Essential Oils in the Control of *Listeria monocytogenes*

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Contributor: György Schneider, Anita Steinbach, Ákos Putics, Ágnes Solti-Hodován, Tamás Palkovics

Listeria monocytogenes is a foodborne pathogen, the causative agent of listeriosis. Infections typically occur through consumption of foods, such as meats, fisheries, milk, vegetables, and fruits. Chemical preservatives are used in foods; however, due to their effects on human health, attention is increasingly turning to natural decontamination practices. One option is the application of essential oils (EOs) with antibacterial features, since EOs are considered by many authorities as being safe.

Keywords: essential oil ; efficacy ; food ; method ; antibacterial ; preservation

1. Methods to Reveal Antilisterial Activity of Essential Oils and Their Active Components

A wide range of methods are available to study the antilisterial activities of essential oils (EOs). The most effective for activity screening is the simple drop plate or paper filter-based disc diffusion method [1], which is used on the lawn of the test organism. Other studies preferred using the agar diffusion assay [2]. To define the lowest EO concentration which inhibits proliferation and which kills *L. monocytogenes*, the terms minimal inhibitory and minimal bactericidal concentration (MIC and MBC) are used. These values can be determined using macro- or microdilution methods, in glass reagent tubes [3][4][5] or in 96-well microdilution plates [6][7], respectively. Due to solubility problems of the hydrophobic EOs and their compounds, the usage of detergents (e.g., Tween-20 and Tween-80) is sometimes required [8]. Transmission and scanning electron microscopy (TEM and SEM) are adequate techniques to visualize the morphological changes accompanying antimicrobial effects [3][9][10][11][12]. To identify active compounds responsible for the antilisterial activity of an EO, bioautography is a proper method. This is based on thin-layer chromatography (TLC) performed on silica gel [13]. Active compounds are identified on the basis of their Rf values, while nonidentifiable active volatile compounds can be cut out from the silica gel and analyzed using the headspace solid-phase microextraction method coupled to gas chromatography–mass spectrometry (HS-SPME/GC-MS) [14]. With this, the purity or percentage composition of antimicrobial active compounds from silica gel can be determined.

For the biofilm-inhibitory or biofilm-degrading capacity of EOs or active compounds, the classical crystal violet staining assay, performed in 96-well microplate format, is most commonly used [7][15][16][17]; however, other methods, such as the TEMPO system (bioMérieux), VIDAS system (bioMérieux), or the discrete element method (DEM), have also been suggested [18]. Polystyrene, polypropylene, polyethylene, glass, and stainless steel are typically tested abiotic surface materials [9][15][19]. SEM and Confocal laser scanning electron microscopy (CLSM) are used to analyze changes in the biofilm integrity as a result of treatment [20].

The molecular changes accompanying the antilisterial activities of EOs and their compounds uncovered using the above methods can be further analyzed with molecular biological tools. Changes in protein profiles are often studied with 1D [3] [10] or with the more detailed 2D polyacrylamide gel electrophoresis (PAGE). A fast alternative of 1D PAGE is capillary electrophoresis, in which expression differences between treated and control samples can be detected in a couple of minutes. A similar quantitative analysis uses liquid chromatography–mass spectrometry (LC–MS/MS) [12].

Further analysis of affected proteins, isolated with 2D PAGE, requires cutting and extraction from the acrylamide gel, followed by separation with liquid chromatography–mass spectrometry (LC–MS/MS) [12][21].

Today, high-throughput molecular biological methods are effectively used to uncover the underlying molecular events of bacterial metabolism in the presence of EOs or their active compounds. Whole-transcriptome analysis (WTA) is a proper approach to get a global view of the level of RNA synthesis [22], while the involvement of individual target genes, such as virulence-associated genes, can be further analyzed more precisely using the reverse transcription quantitative polymerase chain reaction (RT-qPCR) [10][23][24].

Certain enzymatic assays are also preferably used to gain insight into which part of the metabolism is affected on an enzymatic level. Measurement of the level of β-galactosidase and ATPase gives feedback about the energy metabolism of

L. monocytogenes, while the appearance of alkaline phosphatase outside the cell suggests weakened cell-wall integrity [3][10]. Membrane integrity can also be studied by quantifying the appearance of extracellular DNA in the medium [24].

