

Essential Oils in Food Safety and Shelf Life

Subjects: **Microbiology**

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The use of plant extracts (e.g., essential oils and their active compounds) represents an interesting alternative to chemical additives and preservatives applied to delay the alteration and oxidation of foods during their storage. Essential oils (EO) are nowadays considered valuable sources of food preservatives as they provide a healthier alternative to synthetic chemicals while serving the same purpose without affecting food quality parameters. The natural antimicrobial molecules found in medicinal plants represent a possible solution against drug-resistant bacteria, which represent a global health problem, especially for foodborne infections.

natural antimicrobials

food preservatives

biocontrol agents

food safety

1. Introduction

Medicinal plants are a valuable source of new antibacterial, antifungal, and antioxidant compounds due to the large biological and structural diversity of their constituents [1][2][3]. Botanical species and their derivatives, including essential oils (EOs), extracts, and bioactive compounds (BACs), have been discovered to be key contributors to the pharmaceutical, agricultural, and food industries.

Traditionally, medicinal plants are used to treat diseases, but they are also well suited for the food industry as natural antimicrobial preservatives [4][5][6][7]. Chemicals (benzoates, propionates, sorbates, nitrates, nitrites, etc.) commonly used as food additives are reported to be the cause of health problems such as allergies, asthma, liver damage, and cancer [8][9][10][11][12]. These concerns reinforce the interest in using natural antimicrobials in food formulations.

Alcohols, ethers or oxides, aldehydes, ketones, esters, amines, amides, phenols, heterocycles, terpenes (an oxygenated derivative of terpenoids), and polyphenols represent the chemical classes to which belong the constituents of EOs and plant extracts. Some terpene compounds identified in EOs are limonene, β -caryophyllene, β -pinene, α -pinene, α -terpinene, sabinene, β -myrcene, γ -terpinene, cinnamyl alcohol, δ -3-carene, and *p*-cymene. Limonene is mainly distributed in citrus EOs: orange [13], grapefruit [14], and pomelo [15]. Thymol, carvacrol, and eugenol, deriving from *Thymus* [16], *Origanum* [17], and *Ocimum* [18], represent the main phenolic terpenes associated with EOs obtained from medical and aromatic plants (Figure 1).

