



Essential Oils against Fruit Spoilage Fungi

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Abstract

Essential oils (EOs) are natural, low molecular weight secondary metabolites from aromatic plants. They are fat-soluble and soluble liquids in organic solvents, usually with lower density than water. EOs are widely known for their taste, aroma, and antimicrobial activity. Its volatile nature, attributed to secondary metabolites, has antioxidant and antimicrobial potential, representing approximately 90% of the composition. Other non-volatile components (acids, sterols, waxes), present in smaller amounts, can also significantly influence its bioactivity. Recent studies have shown antimicrobial, insecticide, or repellent action on pests using EOs. The use of essential oils proves to be a promising alternative to traditional artificial antifungals since they have effectiveness in the control of fungi related to food deterioration, low toxicity, act as preservatives in food. In this chapter, we will address the main fungi and yeasts responsible for the deterioration of fruits, the main essential oils used in food, and the application of essential oils against fruit-deteriorating fungi.

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5.1 Introduction

Essential oils (EOs) are natural, low-molecular-weight secondary metabolites obtained from aromatic plants. They are aromatic, fat-soluble, and soluble liquids in organic solvents, usually with lower density than water (da Silva et al. 2021; Speranza and Corbo 2010). These substances can be extracted from parts such as leaves, branches, stems, seeds, and leaves (Arasu et al. 2019; Maia et al. 2015; Seshadri et al. 2020a). The main characteristics of these compounds are volatility and low solubility in water (Maia et al. 2015). In addition, some factors, such as genetic factors, climatic conditions, plant maturation stage, processing, concentration, and storage, can affect the composition and, consequently, the final quality of the product obtained (Barbosa 2010; Maia et al. 2015).

With the advance of globalization, there has been a change in the eating habits of populations, while the use of food additives and pesticides in food has intensified. However, consumers have shown concern about the possible harmful effects of these substances, and the uncontrolled use of these can provide the fungal resistance (e.g., Camele et al. 2012; de Corato et al. 2010; Seshadri et al. 2020a; da Silva et al. 2021). Thus, the use of less harmful antimicrobials is effective for the control of fungi and the promotion of food safety (Camele et al. 2012; da Silva et al. 2021). Therefore, these compounds have been widely used to increase the shelf life of processed and fresh foods and maintain sensorial characteristics due to their antifungal activity (Seshadri et al. 2020a; Speranza and Corbo 2010).

Furthermore, another critical factor is the control of post-harvest fungal deterioration. Fruits are highly perishable, and their properties corroborate that there are more significant losses and waste, in addition to their structure providing greater susceptibility to deterioration by fungi (de Botelho et al. 2022). Moreover, fruits have high water and moisture activity and various nutrients, which favor increased deterioration reactions (de Corato et al. 2010). Thus, care must be taken during cultivation, harvesting, and post-harvest to ensure the quality of the final product.

The use of essential oils proves to be a promising alternative to traditional artificial antifungals since they have effectiveness in the control of fungi related to food deterioration, low toxicity, act as preservatives in food, and are generally recognized as safe (GRAS) (Arasu et al. 2019; da Silva et al. 2021; Simas et al. 2017). Furthermore, several studies show the application of essential oils due to antimicrobial activity. The essential oils include clove oil, oregano, basil, thyme, rosemary, ginger, and cinnamon (Cui et al. 2020; Reis et al. 2020; Xu et al. 2022).

In this chapter, we will address the main fungi and yeasts responsible for the deterioration of fruits, the main essential oils used in food, and the application of essential oils against fruit-deteriorating fungi.

5.2 Food Spoilage Fungi

According to de Botelho et al. (2022), fungi are eukaryotic organisms described as heterotrophic, unicellular, and multicellular organisms. The difference between unicellular and multicellular is hyphae formation, a filamentous structure that forms the mycelium. Fungi produce spores that are related to the propagation of the species. These groups of microorganisms could be damaging to human health and cause mycoses and infectious diseases (de Botelho et al. 2022). In addition, there are fungi that cause food spoilage. Food, after being contaminated, can favor the multiplication of undesirable spoilage microorganisms causing damage to the product. Consequently, it causes economic losses and risks to the health of the consumer when mycotoxin-producing fungi occur (Bernardi et al. 2019; Santana Oliveira et al. 2019).