2. Essential Oils with Antilisterial Activities

A broad range of EOs have been investigated for their antilisterial activity in the last two decades. Results of these tests are summarized in **Table 1**. Most of the represented articles focused on the activity of single EOs, but some also investigated synergistic effects when different EOs were combined [25][26]. The significance of this is that most EOs have a strain-dependent effect on *L. monocytogenes* [26][27]; therefore, a combination of different EOs could be an adequate approach against this foodborne pathogen in practice.

Results indicate that members of the Lamiaceae family, involving different *Thymus* and *Oregano* spp., showed the most extended antilisterial activity. Additionally, *Cinnamomun* spp. was proven to be effective. Typically, the major compounds were responsible for the antilisterial activities, especially if they belonged to the groups of mono- and sesquiterpenes. For the antilisterial effects, in most cases, compounds such as carvacrol, thymol, p-cymene, alpha-pinene, terpinene, or citral were responsible [27][28].

Most of the investigated EOs were extracted by steam distillation and originated from different countries and different producers. Especially in earlier studies, the compound composition (determined by gas chromatography) of the investigated EOs was not presented, which is a shortcoming that compromises the comparability of the different studies. This is an important issue as the compound composition of EOs is determined by geographical localization, weather, and time of harvest [29][30], which can influence the test results. A good example is that the compound composition of oregano EO in three studies differed significantly [27][31][32]; in the study of Gottardo, carvacrol content was 91%, whereas, in the studies of Maggio and Pesavento, these values were 68% and 71.8%, respectively. This was also the case with thyme showing differences in major compound content, albeit without influencing the antilisterial activity [19][27][33].

Another problem in the comparability of the results is that the bacterial cell numbers applied in different studies, either for the simple drop plate method or for MIC and MBC determinations, showed discrepancies. Furthermore, during tests, different media were used, e.g., Luria–Bertani (LB), Mueller–Hinton Broth (MHB), Brain Heart Infusion (BHI), and Peptone Yeast glucose (PYG) [1][3][4]. In most cases, in vitro tests were performed at 37 °C, whereas tests were only rarely conducted at lower temperatures under refrigerated conditions, which are mostly applied in food systems. Certainly, it would be a mistake to overstate the importance of the above factors. Moreover, since growth conditions influence gene regulation and, thus, phenotypic heterogeneity in bacteria [34], these factors could also influence the sensitivity of *L. monocytogenes* to EOs.

The antilisterial effect of EOs, summarized in **Table 1**, was mostly investigated using the standard disc diffusion technique [35], as this is the simplest screening method. In positive cases, the diameter of an inhibition zone around the EO spot or disc was typically between 20 and 30 mm, as demonstrated in several cases: black seed oil (31.50 mm) [35], broccoli sprout extract (17.84 ± 0.34 mm) [36], *Citrus medica* L. var. *sarcodactylis* Swingle *citron* oil (23.45 ± 1.23 mm) [22], *Ceratonia siliqua* EO (17 ± 0.3 mm) [6], *Hibiscus surattensis* L. calyces EO (25.26 ± 1.53 mm) [4], and Melaleuca *alternifolia* EO (30 ± 8.8 mm). In addition to this method, the agar diffusion assay [15][22][27], disc volatilization method [37], and plate colony counting [3] in pure, nanoliposome [36], nanocapsule [36], nanoemulsion [1][38][39], and liposome [40] systems were used. The advantage of the use of nanoemulsions is that this formulation is able to increase the biological activity and stability of EOs [41]. Nanoemulsion systems consist of three components: EO, water, and a nonionic surfactant, e.g., Tween-80 [1][39]. These three components are mixed, and then the particle size can be decreased using a sonicator [1].

Considering the tests, researchers have to emphasize again that the effect of EOs can be strain-dependent. A good example demonstrating this was a recent study in which the EO of *Melissa officinalis* was tested on three strains of *L. monocytogenes* (LMG 13305, 16779, and 16780), and three different inhibition zone diameters were found: 38.4 ± 4.2 mm, 54.6 ± 1.3, mm and 48.6 ± 1.7 mm, respectively [17]. In the case of *Schinus terebinthifolius* Raddi, EOs were produced from both ripe and unripe fruits. The inhibition zone of the latter was 35.22 ± 0.79 mm, while that of the former was 40.86 ± 0.31 mm [42].

Another aspect to be considered is which part of the plant and which procedure were used for the extraction. In a recent study, the authors revealed that the antilisterial effect of thyme was the strongest if acetone extract from the leaves was used, while ethanolic extract from the seeds exhibited the lowest antilisterial activity [43].

