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ESSENTIAL OILS AS ANTIMICROBIAL AND ANTI-ADHESION AGENTS AGAINST BACTERIA *SALMONELLA* TYPHIMURIUM AND *STAPHYLOCOCCUS AUREUS*, AND YEASTS *CANDIDA ALBICANS* AND *SACCHAROMYCES CEREVISIAE*

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Abstract: A serious global problem with the increasing resistance of microorganisms to currently used antimicrobials has opened up the promotional research in the identification of new, more effective drugs with a broad spectrum of antimicrobial activities. Plant essential oils, due to the large biological and structural diversity of their components, are known to have many potential benefits. This study aimed to evaluate the antimicrobial and anti-adhesion activity of fifteen essential oils and their compounds against two bacterial and two yeast species responsible for food spoilage and infectious diseases. Antimicrobial activity was determined by testing the minimum inhibitory concentration (MIC), the minimum bactericidal concentration (MBC) and the minimum fungicidal concentration (MFC) of essential oils and compounds. The essential oils of *Cinnamomum zeylanicum* and *Eugenia caryophyllus* showed the highest antimicrobial activity with MICs ranging from 0.078 to 1.25 mg/mL, and 0.039 to 1.25 mg/mL, respectively. On the other hand, essential oils of *Eucalypti aetheroleum* and *Salvia officinalis* had significantly weaker antimicrobial properties than the others. Further, MICs were used to assess the inhibition of adhesion of bacteria *Salmonella* Typhimurium ATCC 25923 and *Staphylococcus aureus* ATCC 14208, and yeasts *Candida albicans* ATCC 10231 and *Saccharomyces cerevisiae* ATCC 9763 in a microtiter plate using the crystal violet staining method. Based on the percentage of adhesion inhibition, yeast *S. cerevisiae* ATCC 9763 showed a high level of antimicrobial resistance. *E. caryophyllus* had the strongest effect with inhibition up to 73%. Consistent with the antimicrobial susceptibility results, the most active anti-adhesion compounds were carvacrol and thymol. Considering the role of biofilm in food spoilage and clinical diseases, inhibition of the initial phase of biofilm formation by natural antimicrobial agents may be an alternative to commonly used synthetic ones.

Key words: *essential oils, antimicrobial agents, anti-adhesion agents, bacteria, yeasts*

INTRODUCTION

Multiple drug resistance of human and plant pathogenic microorganisms has been developed in recent years due to inadequate and non-selective use of synthetic antimicrobial agents commonly used in the treatment of infectious diseases and food preservation (Gulluce et al., 2007). While foodborne

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diseases are still a major health problem in the world, even in well-developed countries, food spoilage caused by various microorganisms is one of the most important challenges for the food industry leading to large economic losses. Pathogenic and spoilage microorganisms can adhere to and grow on food surfaces (vegetables, fruits, meat, seafood, dairy products, beverages), equipment and processing environments, resulting in biofilm formation (Orhan-Yanıkan et al., 2019; Valdivieso-Ugarte, Gomez-Llorente, Plaza-Díaz & Gil, 2019). Many biofilms are present in a variety of microbial infections, including medical environments and can appear on different pieces of equipment and contact surfaces such as stainless steel, glass, rubber or plastic, representing a potential source of infection. Microbial cells present within the biofilm are more resistant to antimicrobial agents than planktonic cells due to the presence of extracellular polymeric substances; thus, conventional antimicrobial agents used as control strategies in the food industry and treatment of infectious diseases have limited antimicrobial effect on the biofilm (Al-Shabib et al., 2017). This resistance involves using extremely high doses of antimicrobial agents that can be harmful due to the presence of toxic or carcinogenic by-products. Moreover, biofilm formation improves the ability of pathogenic microorganisms to survive stresssful conditions such as acidity, refrigeration, and disinfection (Srey, Jahid & Ha, 2013; Campana et al., 2017).

