranium. The EO blend contains major and minor components that somehow affect the antimicrobial response, but these remain to be elucidated.

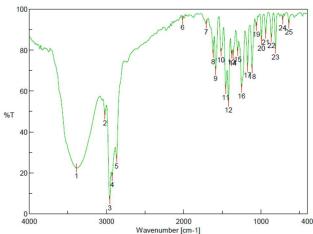


Figure 2. IR spectrum of the selected essential oil blend (B1). Figura 2. Espectro IR da mistura de óleos essenciais selecionada (B1).

The presence of the major and minor components of the EOs could change the antimicrobial response due to synergistic, antagonistic, additive, or indifferent effects (MOUREY; CANILLAC, 2002; IMAEL et al., 2012; PANDEY et al., 2014; NASCIMENTO et al., 2018; WANI et al., 2021). In the studied blends, better antimicrobial responses were observed between the EOs of oregano, rosemary, lemongrass, and eucalyptus without including basil and geranium oils (Figures 3 and 4).

Figure 3 shows the behavior of ternary mixtures of EOs with the presence of oregano EO (0.949 µL) constantly and the absence of basil and geranium about the inhibition or formation of the inhibition halo. In *P. aeruginosa*, it is observed that combinations with greater amounts of rosemary EO produce larger inhibition zones, while in L. monocytogenes, sensitivity decreases, and rosemary EO also influences inhibition. In *E. coli*, a better inhibitory response is observed; the combination of lemongrass, eucalyptus, and rosemary EOs generates a better response. In *S. aureus*, a blend of

rosemary and lemongrass EOs produces a better inhibitory response. In *K. aerogenes* and *S. enteritidis*, low sensitivity is observed.

Table 8. Chemical composition of the selected blend of essential oils.

Tabela 8. Composição química da mistura selecionada de óleos essenciais.

essenciais.			
Chemical compound	LRIe	LRIr	Content (%)
3-methylbutanal	653	654	0,002
tricyclene	925	926	0,002
α-thujene	933	931	0,597
α-pinene	941	939	1,028
camphene	953	954	0,286
sabinene	972	974	0,002
β-pinene	978	979	0,926
1-octen-3-ol	976	982	0,190
6-methyl-5-hepten-2-one	986	987	0,900
octan-3-ol	990	989	0,190
myrcene	992	991	1,744
6-methyl-5-hepten-2-ol	994	992	0,002
octanal	999	999	0,005
α-phellandrene	1002	1003	0,120
α-terpine	1014	1017	0,765
p-cymene	1022	1025	5,549
limonene	1028	1028	0,991
β-phellandrene	1031	1030	0,004
1,8-cineole	1033	1032	2,311
(z)-β-ocimene	1036	1037	0,021
(e)-β-ocimene	1046	1050	0,199
γ-terpinene	1058	1061	3,152
octan-1-ol	1065	1068	0,002
terpinolene	1087	1089	0,104
p-cymenene	1092	1091	0,002
linalool	1096	1097	3,811
3-methylbutyl 3-methylbutanoate	1105	1104	0,002
cis-rose oxide	1111	1109	0,002
trans-rose oxide	1130	1129	0,002
trans-pinocarveol	1140	1140	0,002
exo-citral	1144	1146	0,002
camphor	1146	1148	0,755
citronellal	1155	1153	0,067
δ-terpineol	1166	1166	0,002
borneol	1170	1169	0,863
cis-pinocamphone	1173	1175	0,003
rosefuran epoxide	1176	1177	0,003
terpinen-4-ol	1178	1179	0,488
α-terpineol	1191	1189	1,539
decanal	1202	1202	0,009
verbenone	1202	1202	0,002
β-citronellol	1204	1203	6,518
•	1235	1235	-
thymol methyl ether neral	1239	1240	0,190 0,636
	1246		1
carvone		1243 1253	4,747 5.147
geraniol	1255		5,147
geranial	1268	1267	1,226
bornyl acetate	1284	1287	0,029
thymol	1290	1290	1,614
carvacrol	1296	1299	49,089
α-terpinyl acetate	1348	1348	0,629
α-cubebene	1349	1351	0,001
citronellyl acetate	1355	1353	0,005
eugenol	1361	1359	0,574
α-ilangene	1373	1373	0,001