In most of the recent studies, kinetic assays were also performed in order to reveal the course of the antilisterial effect [11][44][45]. Kinetic curves are necessary if molecular changes on genomic and proteomic levels, accompanying the antilisterial

effect, are intended to be investigated [22]. Through these analyses, the antibacterial mode of action of certain EOs can be revealed.

Since *L. monocytogenes* is able to form biofilms, a number of experiments focused on the antibiofilm capacity of EOs [8] [19]. In such experiments, the biofilm-forming capacities of *L. monocytogenes* strains were hindered by *Cinnamomun zeylanicum* or *Eugenia caryophyllata* EOs [46]. Furthermore, it was also investigated whether EOs have the ability to destroy the already established biofilm on certain surfaces. They found that, within 2 h, clove could already drastically weaken the established biofilm. Such capacity is not typical for all EOs, as demonstrated by Guo et al. After establishing a firm biofilm in 72 h, they treated it with *Citrus Changshan-huyou* EO for 24 h, but the formed biofilm remained intact after treatment [9].

Revealing the antilisterial effect in in vitro studies is inevitable before considering practical uses; from this perspective, the results of screening and exploring the antimicrobial mode of action are all relevant issues, but the real challenge is always how a certain EO with potential antilisterial effects performs under harsh conditions if applied in different food systems.

Table 1. Summary of antilisterial effects of different EOs.

Source of Essential Oil	Major Compounds Investigated in the Study	Applied Method	Experimental Condition	MIC (µL/mL) or Inhibition Zone (mm)	MBC (µL/mL)	Reference
Achillea millefolium	**Caryophyllene, 1,8-cineole, bornyl acetate, 1-terpinen-4-ol, β-pinene, camphor**	**Agar disc diffusion assay, MIC, MBC, biofilm assay**	**BHI broth using a broth microdilution method in the 96-well round-bottomed polystyrene microtiter plates**	**31.3 µL/mL 16 mm**	**62.5 µL/mL**	**Jadhav, Shah et al. [15]**
***Allium sativum* L.**		**Disc diffusion method, MIC,**	**96-well micro-dilution plates with U-bottom wells**	**37.5 µL/mL**		**Razavi Rohani, Moradi et al. [47]**
Allium vineale		**Disc diffusion method, diameter of inhibition zone**	**Turkish Herby Cheese**	**8–15 mm, depending on the strain**		**Sagun, Durmaz et al. [48]**
Brassica oleracea* var. *italica		**Nanoliposome, nanocapsule, AOA, SEM, MIC**	**BSE nanoliposome, ricotta cheese**	**0.8 µL/mL**		**Azarashkan, Farahani et al. [3]**
Caryophyllorum salisque		**Nanoemulsion, MIC, characterization of NEs, agar well diffusion method, inhibition zone, TEM**	**Egyptian Talaga cheese, 96-well plate**	**45.2 ± 34.25 mm**		**Elsherif and Talaat Al Shrief [41]**
Ceratonia siliqua	**Nonadecane, heneicosane, naphthalene, 1,2-benzenedicarboxylic acid dibutylester, heptadecane, hexadecanoic acid, octadecanoic acid, 1,2-benzenedicarboxylic acid, phenyl ethyl tiglate, eicosene, farnesol 3, camphor, nerolidol, *n*-eicosane**	**Agar diffusion method MIC, MFC, MTT test, cytotoxicity assay**	**96-well microplates**	**2.5 µL/mL 17 ± 0.3 mm**		**Hsouna, Trigui et al. [6]**
Chaerophyllum macropodum		**Disc diffusion method, diameter of inhibition zone**	**Turkish Herby cheese**	**7–13 mm, depending on the strain**		**Sagun, Durmaz et al. [48]**
Cinnamomum zeylanicum		**Agar disc diffusion assay MIC, MBC, sensory evaluation**	**Raw minced meat**	**7.5 µL/mL 7–28.7 mm, depending on strain and concentration**	**7.5 µL/mL**	**Pesavento, Calonico et al. [2]**
Cinnamomum cassia	**Cinnamaldehyde, 2-propenal, acrylic acid, benzaldehyde**	**Agar diffusion, MIC, EO microencapsulation, sensory analysis, encapsulation efficiency**	**Italian salami**	**3 µL/mL 0.8–38 mm, depending on the concentration**		**Gottardo, Bidusk et al. [31]**