A range of synthetic antimicrobial agents is currently in use to improve food safety and reduce the growth of food spoiling microbes. However, due to toxicological reasons, the excessive use of synthetic preservatives and conventional disinfectants has been questioned, after the suspicion that synthetic antimicrobial agents are potentially harmful to human health. In this context, the use of essential oils and their bioactive components has become a promising area of research, and consumer demands direct towards novel types of efficient and healthy antimicrobial agents (Purkait, Bhattacharya, Bag & Chattopadhyay, 2018). The antimicrobial activity of the essential oil has been extensively studied against planktonic microorganisms, but the knowledge of their activities against microbial biofilms remains limited (Orhan-Yanıkan et al., 2019; Tomičić et al., 2022). In this regard, this study examined the antimicrobial and anti-adhesion properties of fifteen essential oils and their compounds against several microorganisms, most important for the occurrence of food spoilage and infectious diseases, such as gram-negative bacteria *Salmonella* Typhimurium ATCC 25923, gram-positive bacteria *Staphylococcus aureus* ATCC 14208, and two yeast species *Candida albicans* ATCC 10231 and *Saccharomyces cerevisiae* ATCC 9763.

MATERIALS AND METHODS

Strains and growth conditions

The antimicrobial and anti-adhesion activity of the essential oils was tested against gram-positive bacteria *S. aureus* ATCC 25923 (American Type Culture Collection, Rockville, Maryland, USA), gram-negative bacteria *S.* Typhimurium ATCC 14208 (American Type Culture Collection, Rockville, Maryland, USA), and two yeast species *C. albicans* ATCC 10231 (American Type Culture Collection, Rockville, Maryland, USA) and *S. cerevisiae* ATCC 9763 (American Type Culture Collection, Rockville, Maryland, USA). All strains were obtained from the Culture Collection of the microbiological laboratory at the Institute of Food Technology, University of Novi Sad, Serbia. Reference strains of bacteria were preserved in Trypto-Casein Soy Broth (TSB, Biokar, Beauvais, France), while yeast strains were preserved in Yeast Peptone Dextrose medium (YPD, Sigma-Aldrich, St. Louis, USA) supplemented with 40% glycerol at – 80 °C and revitalized from frozen stocks by cultivation on the Tryptone Soya Agar (TSA, Oxoid CM0131, Hampshire, UK) plates (bacterial strains) and Sabouraud Dextrose Agar (SDA, Sigma-Aldrich, Darmstadt, Germany) plates (yeast strains) for 24 h at 37 °C before performing the assays.

Essential oils

Eleven essential oils (EOs) namely *Cinnamomum zeylanicum*, Lauraceae (cinnamon), *Eucalypti aetheroleum*, Myrtaceae (eucalyptus), *Eugenia caryophyllus*, Myrtaceae (clove), *Lavandula angustifolia*, Lamiaceae (lavender), *Melaleuca alternifolia* Myrtaceae (tea tree), *Mentha piperita*, (peppermint), *Ocimum basilicum*, Lamiaceae (basil), *Origanum vulgare*, Lamiaceae (oregano), *Rosmarinus officinalis*, Lamiaceae (rosemary), *Salvia officinalis*, La-

miaceae (sage) and *Thymus vulgaris*, Lamiaceae (thyme) were purchased from local market in Novi Sad, Serbia (the producer not shown). The pure compounds α-pinene, carvacrol, eugenol and thymol were purchased from Sigma-Aldrich.

Determination of minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC) and minimum fungicidal concentration (MFC)

The minimum inhibitory concentration (MIC) of essential oils and compounds was determined using the broth microdilution method according to the guidelines of CLSI (Clinical and Laboratory Standards Institute (CLSI, 2008)) with a slight modification. Briefly, the tested EOs were first dissolved in dimethyl sulfoxide (DMSO, Sigma-Aldrich, St. Louis, USA) and incorporated into Mueller Hinton Broth (MHB, HiMedia, Mumbai, India) (for bacteria) and Sabouraud Dextrose Broth (SDB, Sigma-Aldrich, Darmstadt, Germany) (for yeast), and then two-fold serially diluted to make a concentration range from 0.039 to 640 mg/mL in 96-well microtiter plates.

The final concentration of DMSO did not exceed 0.5%. Subsequently, 100 μL of working inoculum suspension (1×10^6 CFU/mL) was added to each well. The inoculated plates were then incubated for 24h for bacterial strains and 48h for yeast strains at 37 °C.