1376	1377	0,002
1382	1381	0,029
1392	1392	0,002
1408	1411	0,054
1421	1419	1,844
1442	1441	0,026
1454	1451	0,005
1457	1455	0,298
1461	1462	0,004
1480	1480	0,003
1485	1485	0,002
1502	1501	0,002
1513	1514	0,003
1520	1523	0,003
1548	1550	0,002
1564	1564	0,002
1577	1575	0,005
1585	1582	0,207
1587	1585	0,005
1610	1608	0,002
1651	1653	0,005
		99,697
	1382 1392 1408 1421 1442 1454 1457 1461 1480 1485 1502 1513 1520 1548 1564 1577 1585 1587 1610	1382 1381 1392 1392 1408 1411 1421 1419 1442 1441 1454 1451 1457 1455 1461 1462 1480 1480 1485 1485 1502 1501 1513 1514 1520 1523 1548 1550 1564 1564 1577 1575 1585 1582 1587 1585 1610 1608

LRI_c: índice de retención lineal experimental en DB-5MS. LRI_c: índice de retención lineal de referencia o base de datos en DB-5MS.

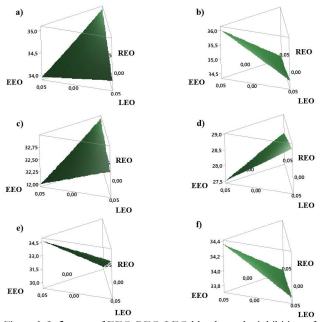
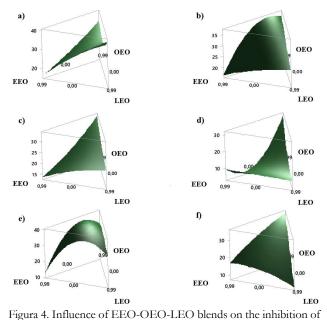


Figure 3. Influence of EEO-REO-LEO blends on the inhibition of bacterial growth. a) *Pseudomonas aeruginosa*; b) *Listeria monocytogenes*; c) *Escherichia coli*; d) *Staphylococcus aureus*; e) *Klebsiella aerugenes*; f) *Salmonella enteritidis*. REO, rosemary; EEO, eucalyptus; LEO, lemongrass; OEO, oregano; BEO, basil; GEO, geranium. Figura 3. Influencia de las mezclas EEO-REO-LEO en la inhibición

del crecimiento bacteriano. a) Pseudomonas aeruginosa; b) Listeria monocytogenes; c) Escherichia coli; d) Staphylococcus aureus; e) Klebsiella aerogenes; f) Salmonella enteritidis. REO, alecrim; EEO, eucalipto; LEO, capim-limão; OEO, orégano; BEO, manjericão; GEO, gerânio.

When the rosemary EO is maintained constant, the absence of basil and geranium reveals that *P. aeruginosa* exhibits sensitivity to oregano and lemongrass and, to a lesser extent, to eucalyptus. In *L. monocytogenes*, high synergy with the EOs is observed, with a broad inhibitory response, showing synergy with oregano and eucalyptus and, to a lesser extent, with lemongrass. In *E. coli*, the inhibitory response is high, showing high synergy with oregano, medium with

lemongrass, and lower with eucalyptus. In *S. aureus*, the inhibitory response occurs mainly with oregano and is lower with eucalyptus and lemongrass. In *K. aerogenes*, synergy with oregano and lemongrass is high and lower with eucalyptus. In *S. enteritidis*, synergy is high with oregano, medium with eucalyptus, and lower with lemongrass (Figure 4).



bacterial growth. a) Pseudomonas aeruginosa; b) Listeria monocytogenes; c) Escherichia coli; d) Staphylococcus aureus; e) Klebsiella aerogenes; f) Salmonella enteritidis. REO, rosemary; EEO, eucalyptus; LEO, lemongrass; OEO, oregano; BEO, basil; GEO, geranium. Figure 4. Influencia de las mezclas EEO-OEO-LEO en la inhibición del crecimiento bacteriano. a) Pseudomonas aeruginosa; b) Listeria monocytogenes; c) Escherichia coli; d) Staphylococcus aureus; e) Klebsiella aerogenes; f) Salmonella enteritidis. REO, alecrim; EEO, eucalipto; LEO, capim-limão; OEO, orégano; BEO, manjericão; GEO, gerânio.