Source of Essential Oil	Major Compounds Investigated in the Study	Applied Method	Experimental Condition	MIC (µL/mL) or Inhibition Zone (mm)	MBC (µL/mL)	Reference
Cinnamomum cassia Blume	*trans*-Cinnamaldehyde, cinnamyl acetate	MIC, biofilm, DEM method	96-well microtiter plates	0.41 ± 0.02 µL/mL		Bermúdez-Capdevila, Cervantes-Huamán et al. [8
Cinnamomum zeylanicum	Eugenol, cinnamaldehyde, cinnamyl acetate, β-phelandrene	MIC, biofilm, DEM method	96-well microtiter plates	4.56 ± 0.2 µL/mL		Bermúdez-Capdevila, Cervantes-Huamán et al. [8
Cinnamomun zeylanicum	Cinnamaldehyde	MIC, biofilm, SEM, Box–Behnken experimental design	Plate, Falcon tubes	1.6 µL/mL		Vidács, Kereke et al. [19]
Cinnamomun zeylanicum		MIC, MBC, biofilm, eDNS, cytotoxicity, qPCR	24-well culture plate	range 10 µL/mL	50 µL/mL	Banerji, Mahamune et al [24]
Citrus Changshan-huyou		Disc diffusion assay, MIC, time-to-kill assay, SEM, TEM, RNA-seq, biofilm assay, SEM, CLSM	Ribo-Zero rRNA Removal Kit	40 µL/mL 25.48 ± 1.41 mm	80 µL/mL	Guo, Gao et al. [
Citrus medica L. var. *sarcodactylis* Swingle		Agar diffusion assay (MIC)	96-well tissue culture plate	40 µL/mL/ 23.45 ± 1.23 mm		Guo, Hu et al. [2
Citrus sinensis		Nanoemulsions (MIC, MBC), disc diffusion assay, time-to-kill assay, antibiofilm assay	MHA, 96 well microtiter plate, 24 well microtiter plate	9 ± 0.31 mm–13 ± 0.75 mm, depending on the concentrations		Das, Vishakha e al. [1]
Citrus limon var. *pompia*		Disc volatilization method, time-to-kill assay, SEM, TEM	Ricotta salata cheese	0.086 µL/mL		Fancello, Petrett et al. [37]
Citrus limon var. *prompia*	Limonene, γ-terpinene, α-terpineol, β-pinene, β-myrcene, citral	Shelf-life evaluation, cell constituent release, crystal violet assay, SEM, sensory evaluations	RTE vegetable salads (carrot, tomato, green oak lettuce, red cabbage)			Parichanon, Sattayakhom et al. [49]
Cuminum cyminum		Nanoemulsion, MIC, characterization of NEs, agar well diffusion method, inhibition zone, TEM	Egyptian Talaga cheese, 96-well plate	50.23 ± 15.7 mm		Elsherif and Talaat Al Shrief [41]
Cymbopogon citratus		Liposome system	Cheese			Cui, Wu et al. [4(
Cymbopogon citratus	Geranial, neral, limonene, geraniol, geranyl acetate	Gene expression assay	PureLink RNA Mini Kit			Hadjilouka, Mavrogiannis e al. [50]
Eugenia spp.	Eugenol, eugnyl acetate, caryophyllene	MIC, biofilm, DEM method	96-well microtiter plates	0.2 ± 0.02 µL/mL		Bermúdez-Capdevila, Cervantes-Huamán et al. [8
Eugenia caryophyllata		Disc diffusion method, MIC	Beef hot dogs	15.6–31.2 µL/mL, depending on the strain		Singh, Singh et al. [28]
Eugenia caryophyllata		MIC, MBC, biofilm, eDNS, cytotoxicity, qPCR	24-well culture plate,	1.5 µL/mL		Banerji, Mahamune et al [24]
Hibiscus surratensis L. calyce	β-Caryophyllene, menthol, methyl salicylate, camphor, germacrene D	Disc diffusion method, MIC, MBC	Broth	0.15 ± 0.05 µL/mL 25.26 ± 1.53 mm	0.083 ± 0.04 µL/mL	Akarca [4]