In all experiments a positive (assay medium without EOs and with reference strains) and a negative control (growth medium without reference strains) were included. Assays were carried out in four independent replicates for each tested microorganism. After incubation, 20 μL of 0.0% resazurin solution (7-Hydroxy-3*H*-phenoxazin-3-one 10-oxide, HiMedia, India) was added as an indicator of bacterial growth to each well of microtiter plate and incubated for 24 h to determine a colour change. The lowest dilution with no colour change was considered as the MIC for that individual oil and compound. On the other hand, for the yeast strains, the absorbance was measured at 630 nm using a microplate reader (Thermo Fisher Scientific, Waltham, Massachusetts, USA). The MICs were determined as the lowest concentration that inhibits the visible growth of the test microorganisms. The minimum bactericidal concentration (MBC) and minimum fungicidal concentration (MFC) were determined by seeding an aliquot of 10 μL suspension of microorganisms in triplicate on Mueller-Hinton agar (MHA, HiMedia, Mumbai, India) plates for bacteria and SDA plates for yeast from wells where no visible growth was observed and cultures was incubated for 24 h at 37 °C (NCCLS, 2003). The lowest concentration of essential oils and compounds that did not yield any growth on the solid medium after the incubation period was recorded as the MBC and MFC.

Adhesion assay

Adhesion assays were performed as previously described (Tomičić, Zupan, Matos & Raspor, 2016) with few modifications. Prior to testing, selected bacterial and yeast strains were grown on TSA and SDA plates for 24 h at 37 °C, respectively. After the incubation, a loopful of actively growing cells of bacteria was suspended in TSB, while yeast cells were suspended in SDB medium. The cell concentration was adjusted to 0.5 McFarland standard turbidity to achieve a final cell concentration of 1×10^7 cells/mL. The assay was initiated by the addition of 200 μL cell suspensions with and without the presence of essential oils and compounds into wells of a 96-well polystyrene microtiter plate (Nunc, Roskilde, Germany), which were then incubated for 24 h at 37 °C. The cells of the tested bacteria and yeast were exposed to 1 × MIC of essential oils and compounds during 24 h of adhesion. The experiments were performed with eight replicates.

The amount of bacterial and yeast cells adhered to the polystyrene plates was measured using the crystal violet (CV) staining method (Tomičić et al., 2016). The amount of adhered cells, that is, the concentration of the released crystal violet was determined by measuring the optical density at 630 nm using a microplate reader (Thermo Fisher Scientific, Waltham, Massachusetts, USA).

Statistical analysis

Descriptive statistical analyses for calculation of the means and standard error of the mean were performed using Microsoft Excel software (Microsoft Office 2013). Results were expressed as mean ± standard deviation (SD). Statistical analysis was performed using the software package StatSoft Statistica, ver. 10 (IBM, Armonk, NY, USA). The effect of essential oils and compounds on bacterial adhesion was analyzed and the statistical significance determined using one-way ANOVA

analysis of variance. A p-value of <0.05 was considered statistically significant.

RESULTS AND DISCUSSION

The growing challenge of microbial resistance to many antimicrobial agents has prompted the search for alternative sources of antimicrobial agents, which are believed to be found abundantly in plants. Some essential oils are known to have various health benefits and have been used as alternative medicines for the prevention and treatment of many infectious diseases, as well as food spoilage microorganisms (Tomičić et al., 2022).