5.2.1 Contamination by Molds and Yeasts that Leads to Spoilage

Fruits and vegetables are essential sources of nutrients for humans; they are good sources of vitamins, minerals, phytochemicals, dietary fiber, and antioxidants. However, about 45% of these foods are wasted annually due to spoilage caused by contaminated environments, inadequate harvesting conditions, unsafe handling and storage processes, and incorrect display methods (Alegbeleye et al. 2022; Saleh and Al-Thani 2019). Also, according to the Food and Agriculture Organization (FAO), about 15 to 50% of the total amount of fruits and vegetables worldwide were lost during the post-harvest process in the year 2011 (Gunny et al. 2021), and 1.3 billion tons of food are lost every year due to spoilage (Kaczmarek et al. 2019).

Food spoilage has numerous socioeconomic implications, as it is related to food scarcity, waste, and hunger in some parts of the world, water stress, unnecessary loss of biodiversity, and increased emissions of greenhouse gases such as carbon dioxide and methane produced by spoilage, which contributes to the climate crisis (Alegbeleye et al. 2022).

Although there are microorganisms naturally present on the surface of fruits and vegetables, which do not cause disease and yet can act as a natural biological barrier to contamination by pathogens or spoilage agents, molds and yeasts, in general, are the leading cause of spoilage of raw fruits (Kaczmarek et al. 2019; Saleh and Al-Thani 2019). Molds can benefit human activities, as they are producers of enzymes, organic acids, and antibiotics and can be used in food production, for example. However, they can spoil various foods, from raw materials to finished products, causing changes in taste, odor, and spoilage. These degradative changes in food products due to microorganism contamination are responsible for losing food quality (Pandey et al. 2021; Quéro et al. 2019). The fermented aroma, sour taste, slimy surface, moisture, soft rot, discoloration, and even visible microbial growth characterize fungal contamination in many types of fruits (Kaczmarek et al. 2019).

Also, they are a significant threat to human health and safety, resulting in food-borne diseases due to the production of mycotoxins that can cause food poisoning and serious health problems to consumers, just as severe economic losses

(Alegbeleye et al. 2022; Atif et al. 2020a; Cai et al. 2022; Pandey et al. 2022). Furthermore, contamination by microorganisms and oxidation triggered by reactive oxygen species (ROS) are generally responsible for the spoilage of fresh vegetable foods and fruits during processing, transportation, and storage (Pandey et al. 2021).

The richness of nutrients, sugar, carbohydrates, free amino acids, and the high water activity make them suitable for microbial growth and survival. However, bacterial spoilage in this food category is limited due to their low pH since bacteria typically grow at neutral pH. Thus, spoilage of these foods is usually associated with aciduric bacteria, molds, and yeasts (Alegbeleye et al. 2022; Roumani et al. 2022).

As the physiological ripening process of fruits and vegetables progresses toward senescence and possible spoilage, metabolic and developmental activity is maintained. During this process, various physiological and compositional changes occur that make fresh fruits and vegetables a suitable substrate for the proliferation of microorganisms. However, the chances of microbial spoilage becoming established depend on certain factors, such as surface morphology and topography, exudates from the plant surface, stage of development, and post-harvest handling. As soon as foods are removed from their natural supply of nutrients, their quality begins to decline. Therefore, the period of ripening and senescence is the most susceptible to spoilage (Alegbeleye et al. 2022).

Spoilage microorganisms can be introduced into fruits and vegetables under different circumstances, in the farm, post-harvest handling, and processing, all of which can predispose these foods to spoilage. In addition, plants are usually grown in unprotected natural environments, where soils are teeming with life, making contamination possible (Alegbeleye et al. 2022).

Many soil microorganisms can be transferred to fruit or vegetable surfaces during food production or processing. Soil particles, dust, air, and water used for irrigation are common vectors and can carry fungal mycelium, spores, and live bacterial cells. Microorganisms can grow on fresh produce and have developed sophisticated biochemical mechanisms to break down food components, which provide them with the energy sources needed for growth (Kaczmarek et al. 2019).