Source of Essential Oil	Major Compounds Investigated in the Study	Applied Method	Experimental Condition	MIC (µL/mL) or Inhibition Zone (mm)	MBC (µL/mL)	Reference
Laurus nobilis		Liposome-coated, MIC, MBC, SEM	Silver carp (*Hypophthalmicchthys molitrix*)	45 µL/mL	50 µL/mL	Aala, Ahmadi et al. [51]
Melaleuca alternifolia	Terpinen-4-ol, gamma-terpinene, α-terpinene, α-terpineol, terpinolene, α-pinene	Agar disc diffusion method, MIC, death–time curve, SEM	Ground beef, 96-well microplates	0.10 µL/mL 30 ± 8.8 mm	0.15 µL/mL	Silva, Figueiredo et al. [52]
Melissa officinalis	β-caryophyllene, *cis*-1,2-dihydroperillaldehyde, caryophyllene oxide, geranyl acetate, citronellal, β-citronellol, photocitral A, (*E*)-methyl geranate, β-linalool	Agar disc diffusion assay, MIC, time-kill curves determination, quorum sensing	Watermelon	0.5 µL/mL 38.4 ± 4.2–54.6 ± 1.3 mm, depending on the strain		Carvalho, Coimbra et al. [1]
Mentha piperita	Pulagone, isomenthone, piperitenone, menthone, piperitone	High-pressure processing, separately	Ayran (yoghurt)			Evrendilek and Balasubramania [53]
Moringa oleifera	Palmitic acid, phytol, ethyl palmitate	MIC, double dilution method, virulence gene activity, time-to-kill curve, LSCM analysis	Mozzarella cheese, cheddar cheese, parmesan cheese, camembert cheese	10 µL/mL		Cui, Li et al. [10]
Moringa oleifera	Palmitic acid, phytol, ethyl palmitate, hexadecanal	MIC, MBC, moringa–chitosan nanoparticles, FTIR, SEM, AFM, color and sensory evaluation, time-to-kill curve	Fresh hard cheese (cheddar)	10 µL/mL	10 µL/mL	Lin, Gu et al. [5]
Nigella sativa	Carvacrol, thymol, thymohydroquinone, thymoquinone, limonene, carvone, *p*-cymene, γ-terpinene	Standard disc diffusion technique	Am1 plate	28.2 ± 2.0–39.5 ± 1.1 mm, depending on the strain		Nair, Vasudevan et al. [35]
Origanum majorana	Terpinene-4-ol, γ-terpinene, β-phellandrene	MIC, biofilm, SEM, Box–Behnken experimental design	Plate, Falcon tubes	6.3 µL/mL		Vidács, Kereke et al. [19]
Origanum vulgare	Monoterpene, carvacrol, p-cymene, sesquiterpenes	Agar disc diffusion assay MIC, MBC, sensory evaluation	Raw minced meat	0.062–0.12 µL/mL, depending on the strain 10.3–27.3, depending on strain and concentration	0.062–0.12 µL/mL, depending on the strain	Pesavento, Calonico et al. [2]
Origanum vulgare	Carvacrol, *o*-cymene, thymol	Petri dish, confocal laser scanning, MIC,	GEN III microplates	2.50 µL/mL		Maggio, Rossi et al. [32]
Origanum	Carvacrol, linalool, p-cymene	Thermal inactivation by sous-vide processing, D and z-values	Atlantic salmon (*Salmo salar)*			Dogruyol, Mol et al. [54]
Origanum vulgare	Thymol, carvacrol	Inoculation of chicken fillets, sensory evaluation, changes in shelf-life study	Fresh chicken breast meat fillets			Khanjari, Karabagias et a [55]
Origanum vulgare	Carvacrol, p-cymene, caryophyllene, terpinene	Agar diffusion, MIC, EO microencapsulation, sensory analysis	Italian salami	3 µL/mL 0.8–38 mm, depending on the concentration		Gottardo, Bidusk et al. [31]