Accordingly, the efficacy of 15 essential oils and their compounds against planktonic cells of reference strains of bacteria *S.* Typhimurium ATCC 14028 and *S. aureus* ATCC 25923, and yeast *C. albicans* ATCC 10231 and *S. cerevisiae* ATCC 9763 was first assessed by determining the MIC and MBC/MFC, as shown in Table 1. As can be seen, most of them exhibited significant antimicrobial activity against gram-positive and gram-negative bacteria, and especially against yeast. The MIC, MBC and MFC values ranged from 0.039 to 320, 0.625 to 320 and 0.039 to 40 mg/mL for the tested strains, respectively. The results of MIC indicated that the most sensitive strain was the *C. albicans* ATCC 10231 followed by *S. cerevisiae* ATCC 9763, *S. aureus* ATCC 25923 and the most resistant one was *S.* Typhimurium ATCC 14028. There is evidence in the literature that gram-negative bacteria are more resistant to the effect of plant essential oils than gram-positive bacteria (O'Bryan, Pendleton, Crandall & Ricke, 2015; Semeniuc, Pop & Rotar, 2017). The presence of an outer membrane is one of the features that differentiate gram-negative from gram-positive bacteria. Gram-negative bacteria are surrounded by a thin peptidoglycan cell wall, which itself is surrounded by an outer membrane containing lipopolysaccharide that creates a barrier toward hydrophobic compounds such as those found in EOs and allows gram-negative bacteria to be more resistant to EOs and other natural extracts with antimicrobial activity (Nazzaro, Fratianni, De Martino, Coppola & De Feo, 2013). Planktonic cells showed wide variability in resistance to essential oils, with *C. zeylanicum* and *E. caryophyllus* being the most effecttive, followed by *O. vulgare*, *T. vulgaris* and *O. basilicum*. Cinnamon bark oil is one of the most effective EOs against common foodborne pathogens (Valdiviesо-Ugarte et al., 2019). It should be noted that *C. zeylanicum* in our study showed a considerable antibacterial effect against *S. aureus* at the concentration of 0.312 mg/mL similar to the data obtained by Firmino et al. (2018). As in the case of the antibacterial activity, *E. caryophyllus* was found to have substantial antifungal potential against *C. albicans* ATCC 10231 and *S. cerevisiae* ATCC 9763 with MIC values of 0.039 and 0.078 mg/mL, according to microdilution assay, and for MFC values of 0.039 and 0.156 mg/mL (Table 1). On the other hand, the lowest antimicrobial activity was determined for the essential oils of *E. aetheroleum*, *R. officinalis* and *S. officinalis*. However, *R. officinalis* showed slightly lower antifungal activity than *E. aetheroleum*, and it was also stronger than *S. officinalis*. No significant differences were observed between the *R. officinalis* and *S. officinalis* against yeast.

The major phenolic compounds carvacrol and thymol are well known for their higher biological activity compared to other components such as α-pinene and eugenol, the major metabolites of some essential oils (Swamy, Akhtar & Sinniah, 2016; Cáceres, Hidalgo, Stashenko, Torres & Ortiz, 2020). It can be seen that thymol exhibited high antimicrobial activity with MICs ranging from 0.078–0.3125 mg/mL, while carvacrol showed stronger antimicrobial activity with MBC at 0.156 mg/mL and MFC at 0.039-0.078 mg/mL.

Although the mechanism of action of thymol and carvacrol has not been completely elucidated, it appears to act through cell membrane disruption, leading to an increase in cell membrane permeability, decrease membrane potential, dissipation of the pH gradient, intracellular leakage of nutrients, and subsequent cell death (Nazzaro et al., 2013; Cáceres et al., 2020). In previous research, essential oils of *O. vulgare* and *T. vulgaris* had remarkable antimicrobial effects, which were associated with the presence of phenolic compounds (Mith et al., 2014; Sakkas & Papadopoulou, 2017). Also, Giordani et al. (2004) reported that *C. albicans* was inhibited by the *T. vulgaris* which is rich in thymol, and *O. vulgare* which is rich in carvacrol, also in accordance with results obtained in our

Table 1.
Antimicrobial activity of essential oils and their compounds against bacteria and yeasts represented by the values of minimum inhibitory concentration (MIC) and minimum bactericidal/fungicidal concentration (MBC/MFC)