Microbial contamination of fruits and vegetables can come from various sources, including the farm environment, post-harvest handling, raw materials, and contact with processing equipment. In addition, food contamination can occur after harvest and processing. Several examples occur, such as significantly higher bacterial counts after processing, such as shredding, cutting, slicing, rinsing, and packaging, as debris and microorganisms can accumulate on the equipment that performs these operations. If effective hygiene practices are not followed during fresh produce processing, new sources of contamination can be introduced (Kaczmarek et al. 2019; Seshadri et al. 2020a).

Furthermore, it is worth noting that specific post-harvest practices can promote the establishment of existing microorganisms or introduce new contamination (Alegbeleye et al. 2022). Thus, misbehaving practices in fruit handling, such as packing in plastic cages, stacking cages that cause pressure on the fruit leading to mechanical damage, high temperature during marketing, and pecking or biting by insects and wild animals that damage the outer cover of the products. This can cause

damage such as wounds, abrasion, hematomas, perforations, cracks, breaks, or compression of the cellular and tissue structure that accelerate the contamination process and this favors the growth of microorganisms, such as fungi, prematurely. In addition, when the fruit is affected by molds and the fungal hyphae spread in the intercellular spaces and inside the parenchyma cells, they secrete enzymes that cause tissue maceration, eventually leading to fruit compromise (Alegbeleye et al. 2022; Moustafa 2018).

Applying good agricultural practices could reduce food loss from spoilage at all stages of vegetable production. The Federal Drug Administration (FDA, 2008) has a guide to minimize microbial food safety hazards for fresh vegetables (Guide to Minimize Microbial Food Safety Hazards of Fresh-cut Fruits and Vegetables) that recommends good practices for food producers, packers, and transporters to reduce the common microbiological hazards often associated with their operations (Kaczmarek et al. 2019).

It is also worth noting that several sources of contamination precede the harvest, such as seeds, agricultural water, soil, manure, biological alterations, livestock/wildlife, and humans. Contamination at this stage is crucial because it can determine the establishment of pathogens and spoilage within plant product tissues, making them impossible to remove by sanitizers or other decontamination strategies (Alegbeleye et al. 2022).

5.2.2 Main Molds and Yeasts that Spoilage Fruits

Plant-derived foods are often subject to spoilage by molds and yeasts, and contamination can occur from various sources described above (Table 5.1).

Members of the phylum *Ascomycota*, *Basidiomycota*, and *Zygomycota* are some of the most prevalent fungi. In addition, species of *Penicillium*, *Alternaria*, *Botrytis*, *Fusarium*, *Cladosporium*, *Phoma*, *Trichoderma*, *Lasiodiplodia*, *Aspergillus*, *Alternaria*, *Rhizopus*, *Aureobasidium*, and *Colletotrichum* are often responsible for spoilage of fresh produce and production of mycotoxins that impact product quality, can cause health risks to consumers, and socioeconomic losses (Alegbeleye et al. 2022; Kaczmarek et al. 2019). So, *Alternaria*, *Aspergillus*, *Penicillium*, and *Fusarium* are the main genera of spoilage fungi present in the postharvest stage (Atif et al. 2020a; Sanzani et al. 2016; Seshadri et al. 2020a), in which they secrete

Table 5.1 Fruit spoilage fungi and their effects

Fungi/Yeasts	Characteristics	Contaminated fruit	Reference
Genus <i>Alternaria</i>	Tenuazonic acid	Wide range of fruits, tomatoes	Moss (2008)
Genus <i>Aspergillus</i>	Black mold	Grapes, apricots	Alegbeleye et al. (2022)
Genus <i>Fusarium</i>	Fruit rot and crown rot	Banana	Alghuthaymi et al. (2020)
Genus <i>Penicillium</i>	Produced toxic patulin, blue and green rot of citrus	Pears and apples, citrus fruit	Neri et al. (2010)

a broad spectrum of enzymes such as amylases, cellulases, and pectinases that cause softening of plant tissues (Alegbeleye et al. 2022).