4. DISCUSSION

The physical characteristics and composition of an essential oil vary depending on its origin, whether due to the source of the oil (flowers, leaves, root), the method of extraction, or agronomic characteristics such as harvest time, soil type, irrigation, plant maturity (Djapic, 2023), water stress (Sarmoum et al., 2019), among others (Nguyen et al., 2023), which explains the diversity of results in the physical and chemical composition of the oils studied. The density values of rosemary and oregano essential oils are similar to those of 0.89 and 0.93 g mL⁻¹, and the RI values of 1.46 and 1.47, respectively, expressed by Argote-Vega et al. (2017).

In the basil EO, Perveen et al. (2020) indicate that the major components are linalool (28.70%), methyl chavicol (38.2%), 1,8-cineole (8.5%), and other minor components. Therefore, the EO in this study would belong to the methyl chavicol/linalool classification. According to Kholiya et al. (2022), basil contained linalool as the major component, reaching 84%. The yield and composition of the EO were dependent on the flowering stage, the effect of planting density, and the effect of drying. Different constituents could be due to different environmental and geographical conditions, cultivation techniques, genetic factors, and plant nutritional status (HANIF et al., 2011).

In the geranium EO, compared to the results of Boukhaten et al. (2013), the citronellol content (30.2%) is

similar, while geraniol (7.6%) and linalool (3.2%) are lower in the sample of this research. On the other hand, citronellium formate shows only 2.5%, in contrast to the 9.3% reported by Boukhatem et al. (2013). In the study by Saraswathi et al. (2011), the major components are citronellol (29.90%), transgeraniol (18.03%), 10-epi-γ-eudesmol (8.27%), isomenthone (5.44%), linalool (5.13%), geranyl acetate (4.52%), γ-cadinene (2.89%), geranyl butyrate (2.53%), geranyl tiglate (2.50%) and gemacrene D (2.05%). Džamić et al. (2014) observed citronellol (24.54%), geraniol (15.33%), linalool (9.80%), 6,9-guaiadiene (7.08%), and isomenthone (2.86%) among the major components, which are similar to the citronellol, geraniol, and linalool contents of this study.

In the oregano oils analyzed by Salvo et al. (2018), in samples from different origins, concentrations ranged from 0.89 to 31% of carvacrol, 14.16 to 37.42% of thymol, 3.02 to 19.48% of p-cymene, 1.68 to 28.96% of β-terpineol, and 3.19 to 19.09% of p-cymene, among other minor components. In a work (Almas et al., 2021) on the essential oils of *E. globulus*, it is observed that the largest component is 1,8-cineole (51.62%) as the main constituent, a value similar to the 58.8% found in this study. In the case of lemongrass, the values differ from those presented by Tang et al. (2024), who showed (+)-citronellal (35.60%), geraniol (21.84%), and citronellol (13.88%) as the major compounds in the EO. Akinkunmi et al. (2016) reported a nerol content of 32% and geraniol of 51.70% when the EOs were fresh, since these compounds can transform into others over time.

The different components of the EOs influence their physical and chemical properties, as observed through GC-MS and FTIR composition.

Regarding the MIC, the 5% samples represent the minimum concentration that should be added to achieve inhibition. This value of 50,000 µg mL⁻¹ is similar to the value presented by Argote-Vega et al. (2017), who indicated that for rosemary and *S. aureus*, the MIC is 28.48 mg mL⁻¹, while wild oregano requires 37.2 mg mL⁻¹.

Each of the EOs showed antimicrobial and antifungal activity. Thus, lemongrass EO possesses antimicrobial characteristics, particularly inhibiting S. enteritidis, as Azizah et al. (2023) indicated. According to the analysis performed by Majda et al. (2022), rosemary EO contained 51.62% 1,8-cineole, 18.94% α -pinene, and 10.65% α -campholene aldehyde; however, its antimicrobial activity against E. coli was ineffective.