	Bacteria				**Yeasts**			
	***Salmonella* Typhimurium ATCC 14028**		***Staphylococcus aureus* ATCC 25923**		***Candida albicans* ATCC 10231**		***Saccharomyces cerevisiae* ATCC 9763**	
Essential oils	MIC (mg/mL)	MBC (mg/mL)	MIC (mg/mL)	MBC (mg/mL)	MIC (mg/mL)	MFC (mg/mL)	MIC (mg/mL)	MFC (mg/mL)
Cinnamomum zeylanicum	1.25	1.25	0.31	0.63	0.08	0.16	0.16	0.16
Eucalypti aetheroleum	320.00	320.00	320.00	320.00	2.50	5.00	10.00	10.00
Eugenia caryophyllus	1.25	1.25	0.63	1.25	0.04	0.04	0.08	0.16
Lavandula angustifolia	160.00	160.00	80.00	160.00	2.50	2.50	5.00	10.00
Melaleuca alternifolia	20.00	40.00	10.00	20.00	1.25	2.50	2.50	2.50
Mentha piperita	80.00	80.00	5.00	10.00	0.63	0.63	1.25	1.25
Ocimum basilicum	20.00	40.00	20.00	20.00	0.31	0.31	0.63	1.25
Origanum vulgare	10.00	10.00	2.50	2.50	0.16	0.16	0.31	0.31
Rosmarinus officinalis	80.00	80.00	160.00	160.00	10.00	10.00	10.00	10.00
Salvia officinalis	160.00	160.00	40.00	80.00	20.00	20.00	40.00	40.00
Thymus vulgaris	10.00	10.00	20.00	20.00	0.16	0.31	0.63	0.63
Compounds								
α-Pinene	1.25	2.50	2.50	2.50	0.31	0.31	0.63	0.63
Carvacrol	0.08	0.16	0.16	0.16	0.04	0.04	0.04	0.08
Eugenol	0.63	0.63	0.16	0.31	0.16	0.16	0.16	0.16
Thymol	0.16	0.31	0.31	0.31	0.08	0.08	0.16	0.16