5.2.2.1 *Alternaria*

Alternaria is a fungal genus that has saprobic, endophytic, and pathogenic species and includes over 250 species. This group is related with a large range of serious plant pathogens that cause losses in large agricultural crops (de Botelho et al. 2022; de Lima et al. 2016).

One of the important species in this genus is *Alternaria alternata* that is a prevalent phytopathogen, causing serious foodborne spoilage and producing mycotoxins (Wang et al. 2019). Black spot caused by *Alternaria alternata* is associated to significant losses during storage and transportation of fruits (Guo et al. 2022). *Alternaria alternata* species are primarily responsible for causing brown spots on citrus fruits, also occur in a saprotrophic form on harvested fruits and vegetables, and can produce mycotoxins that exert poisonous effects after human consumption (Sanzani et al. 2016). The most important and frequent *Alternaria* mycotoxins are alternariol (AOH), and alternariol monomethyl ether (AME). The mycotoxins occur in derived from cereals, fruits, and vegetables, contaminating food and feedstuffs and could cause genotoxicity, cytotoxicity, mutagenicity, and carcinogenic (Wang et al. 2019).

5.2.2.2 *Aspergillus*

According to de Botelho et al. (2022), *Aspergillus* is an anamorph genus belonging to the phylum Ascomycota. There are more than 250 species of saprophytic mold in this genus, and they present asexual spores called aspergillum. *Aspergillus* can occur in different types of environments as they adapt to various growing conditions such as temperature from 6 to 55 °C and relatively low humidity. In food, they are frequently found in spoiled products, such as grains, cereals, vegetables, fruits, juices, and cakes (de Botelho et al. 2022).

Some species of the genus *Aspergillus* can affect entire plant organs and seeds and produce mycotoxins due to their secondary metabolism. Infection by these fungi usually occurs in the field, where the mycelium is confined to specific tissues (seed or fruit) (Sanzani et al. 2016).

One of the main species of this genus is *Aspergillus flavus*. This species is a facultative parasite that naturally occurs as a saprophytic soil fungus and contaminates various food crops such as cottonseed, maize, peanuts, and tree nuts (de Botelho et al. 2022; Chiotta et al. 2020; Tian et al. 2022). However, it is important to mention that *A. flavus* produces aflatoxin, which is a carcinogenic and mutagenic secondary metabolite and affects animal and human (Tian et al. 2022). For control *A. flavus* and aflatoxin contamination is important to avoid the growth of spores and mycelia and the inactivation of aflatoxins. Usually, to achieve these results it is necessary to use substances such as synthetic fungicides, ozone fumigation, irradiation, cooking processes, and controlled environmental conditions. However, many of these strategies are difficult, expensive, and inefficient. Then, considering this and the consumer's interest in ingesting products as natural as possible, essential oils could be a good option (Tian et al. 2022; de Botelho et al. 2022).

5.2.2.3 *Penicillium*

Penicillium is one of the most common genus belonging to Trichocomaceae and comprises 483 species (Sanzani et al. 2016; de Botelho et al. 2022). *Penicillium* occurs in a wide range of habitats, such as soil, vegetation, air, indoor environments, and various foodstuffs. They are decomposers of organic matter and destructive rot-causing agents in the food industry, producing a wide range of secondary metabolites (Sanzani et al. 2016). Also, some species are important in food industry to produce cheeses and fermented sausages and in medicine due to production of penicillin that is a secondary metabolite with biological activity (de Botelho et al. 2022).

However, *Penicillium* spp. are the major cause of deterioration and decomposition of many plant products after harvest, mainly fruits (El-Samawaty et al. 2021). Additionally, some species could produce toxic secondary metabolite as ochratoxin A (OTA) that widely contaminates food and feed (Ferrara et al. 2022). OTA could be synthesized by *Aspergillus*, *Penicillium*, *A. ochraceus*, *P. nordicum* e *P. verrucosum* and are founded in cereals, herbs, oilseeds, figs, beef jerky, fruits, and wine. The main toxic effects and diseases caused by this mycotoxin is kidney and liver damage, loss of appetite, nausea and vomiting, immune system suppression, and carcinogenic (El-Sayed et al. 2022).