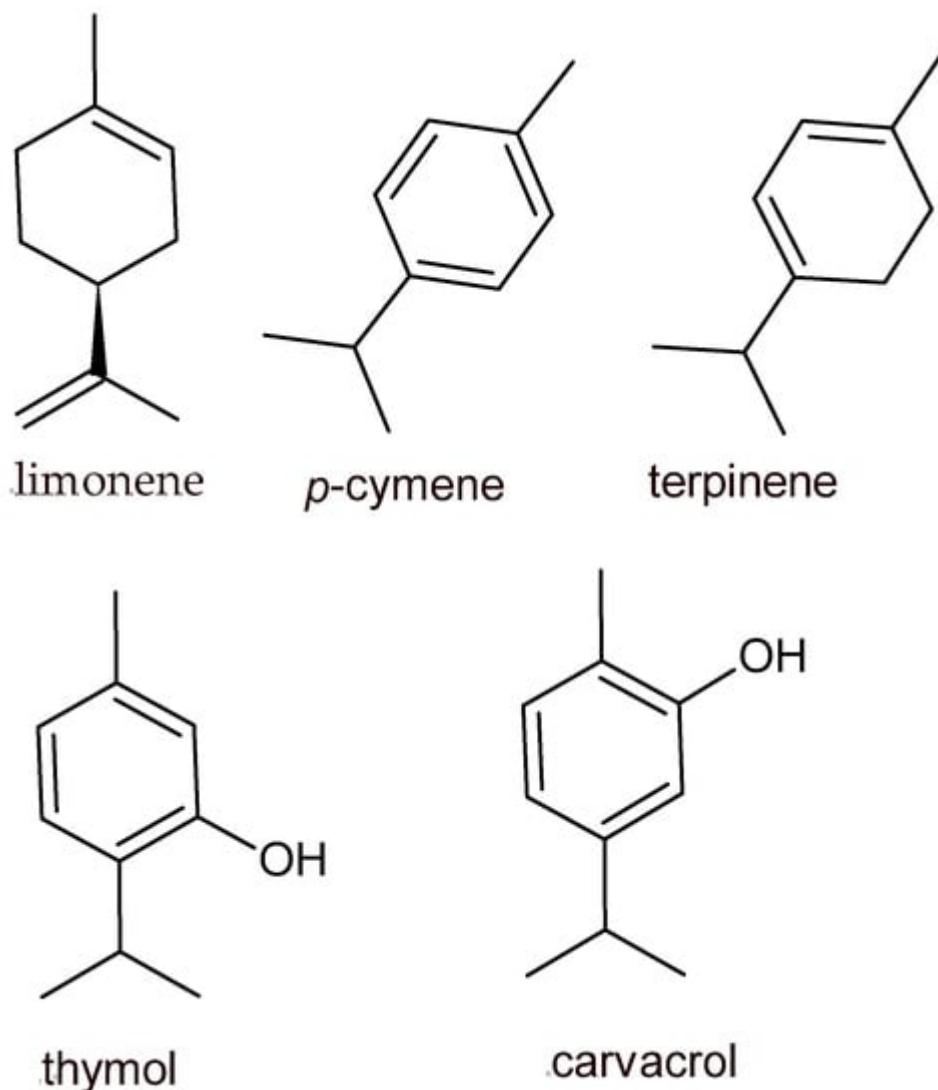


Figure 1. Chemical structures of the most applicable EO components in foods.

A large variety of these compounds are recognized to have strong and effective antimicrobial activity [19][20][21][22]. EOs from the *Lamiaceae* family aromatic plants (i.e., oregano and thyme) and their constituents (like carvacrol and thymol) have been described in the literature as potential preservatives with significant effects on food shelf-life [23][24][25][26][27][28][29].

The lipophilic nature of EOs is directly related to their antibacterial activity. EOs destabilize the membrane potential of bacterial cells by disrupting the permeability of the plasma membrane [30]. Degradation of membrane lipid fractions by the EO component thymol leads to destabilization of membrane permeability [31]. EOs affect quorum sensing signaling and can inhibit cell-to-cell communication, e.g., biofilm formation in bacterial cells [32]. Bouyahya et al. (2019) [33] reported the antimicrobial activity of *Origanum compactum* EO in the context of disrupting cell membrane stability and integrity and increasing membrane permeability, leading to leakage of cellular material (DNA and RNA). Moreover, the effect of EOs on inhibiting biofilm formation in bacterial communities triggers the breakdown of their sensing communication [34].

Several EOs, including those of thyme, marjoram, oregano, basil, ginger, lemongrass, and clove, were shown to be highly effective in inhibiting spoilage bacteria in meat [35], dairy products [36][37][38][39][40], and beverages [41]. It is generally accepted that plants are an important source of antimicrobial metabolites, including flavonoids, phenolic compounds, tannins, terpenoids, saponins, and alkaloids. Other than being endowed with antibacterial and antioxidant properties, BACs may help to enhance the food's sensory and organoleptic properties and its acceptability, including its shelf life [42].

Many EOs and plant extracts have the GRAS (generally recognized as safe) status obtained by the Food and Drug Administration (FDA) and the European Food Safety Authority (EFSA), (Figure 2). Therefore, they can be applied in the food industry, meeting in this way the consumer requirement for natural food preservatives [4].