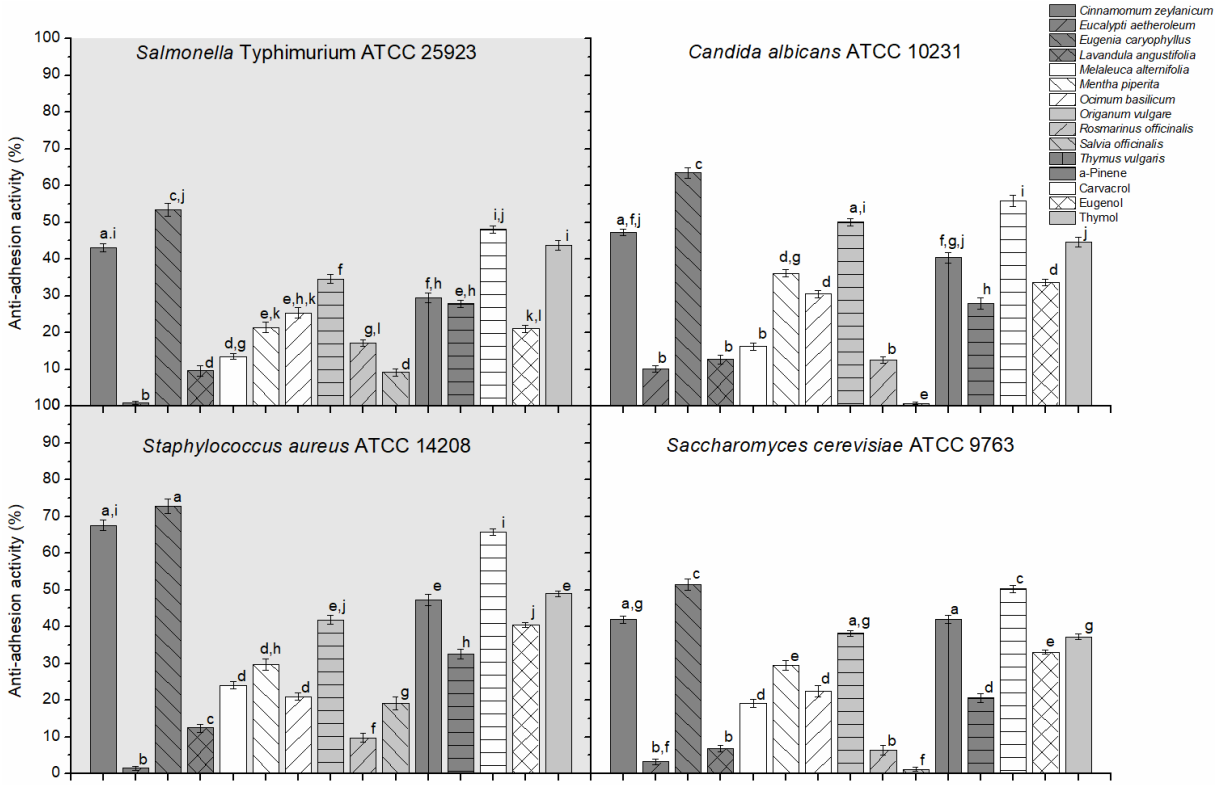

Figure 1. The effect of MIC concentration of essential oils and main EO compounds on the initial phase of biofilm formation of bacteria *S.* Typhimurium ATCC 25923 and *S. aureus* ATCC 14208, and yeast *C. albicans* ATCC 10231 and *S. cerevisiae* ATCC 9763 (n=8). *Different letters (a, b, c, d...) mark significant differences between essential oils and compounds ($p < 0.05$)

research. In another study including nine essential oils, oregano and thyme displayed a powerful antifungal activity against clinical strains of *Candida spp.* (Tomičić et al., 2022)

Thyme essential oil is one of the ten most commercial essential oils worldwide, used as a natural food preservative and aromatic additive to various foods and beverages, while oregano essential oil as a food preservative is quite limited because of its strong odour, which adversely affects the organoleptic properties of food and could result in low consumer acceptance (Cosentino et al., 1999; Sakkas & Papadopoulou, 2017; Valdivieso-Ugarte et al., 2019).

One important concern in the food industry and medicine is the presence of biofilms. Bacteria and yeast can easily adhere to the surface of food processing equipment and medical devices, forming a biofilm. Microbial biofilm, a matrix of extracellular polymeric substances, has become one of the main factors for the rapid emergence of resistant microorganisms to conventional antimicrobial agents (Valdivieso-Ugarte et al., 2019; Tomičić et al., 2022). Therefore, the next step of this study was to evaluate the anti-adhesion activity of MIC of essential oils and main compounds against selected bacteria and yeast.

A high inhibitory effect on the initial phase of biofilm formation was observed for most of the tested essential oils as shown in Figure 1. As evident, they were less active against gram-negative than gram-positive bacteria. Based on the percentage of adhesion inhibition, yeast *S. cerevisiae* ATCC 9763 showed the least sensitivity to the tested essential oils and compounds. Likewise, for planktonic cells, *E. caryophyllus* showed the strongest effect with a percentage of inhibition of 53.5, 73, 63.5 and 51.5% on *S.* Typhimurium ATCC 14208, *S. aureus* ATCC 25923, *C. albicans* ATCC 10231 and *S. cerevisiae* ATCC 9763, respectively. The essential oils of *C. zeylanicum*, *O. vulgare* and *T. vulgaris* also showed a strong anti-adhesion effect. Moreover, *M. piperita* and *O. basilicum* were effective against all tested bacteria and yeast by inhibiting the adhesion up to 36% and 30.5%, respectively. More recently, Tomičić et al. (2022) concluded that the essential oils of

Cinnamomum verum, *O. vulgare* and *T. vulgaris* possessed promising anti-adhesion activity reducing the initial phase of biofilm formation and pre-formed 24 h biofilm of *Candida glabrata* in the range of 40 to 73% when 3/4 of the MIC concentration was tested, suggesting that the use of natural antimicrobial agents could provide alternative or supplementary ways for the treatment of pathogenic yeast. These EOs strongly inhibited the initial phase of biofilm formation, highlighting them as excellent candidates for the discovery of new natural agents and the best candidate for further studies in the design and development of drugs against microbial resistance. On the other hand, the anti-adhesive properties of the essential oils of *E. aetheroleum* and *S. officinalis* were significantly lower than the others. With a MIC value of 320 mg/mL, eucalyptus EO did not show any anti-adhesion effect against bacteria, while MIC values of sage EO showed a weak effect on yeast. Further, it is important to highlight that peppermint EO inhibited the adhesion of *C. albicans* and *S. cerevisiae* more efficiently than the essential oils of eucalyptus, lavender, tea tree, basil, rosemary and sage. No significant differences were observed between lavender and rosemary essential oils against yeast.