Another species that could affect food is *Penicillium expansum* that causes blue mold rot and this problem is an international concern and is worldwide distributed. Furthermore, this is one of the most important post-harvest diseases of some fruits, such as apples. Various strains of *P. expansum* are able to produce patulin, a mycotoxin (Bahri et al. 2019; Luciano-Rosario et al. 2020).

5.2.2.4 *Fusarium*

Fusarium is a genus of molds that could be founded in different places as in soil, water, plants, and in the air. *Fusarium* species can adapt to a varied range of habitats but are mainly distributed in the tropical and subtropical regions (de Botelho et al. 2022; Perincherry et al. 2019). Some species of these genus are adaptable and resistant to antifungal compounds and many species are causing diseases in plants, such as wilt and canker (Perincherry et al. 2019). However, one of the main concerns regarding this group of fungi is the production of mycotoxins that could contaminate food products. Some known species that produce mycotoxins are *F. graminearum*, *F. oxysporum*, *F. sporotrichioides*, *F. verticillioides*, and *F. proliferatum* (El-Sayed et al. 2022; Perincherry et al. 2019).

The action of this fungus is characterized by the colonization of the host as biotrophic fungi, and then production of toxins and enzymes. *Fusarium* species produce mycotoxins and are most important in cereals (wheat, oats, barley, maize) and tropical fruit crops (banana and pineapple, lentils, tomato, and peas). Examples of species that produce mycotoxin are *Fusarium verticillioides* and *F. culmorum*. These species produce fumonisins that cause human encephalomalacia, pulmonary edema, carcinogenicity, neurotoxicity, liver damage, heart failure, and esophageal cancer (El-Sayed et al. 2022; Perincherry et al. 2019). *F. oxysporum* produces tricothecenes as T2 toxin and deoxynivalenol (DON) and these mycotoxins are found in cereals, feed, silage, legumes, fruits, and vegetables. These mycotoxins could cause

immune suppression, cytotoxicity, skin necrosis, hemorrhage, anemia, granulocytopenia, oral epithelial lesions, hypotension, and coagulopathy (El-Sayed et al. 2022).

5.2.2.5 Yeasts

Yeasts are eukaryotic fungi, majority unicellular and reproduce by budding (Zhang et al. 2020). Among yeast species, the genera *Saccharomyces*, *Candida*, *Torulopsis*, and *Hansenula* have been the main ones associated with fruit and juice fermentation. Some other yeast species that can cause loss of product quality include *Rhodotorula mucilaginosa*, *Rhodotorula glutinis*, *Zygosaccharomyces bailii*, *Zygosaccharomyces bisporus*, and *Zygosaccharomyces rouxii* (Kaczmarek et al. 2019).

One of the major yeast genera is *Candida* that includes about 200 species. This genus could be found in distinct environments as in the human microbiota, in animals, in the skin, and mucous membranes. This group is considered opportunistic pathogenic fungi, and some species cause human infections (de Botelho et al. 2022).

As yeasts are ubiquitous, microorganisms could occur in raw materials that are utilized for food manufacture. Usually these microorganisms are associated to inappropriate practice in food production that facilitated the cross contamination. Spoilage caused by yeasts can generally be related to the action of enzymes (cellulases, lipases, and proteases), gas production, discoloration, and off-flavors in food. The most common species in spoilage fruit are *Hanseniaspora*, *Pichia*, and *Wickerhamomyces* (Osimani et al. 2022).

5.3 Plant Essential Oils

5.3.1 Definition

Essential oils (EOs) are natural products of several volatile molecules (El Asbahani et al. 2015; Huang et al. 2021). These compounds are widely known for their taste, aroma, and antimicrobial activity (Salvi et al. 2022). EOs are commonly distributed in Myrtaceae, Lamiaceae, Rutaceae, Umbelliferae, Lauraceae, Zingiberaceae, Asteraceae, and other plants (Ni et al. 2021). EOs are extracted from different plant parts, such as a leaf, flower, root, stems, and others non-woody organs (Arasu et al. 2019; Ni et al. 2021). Obtaining methods can be by distillation, mechanical pressure, or solvent extraction (Hou et al. 2022). The method can influence the yield, quality, and final product composition (Skendi et al. 2022).