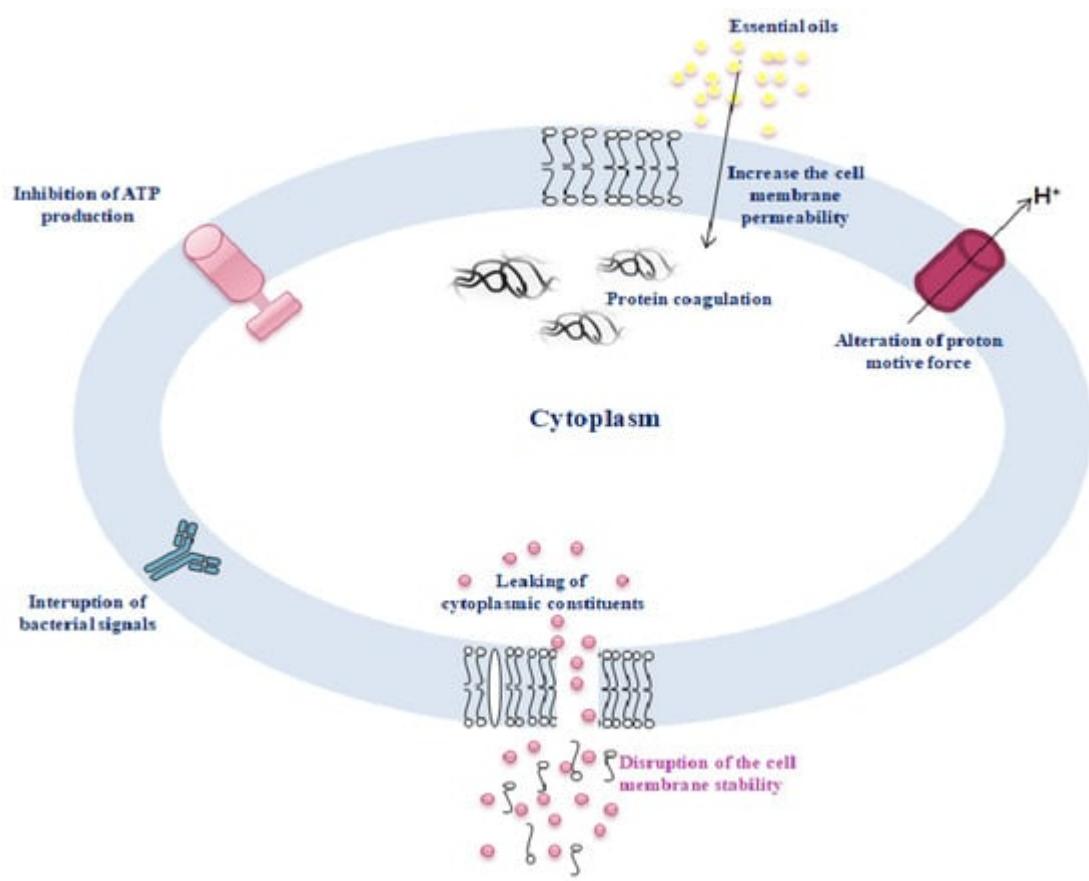


Figure 2. The antimicrobial mechanisms of action of essential oils.

However, despite EOs positive effect, their practical use as effective antimicrobial agents in the food sector is still a challenge due to their volatile characteristics, hydrophobicity, and low stability. Nowadays, new technologies and delivery strategies such as nanoencapsulation, encapsulation in active packaging, or polymer-based coatings have efficiently addressed these issues and improved the efficacy and gradual release of EOs [43]. Eventually, EO application can be limited by the sensory impact it imparts when applied at the required active concentrations. Therefore, a possible solution to overcome this aspect is their combination with other natural strategies.

Finally, LABs can produce different classes of antimicrobial peptides called bacteriocins. These molecules are antimicrobial peptides produced by specific bacterial species to protect themselves from other bacteria by inhibiting or killing them without harming themselves. The bacteriocins action mechanism is mainly associated with the permeabilization of the cell membrane, but it is reported that they can also act on protein metabolism, DNA and RNA, and quorum sensing [44][45]. The application of bacteriocins in the food sector has increased in recent years. They have attracted noticeable attention for their potential application as natural and safe food preservatives since they are easily digestible in the gastrointestinal tract. Nisin represents the most known and studied bacteriocin; it is produced by *Lactococcus lactis*, and its use was authorized by the Food Drug and Administration (FDA) in more than 50 countries [46]. Three ways of bacteriocin applications in foods are reported: inoculation of bacteriocin-producing LABs directly in the food products (starter or protective cultures), application as a food additive, and incorporation in food packaging of the purified or partially purified bacteriocin.

2. Use of Essential Oils and Biocontrol in Minimally Processed Fruit and Vegetables

Over recent years, the market for minimally processed fruits and vegetables has increased steadily. Their popularity is due to their convenience, reduced waste production, and high nutritional content. In fact, these foods represent a source of valuable compounds such as vitamins, minerals, fibers, and antioxidants and may help prevent chronic diseases. Nevertheless, fruits and vegetables are a suitable matrix for microbial proliferation [47][48]. In fact, these foods can be easily spoiled due to the high nutrient and water content and the numerous processing steps that fresh produce undergoes (i.e., peeling, cutting, or slicing), which can promote microbial proliferation by releasing nutrients and transporting the microbial population present on the surfaces of vegetables and fruits into the cut ones [49][50].