According to the results of antimicrobial susceptibility, the most active compounds were carvacrol and thymol, which showed the most remarkable anti-adhesion effect against bacterial and yeast strains with a percentage of inhibition up to 66% and 49%, respectively. These results are consistent with previous studies (Knowles, Roller, Murray & Naidu, 2005), which demonstrated that carvacrol inhibits matrix formation during the early stages of biofilm development. Other studies have shown that the anti-biofilm activity of carvacrol is associated with disruption of quorum sensing (Knowles et al., 2005; Trevisan et al., 2018). It seems likely that there is a relationship between the high activity of tested oils and the presence of phenol components such as thymol and carvacrol.

CONCLUSIONS

Our results demonstrated that the tested essential oils and compounds exhibited anti-microbial activity and anti-adhesion potential against gram-negative bacteria *S.* Typhimurium, gram-positive bacteria *S. aureus*, and two yeast species *C. albicans* and *S. cerevisiae*. Among the essential oils, *E. caryophyllus* was the most effective, while *C. zeylanicum*, *O. vulgare* and *T. vulgaris* also showed strong antimicrobial and anti-adhesion potential. It can be also concluded that the activity was more pronounced against gram-positive bacteria than gram-negative bacteria, and the most resistant strain was yeast *S. cerevisiae*. This research should help to clarify the application of these essential oils for the treatment of many infectious diseases and food preservation in the future.

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ETARSKA ULJA KAO ANTIMIKROBNA I ANTIADHEZIVNA SREDSTVA PROTIV BAKTERIJA *SALMONELLA* TYPHIMURIUM I *STAPHYLOCOCCUS AUREUS*, I KVASACA *CANDIDA ALBICANS* I *SACCHAROMYCES CEREVISIAE*

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Sažetak: Rastući globalni problem usled povećane rezistencije mikroorganizama na najčešće korišćena antimikrobna sredstva, podstakao je istraživanja u cilju identifikacije novih, efikasnijih lekova širokog spektra delovanja. Poznato je da etarska ulja biljaka, zbog velikog biološkog i strukturnog diverziteta svojih komponenti, imaju mnoge potencijalne prednosti. Cilj ovog istraživanja bio je da se proceni antimikrobni i antiadhezivni efekat petnaest etarskih ulja i njihovih komponenti protiv dve bakterijske vrste i dve vrste kvasaca uzročnike kvarenja hrane i zaraznih bolesti. Antimikrobna aktivnost je određena ispitivanjem minimalne inhibitorne koncentracije (MIC), minimalne baktericidne koncentracije (MBC) i minimalne fungicidne koncentracije (MFC) etarskih ulja i komponenti. Rezultati su pokazali da su etarska ulja *Cinnamomum zeylanicum* i *Eugenia caryophyllus* imala najveću antimikrobnu aktivnost sa vrednostima MIC u rasponu od 0.078 do 1.25 mg/mL, odnosno 0.039 do 1.25 mg/mL. S druge strane, etarska ulja *Eucalypti aetheroleum* i *Salvia officinalis* su pokazala znatno slabija antimikrobna svojstva od ostalih. U daljem istraživanju, koncentracije MIC su korišćene za procenu inhibicije adhezije bakterija *Salmonella* Typhimurium ATCC 25923 i *Staphylococcus aureus* ATCC 14208, i kvasaca *Candida albicans* ATCC 10231 i *Saccharomyces cerevisiae* ATCC 9763 korišćenjem metode bojenja kristal violetom u mikrotitar pločama. Na osnovu procenta inhibicije adhezije, kvasac *S. cerevisiae* ATCC 9763 je pokazao visok stepen antimikrobne rezistencije. Pored toga, *E. caryophyllus* je imalo najjači efekat sa inhibicijom adhezije do 73%. U skladu sa rezultatima antimikrobne osetljivosti, najaktivnija antiadhezivna jedinjenja bila su karvakrol i timol. S obzirom na ulogu biofilma u kvarenju hrane i infektivnim bolestima, inhibicija početne faze formiranja biofilma prirodnim antimikrobnim agensima može biti alternativa uobičajeno korišćenim sintetičkim agensima.

Ključne reči: *etarska ulja, antimikrobna sredstva, antiadhezivna sredstva, bakterije, kvasci*

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