Source of Essential Oil	Major Compounds Investigated in the Study	Applied Method	Experimental Condition	MIC (µL/mL) or Inhibition Zone (mm)	MBC (µL/mL)	Reference
Origanum vulgare subsp. *hirtum*	α-thujene, p-cymene, gamma-terpinene, thymol, carvacrol	Spreading, sensory evaluation	Feta cheese			Govaris, Botsoglou et al [56]
Origanum vulgare L.	Carvacrol	Carvacrol encapsulation on chia mucilage nanoparticle (CMNP) and flaxseed mucilage nanoparticle (FMNP) Carvacrol was encapsulated in mucilage (chia and flaxseed) using the BIC, time-to-kill assay	96-well microplate			Cacciatore, Maders et al. [44
Phoenix dactylifera L.	3,4-dimethoxytoluene, 5,9-undecadien-2-one, 9-octadecenoic acid, 2,6-dimethoxytoluene	Inhibition zones, inhibition activity, agar well diffusion assay	Chicken meat	13 mm		Al-Zoreky and A Taher [57]
Picea excelsa	β-Pinene, α-pinene, limonene, camphene, delta-3-carene, β phellandrene, 1,8-cineole, traces of sabinene, α-terpineol, terpinen-4-ol	MIC, LBC, MBC	Broth, plate	0.15 ± 0.02–0.67 ± 0.26 µL/mL	2–6 µL/mL	Canillac and Mourey [58]
Picea excelsa	β-Pinene, α-pinene, limonene, camphene	MIC, MBC, simplified method, kinetic studies	Cheese	0.25–0.26 µL/mL	2–2.1 µL/mL	Canillac and Mourey [59]
Pimenta dioica		Disc diffusion method, MIC	Beef hot dogs	15.6–31.2 µL/mL, depending on the strain		Singh, Singh et al. [28]
Plectranthus amboinicus (Lour.) Spreng.	Thymol, p-cymene, β-myrcene, α-terpinolene	MIC, MBC, time-to-kill assay, bacterial anti-adhesion assay	Beef patties	2 µL/mL	4 µL/mL	Dutra da Silva, Bernardes et al [16]
Prangos ferulacea		Disc diffusion method, diameter of inhibition zone	Turkish Herby cheese	8–13 mm, depending on the strain		Sagun, Durmaz e al. [48]
Prunus armeniaca	Benzaldehyde, benzoic acid, mandelonitrile	Chitosan films, sensory evaluation	Spiced beef			Wang, Dong et a [60]
Rosmarinus officinalis	1,8-cineole, α-pinene, sesquiterpenes	Agar disc diffusion assay, MIC, MBC, sensory evaluation	Raw minced meat	5–30 µL/mL, depending on the strain 6–19.7 mm, depending on strain and concentration	5–30 µL/mL, depending on the strain	Pesavento, Calonico et al. [2
Rosmarinus officinalis		Disc diffusion method, MIC	Beef hot dogs	62.5–125.0 µL/mL		Singh, Singh et al. [28]
Syzygium aromaticum		Petri dish	Chicken frankfurters			Mytle, Anderson et al. [61]
Syzygium aromaticum		MIC, MBC, plate colony counting, time-to-kill analysis, TEM	PYG liquid medium	0.5 µL/mL	1 µL/mL	Cui, Zhang et al [3]
Salvia officinalis		Agar disc diffusion assay MIC, MBC, sensory evaluation	Raw minced meat	60 µL/mL 6–15.7 mm, depending on strain and concentration	60 µL/mL	Pesavento, Calonico et al. [2
Salvia officinalis		Disc diffusion method, MIC	Beef hot dogs	125.0–250.0 µL/mL		Singh, Singh et al. [28]