Currently, the use of chemical sanitizers in the washing step, as well as the maintenance of refrigerated temperatures during processing and storage, are the only stages capable of reducing and controlling the microbial population in minimally processed vegetables [51]. However, the use of chemical sanitizers, especially chlorine-based ones, is not always effective, can result in the production of toxic molecules, and is not well accepted by consumers [52]. Because of these drawbacks, researchers are looking to alternative sanitizers (i.e., ozone, hydrogen peroxide, and peroxyacetic acid), physical treatments (UV light, ultrasound, and gamma rays), and natural food additives. As a result of their antibacterial and antioxidant properties as well as their GRAS classification, EOs have the potential to extend the shelf life of several foods, including fresh-cut fruits and vegetables. EOs can be used in various stages of food and vegetable processing, including the washing step, directly on the product, or in the packaging. In fact, the incorporation of EOs and their components in active food packaging is of high relevance. Active packaging is a solid matrix from which EOs are gradually released during food storage. Generally, various methods, including direct incorporation, coating, and surface modification, are applied to incorporate EOs into active packaging [53]. Antimicrobial active packaging is designed to enhance product safety and shelf life by inhibiting or reducing microbial growth in packaged foods [54][55][56]. In this context, it

is fundamental to use suitable solvents or polymeric carriers, as EOs are always used in diluted form, which should be food-grade and not interfere with the antimicrobial and antioxidant activities of EO constituents [57].

Other researchers found that dipping sliced apples in a solution containing dissolved hexanal (250 ppm) or the combinations hexanal/2-(E)-hexenal and citral/2-(E)-hexenal (125 ppm of each compound) increased quality parameters and extended the shelf life by reducing the proliferation of naturally occurring yeast [47]. In addition, sliced apples treated with hexanal/2-(E)-hexenal, and citral better retained both color and textural properties. The combination with an active, modified atmosphere (7% O₂ and 0% CO₂) enhanced the safety and shelf life of minimally processed apples washed with a solution containing a mixture of hexanal/2-(E)-hexenal. Other researchers discovered that active packaging containing thymol and eugenol inhibited the growth of mesophilic bacteria and yeasts and consequently extended the shelf life of table grapes stored in a modified atmosphere (MAP) compared to the control group [55].

The use of EOs has great potential, even in minimally processed vegetables. In fact, herbal EOs and their constituents were studied as alternative natural disinfectants for reducing the presence of spoiling and pathogenic bacteria. Due to the strong antimicrobial activity in vitro of oregano (*Origanum vulgare*) and thyme (*Thymus vulgaris*) EOs and their main constituents (carvacrol and thymol), these compounds are the most suitable to be used in minimally processed vegetables [30][44][58][59][60].

For example, the inclusion of oregano and thyme EOs at a concentration of 250 ppm in the washing solution of minimally processed lamb's lettuce resulted in a product shelf life comparable to the one obtained by using chlorine (120 ppm) in the washing step [59]. In fact, the use of EOs resulted in a preliminary decrease in the microbial population. In addition, the color and turgidity of the lettuce were not affected throughout the storage period while the sensorial properties were not negatively affected [59].

When used in the washing solution, oregano oil (*Origanum onites*) was able to inactivate *Salmonella typhimurium* inoculated on iceberg lettuce. The effectiveness of oregano EO treatment (75 ppm) was comparable to that of chlorine treatment at 50 ppm [61].

When oregano EO was applied to ready to eat lettuce and carrots, the initial decontamination effect was comparable to that of chlorine [62]. Furthermore, oregano EO did not negatively affect the color, texture, or water activity of the samples. However, sensory acceptance of the product treated with oregano EO was only observed for carrots. Volatile antimicrobial compounds such as borneol, carvacrol, cinnamaldehyde, eugenol, menthol, thymol, and vanillin were able to inhibit the growth of *Bacillus cereus* inoculated in carrot juice. Direct application of EOs in the food system has some drawbacks due to their strong odor, chemical reactivity, hydrophobicity, low solubility, and potential negative interaction with the food matrix, leading to alteration of organoleptic properties. Several technological approaches have been tested for EO delivery in food. EO encapsulation is a novel and advanced delivery system. It provides enhanced antimicrobial efficacy as well as control over the release of EO flavors into the food system [63].

3. Application of Essential Oils as Meat and Dairy Preservatives

3.1. Antimicrobials as Meat Preservatives

Fresh meat and fresh meat produce are extremely perishable and prone to oxidative and microbial spoilage. These products are easily subject to oxidation, which reduces nutritional value, affecting lipids, proteins, myoglobin (pigments), texture, and flavor [64]. Plant extracts are a natural source of compounds with antioxidants and antimicrobial activity that can be used as an alternative to synthetic ones. EOs and their constituents have been successfully used in meat and meat products to prevent oxidation, degradation, and microbial proliferation [65][66][67]. Promising results can be achieved with EOs from the *Lamiaceae* family, including oregano, thyme, sage, and rosemary, which are commonly used as flavoring agents and prevent oxidative degradation. The antioxidant activity of EOs is well documented and can be attributed to phenolic compounds including eugenol, carvacrol, and thymol, which act as hydrogen donors and scavengers of free radicals [68][69].