According to Cebi (2021), geranium EO contains geraniol, citronellol, and other terpene compounds. This EO exhibits a stretching band corresponding to the O-H bond around 3400 cm⁻¹; between 2800 and 3000 cm⁻¹, it exhibits a C-H stretching band in the methyl and methylene groups; while at 1735 cm⁻¹, it exhibits a C=O stretching band corresponding to the carbonyl groups. At 1600 cm⁻¹, a band due to C=C is observed, and between 1050 and 1150 cm⁻¹, a C-O band related to the ether and ester bond.

The EO mixtures obtained in their composition contain more than 90% oregano EO, with a majority content of phenols, alcohols, ethers, aldehydes, hydrocarbons, and other components. The blend B4, with 85% oregano EO, exhibited slightly lower inhibition halo values; however, these values are still significant, demonstrating the inhibitory capacity against the microorganisms studied. This is explained by the fact that EOs can penetrate the interior of bacterial cells, leading to their death. This characteristic is attributed to various components in the EOs, including lipophilic

functional groups (RAUT; KARUPPAYIL, 2014; WANI et al., 2021). The components of the optimized mixture mainly contain carvacrol, p-cymene, linalool, (B)-caryophyllene, and eucalyptol or 1,8-cineole, along with other minor components that provide antimicrobial properties.

Several researchers have demonstrated the presence of synergy, antagonism, or additivity between EOs. This study observed a synergy between oregano and rosemary as an antimicrobial response against the microorganisms studied. Thus, synergy was observed between oregano and rosemary (HONÓRIO, 2015), although antagonism was also observed between their main components, carvacrol and myrcene, according to Galluci et al. (2009). Additionally, synergy has been observed between carvacrol and p-cymene according to Ultee et al. (2000), carvacrol and linalool, according to Imael et al. (2012), carvacrol and citral according to Cao et al. (2021), carvacrol and 1,8-cineole according to De Sousa et al. (2012), oregano and basil have shown an additive effect according to Gutierrez et al. (2008), carvacrol and thymol according to Kissels et al. (2017), as well as it has been observed between O. vulgare and O. basilicum. Antagonism has been evidenced between thyme oil and A. eucalyptus (MEHRABANI et al., 2022). These observed results will depend on the components present in the EOs and, therefore, in their mixtures, with the major and minor components somehow affecting the antimicrobial response.

Fratini et al. (2021) demonstrated the synergistic antimicrobial effect of a mixture of four EOs rich in thymol, carvacrol, p-cymene, and cinnamaldehyde, the results of which were more effective than those obtained individually. Helal et al. (2019) also showed that EOs from seven aromatic plants from the Asir region exhibited significant activity against the fungus *Candida albicans*.

Ouedrhiri et al. (2021) observed synergistic effects among several EOs. Leite et al. (2022) demonstrated the benefit of including a mixture of oregano and copaiba EOs (0.025% each) in ground beef burgers. These samples showed better color during the exhibition, were more tender, and had greater antioxidant activity. However, the samples with oregano EO had the lowest acceptability. Therefore, it is necessary to have the sensory acceptability of the EO.

5. CONCLUSIONS

Antimicrobial activity can vary depending on the type of microorganism and can be achieved with pure essential oils and blends. However, given the diversity of microorganisms to which an essential oil may be exposed, it is crucial that the selected essential oil or blend can effectively respond to these different demands.

The main component of the blends was carvacrol; however, the antimicrobial activity results depend on the presence of minor components. Therefore, given the diversity of components, it is advisable to know the content of the oils before blending them. Additionally, due to the characteristic odors and flavors they produce, it is advisable to test them organoleptically to determine their acceptance before use.

The blend B1 exhibited antimicrobial activity against the microorganisms tested and was also accepted by the evaluators, making it a potential additive. The resulting blend could have a wide range of practical applications, from packaging, films, food, and more, thus contributing to infection prevention and extending product shelf life. Furthermore, blending essential oils with antimicrobial

properties could be a natural and biodegradable alternative to synthetic antimicrobials, aligning with sustainability principles and environmental friendliness.

6. REFERENCES

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Conflict of interest: The authors declare that they have no conflict of interest.



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