Source of Essential Oil	Major Compounds Investigated in the Study	Applied Method	Experimental Condition	MIC (µL/mL) or Inhibition Zone (mm)	MBC (µL/mL)	Reference
Salvia rosmarius		Liposome-coated, MIC, MBC, SEM	Silver carp (*Hypophthalmicchthys molitrix*)	5 µL/mL	10 µL/mL	Aala, Ahmadi et al. [51]
Salvia officinalis L.	β-Pinene, camphor, β-thujene, 1.8-cineole, α-humulene, endoborneol	Sous-vide cook-chill (SVCC), MIC	*Maronesa* male bovines	31.25 µL/mL		Moura-Alves, Gouveia et al. [6
Satureja horvatii	*p*-Cymene, thymol, thymol methyl ether, γ-terpinene, α-pinene, α-terpinene	MIC, MBC, MYC, modified micro-dilution technique, sensory evaluation	96-wells microplates, pork meat medium	0.57 ± 0.03 µL/mL	1.15 ± 0.01 µL/mL)	Bukvicki, Stojkovic et al. [6
Schinus terebinthifolius Raddi	Monoterpenes such as α-pinene, β-Pinene, myrcene, limonene, *D*-germacrene	Disc diffusion method, MIC, MBC, inhibition zone	Cheese	6.799–6.820 µL/mL 35.22 ± 0.79–40.86 ± 0.31 mm	6820–13.598 µL/mL	da Silva Dannenberg, Funck et al. [42]
Tetraastris catuaba	β-Caryophyllene, α-copaeno, α-himachalene, *iso*-sylvestrene, linalool butanoate, α-pinene, guaiene	Nanoemulsion, thermal analysis, stability test, TEM, biofilm assay, SEM	Microtiter plate, BHI agar			Silva, de Souza Arruda et al. [38
Trachyspermum ammi	Thymol, p-cymene, γ-terpinene, α-terpinene, α-thujene	Nanoemulsions, MIC, MBC	Turkey fillet preparation	8 µL/mL		Kazemeini, Azizian et al. [39
Thymbra capitata		MIC, MBC, WGS, antibiotic susceptibility test, *Thymbra capitata* evolution asay	Skimmed milk	0.15–0.30 µL/mL, depending on the strain	0.20–0.40 µL/mL, depending on the strain	Berdejo, Pagan et al. [64]
Thymus capitatus	Carvacrol, p-cymene	MIC, biofilm, DEM method	96-well microtiter plates	2.56 ± 0.17 µL/mL		Bermúdez-Capdevila, Cervantes-Huamán et al. [8
Thymus eriocalyx	Thymol, α-phellandrene, *cis*-sabinene hydroxide, 1,8-cineole, α-pinene	Disc diffusion method, MIC, bactericidal kinetics, TEM	BHI, MH	0.25 µL/mL 19–44 mm, depending on the concentration		Rasooli, Rezaei et al. [11]
Thymus x-porlock	Thymol, α-phellandrene, *cis*-sabinene hydroxide, 1,8-cineole, α-pinene	Disc diffusion method, MIC, bactericidal kinetics, TEM	BHI, MH	0.25 µL/mL/19–40 mm, depending on the concentration		Rasooli, Rezaei et al. [11]
Thymus vulgaris	Monoterpenes and sesquiterpenes, p-cymene, thymol	Agar disc diffusion assay MIC, MBC, sensory evaluation	Raw minced meat	0.25 µL/mL 11–33.5 mm, depending on the strain and concentration	0.25 µL/mL	Pesavento, Calonico et al. [2
Thymus vulgaris	p-Cymene, γ-terpinene, thymol, carvacrol, β-bisabolene	Spreading, sensory evaluation	Feta cheese			Govaris, Botsoglou et al [56]
Thymus vulgaris		Disc diffusion method, MIC	Beef hot dogs	7.8–15.6 µL/mL, depending on the strain		Singh, Singh et al. [28]
Thymus vulgaris	γ-Terpinene, thymol, p-cymene	MIC, biofilm, SEM, Box–Behnken experimental design		6.3 µL/mL		Vidács, Kerekes et al. [19]

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Thymus vulgaris L.	Carvacrol	Carvacrol encapsulation on chia mucilage nanoparticle (CMNP) and flaxseed mucilage nanoparticle (FMNP), carvacrol was encapsulated in mucilage (chia and flaxseed) using the BIC, time-to-kill assay	96-well microplate			Cacciatore, Maders et al. [44]
Thymus vulgaris L.	α-Pinene, p-cymene, thymol, linalool, γ-Terpinene	MIC	Cheese, microtiter plate	2.5 µL/mL		de Carvalho, de Souza et al. [33]
Thymus zygis		Disc diffusion method, vapor-phase antimicrobial activity determination, MIC, time-to-kill curves, motility assay, biofilm	Chicken juice, lettuce leaf model, ZHT-treated skim, milk, spinach	0.5 µL/mL 41.55 ± 2.63–55.04 ± 3.64 mm, depending on the strain		Coimbra, Carvalho et al. [4
Zataria multiflora Boiss	α-Pinene, p-cymene, α-terpinene, eucalyptol, α-terpineol	Sensory analysis, MBC, gene expression assay (RNS extraction, purification, RT-PCR)	Broth and minced rainbow trout	0.31–0.9 µL/mL, depending on the temperature	0.625–1.25 µL/mL, depending on temperature	Pilevar, Hossein et al. [23]
Ziziphora clinopodioides	Carvacrol, thymol, *p*-cymene, *γ*-terpinene	Chitosan–gelatin film, sensory evaluation	Minced rainbow trout			Kakaei and Shahbazi [65]

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