Fasseas and colleagues (2007) [70] measured the antioxidant activity of pork and beef meats (raw and cooked) treated with sage and oregano EOs over a 12 day storage period at 4 °C. Compared to controls without added EOs, the EO-treated meat (raw and cooked) had higher TBA (thiobarbituric acid) scores and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity.

Meat contaminated with spoilage microorganisms shows color changes, the development of uncharacteristic and undesirable odors and tastes, and the formation of superficial slime [71][72]. Bacteria belonging to the genera *Pseudomonas*, *Acinetobacter*, *Staphylococcus*, *Brochothrix*, *Moraxella*, *Micrococcus*, and *Flavobacterium*, as well as LAB and genera belonging to the *Enterobacteriaceae* family, are reported to be involved in the spoilage of meat and meat derivatives [72][73].

Cell loads of foodborne pathogen *L. monocytogenes* deliberately inoculated on ham (3 log CFU/g) were decreased by 10 and 19% after supplementation of *Cinnamon cassia* and oregano EOs, respectively [74]. *Cinnamon cassia* EO was also effective in promoting the shelf life of ground lamb meat throughout refrigerated storage at 4 °C. Compared to control samples, supplementation of *Cinnamon cassia* EO (0.5%) reduced from the 4th to the 16th day the lactic acid bacteria spoilage population up to 1.9 log CFU/g and *Enterobacteriaceae* up to 1.1 CFU/g [75]. A reduced microbial spoilage population was also observed in minced meat with rosemary, thyme, and oregano EOs at concentrations between 1 and 1.5% (v/w). Compared to the control group, treated samples showed reduced loads of LAB, molds, and yeast [76]. Thyme EOs tested at two different concentrations (0.02 and 0.05 w/w) inhibited the growth of coliform in chicken hamburgers [77].

The use of EOs as antioxidants and preservatives for meat is dependent on the bioactivity and stability of their components. Most EO constituents lose physical stability when they interact with meat constituents [78], probably due to the binding ability of fats and proteins in meat to volatiles in EOs [78]. When encapsulated EOs are added to edible coatings, plastic films, or meat during marinating, their stability increases significantly. The possibility of

using active packaging supplemented with natural antioxidants and antimicrobials has recently been investigated [79][80].

The stability, antioxidant, and antimicrobial properties of rosemary EO can be enforced by encapsulation with nanogel. Compared to samples treated with free rosemary EO, rosemary EO encapsulated in chitosan-benzoic acid-based nanogels showed greater efficacy in reducing the cell load of *Salmonella typhimurium* inoculated on beef chops during storage at 4 °C [63]. Chitosan-based coatings also improved the performance of oregano EO in reducing lipid oxidation in dry fermented sausages in comparison to control samples after 7 months of storage [81].

LABs can be added for two different purposes: as starting materials for producing fermented meat products or as a bioprotective culture used only to compete with naturally occurring microflora and produce antimicrobial peptides [82].

The constant release of antimicrobial peptides from the packaging to the product surface could help to maintain the bacteriocin concentration at the optimal level to carry out their bioprotective effect [83]. In various studies, films containing the bacteriocin nisin have been successfully used to preserve meat products [82]. Pullulan film containing Nisin Z, which was used for the packaging of refrigerated vacuum-packed raw beef and deli ham, strongly inhibited various foodborne pathogens, including *Salmonella* spp., *S. aureus*, *L. monocytogenes*, and *E. coli* [84]. Furthermore, the efficacy of this nisin/pullulan film was improved by the addition of lauric alginate [84]. Reduced growth of *S. aureus* on chilled sliced beef was observed when packaged with an alginate-based palmitoylated film supplemented with nisin [85]. Nisin-internalized cellulose envelopes demonstrated anti-*Listeria* properties on chilled vacuum-packed frankfurters (sausages) [86][87] and vacuum-packed hot dogs wrapped with a plastic film enriched with nisin. A combination of nisin and Nisaplin® (a commercial product from DuPont) adsorbed in cellulose-based packaging paper was shown to inhibit *S. aureus*, *L. innocua*, and LAB on cooked ham [88].

3.2. Essential Oils as Dairy Preservatives

Plants and spices have been used in cheese production since ancient times, often linked to local traditions. Traditionally, spices and herbs or their extracts were rubbed directly on the cheese. In fact, herbs and spices can be applied as antioxidant, antimicrobial, flavoring, enriching, and functionalizing ingredients, which may improve the appearance and appeal of the product [89].

Although many dairy products undergo heat treatment during their manufacture, they are perishable and can be easily contaminated by microorganisms, resulting in food spoilage, consumer health risks, and a shortened product shelf life. Furthermore, as they are rich in lipids, oxidative processes can lead to a loss of flavor, nutrients, and color, the development of off-flavors, and the accumulation of compounds that may be of concern to human health [90]. For that reason, there is growing interest in the application of plants, extracts, and EOs as natural preservatives in the dairy sector as an alternative to synthetic preservatives [91]. The antimicrobial activity of natural products is commonly assessed against the main pathogens and spoilage bacteria commonly found in the dairy

sector, such as *L. monocytogenes*, *S. aureus*, *E. coli*, *Salmonella* spp., and *Pseudomonas* spp., as well as yeasts and molds, such as *Penicillium* and *Aspergillus* [89].

Natural products have also been proven in additional types of dairy products, including milk. Jemaa et al., 2017 [92] showed that EO of *Thymus capitatus* ameliorated pasteurization effectiveness in maintaining raw milk quality. Incorporation of EOs from basil, peppermint, and zataria in the formulation of probiotic yoghurt increased the inhibition of *E. coli* and *L. monocytogenes* compared to yoghurt without EOs. However, only the EOs of basil and peppermint showed good antioxidant and antiradical activity along with good sensory acceptability [93]. The application of *Echinophora platyloba* EO and lycopene was effective as natural preservatives for dairy products with high fat content, such as butter and cream. Pasteurized cream treated with a mixture of EO and lycopene (0.5% and 50 ppm, respectively) showed improved shelf life (both from a microbiological and chemical point of view) compared to control samples. Sensory evaluation results showed that all the samples have satisfactory overall acceptability, although the greatest sensorial features were detected in creams prepared with a combination of low concentrations of *Echinophora* EO and lycopene (0.1% and 20 ppm, respectively) [94]. Other authors extended the shelf life of butter using thyme and cumin EOs [95], while Ozkan et al. (2007) [96] explored the potential of *Satureja cilicica* EO as a natural flavoring and antioxidant in the same dairy product.

Although many studies highlight the beneficial properties of herbal preservatives, other papers report that dairy fats, carbohydrates, or proteins may interact with them and reduce their functional properties. Consequently, higher amounts of EOs are necessary to attain the desired effect, with subsequent drawbacks related to sensorial impacts. To overcome this aspect, different approaches have been investigated.

Nisin is permitted as a food additive for processed cheese by the FAO/WHO Codex Committee at a concentration of 12.5 mg/kg product, while the US FDA is permitted to use up to 250 mg/kg [97]. It is generally known that the effectiveness of nisin is dependent on the dairy products considered. In particular, neutral-pH dairy products made from whole milk are not suitable for nisin use [98].

4. Conclusions

Essential oils and bioactive compounds can be applied as food preservatives due to their antimicrobial and antioxidant activities, which prevent spoiling processes and guarantee food safety. On the other hand, an additional strategy to increase the safety and shelf life of various types of foods, including minimally processed fruits and vegetables, vegetable beverages, meat, and dairy products, can be represented by the use of protective cultures, especially lactic acid bacteria, which are able to produce antimicrobial compounds such as bacteriocins in addition to organic acids and hydrogen peroxide. Recently, innovative new technologies and delivery strategies such as nanoencapsulation or polymer-based coatings have improved the efficacy and allowed the controlled release of natural antimicrobials. In addition, the use of EOs and bioactive plant molecules in combination with bioprotective cultures or bacteriocins can exert an additive or synergistic effect and reduce the applied concentration of EO. The combination of these antimicrobial agents represents an interesting strategy to increase the shelf life and safety of food due to their antimicrobial and antioxidant properties. The synergistic combinations of EOs and LAB

metabolites such as bacteriocins or nisins allow the exploration of promising ways to overcome both the narrow range of antimicrobial action and the unpleasant sensory properties of foods.

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