5.3.2 Characteristics

Essential oils are formed by different substances, such as aldehydes, terpenes, phenolics, esters, alcohols, and ketones (Arasu et al. 2019). Its volatile nature, attributed to secondary metabolites, has antioxidant and antimicrobial potential (Salvi et al. 2022), representing approximately 90% of the composition. Other non-volatile

components (acids, sterols, waxes), present in smaller amounts, can also significantly influence its bioactivity (Luque de Castro et al. 1999).

According to Ni et al. (2021), phenolic compounds are the most important for the antibacterial activity of EOs. Terpenes, phenols, esters, aromatic compounds, aldehydes, or terpenoids cause inhibition of microorganisms' growth in food and preventing spoilage of food.

Among the volatile substances found, monoterpenoids and sesquiterpenoids are the main constituents of EOs (Fotsing Yannick Stephane and Kezetas Jean Jules 2020). Terpenes and other compounds give EOs antimicrobial, antioxidant, and antifungal properties (Arasu et al. 2019). Terpenoids can affect the fatty acids of the cell membrane of microorganisms, thus altering their permeability and, consequently, causing leakage of intracellular substances (Ju et al. 2019). Phenolic compounds, conversely, have important antibacterial activities, such as disruption of the cytoplasmic membrane, changes in the flow of electrons, and coagulation of cell contents (Dhifi et al. 2016). It is important to highlight that the compound action in the essential oil will also depend on the type of target microorganisms, due to their composition and membrane thickness (Ju et al. 2019).

Additionally, other functional groups present in EOs have become important as hydrocarbons (α -pinene and sabinene), oxides (linalool oxide, and cineol), lactones (citroptene), esters (eugenol acetate), alcohols (linalool and geraniol), phenols (thymol and eugenol), ketones (camphor, and carvone), and aldehydes (citral, citronellal, and cinnamaldehyde) (Ni et al. 2021).

Recent studies have shown antimicrobial, insecticide, or repellent action on pests using EOs (Barros et al. 2022; Seshadri et al. 2020a; Wang et al. 2022; Xu et al. 2022). These positive results indicate that EOs can be used as natural preservative agents in the food industry. In addition, their use can attract consumers to be aware of food safety and reduce chemical agents in the food chain (Miao et al. 2020; Li et al. 2022).

5.3.3 Main Plants' Essential Oils Applied in Food Processing

There Are Many EOs that Are Tested in Food in Studies. Among them Are Lavender EO, Thyme EO, Peppermint EO, Cajuput EO, Cinnamon EO, Clove EO, Eucalyptus EO, and Sage EO (Wińska et al. 2019). Next, some Studies with the Application of these Different EOs Will Be Presented

Studies have shown that cinnamon essential oil is used alone, combined with other EOs, or with other preservation methods/agents for conservation and sanitization purposes. For example, Seshadri et al. (2020a) observed antimicrobial action against bacteria and fungi in vitro and in vivo analyses (in guavas) combining EOs extracted from the bark of *C. lourerii* and the plant *Evolvulus alsinoides*. In another study, using EO from *C. zeylanicum* and *Cymbopogon citratus* efficiently reduced fungal growth and total inhibition of the pathogens *C. musae*, *F. incarnatum*, and *F. verticillioides* (Kamsu et al. 2019). Xu et al. (2022) also observed significant antimicrobial effects when combining cinnamon EO with intermittent mild heating, resulting in a reduction of 4 log CFU/g in alfalfa seeds.

The use of EOs can also be used to delay post-harvest deterioration. For example, in in vivo tests, peppermint essential oil inhibited 100% of the development of *Colletotrichum gloeosporioides*, which is responsible for anthracnose in papayas (Ayón Reyna et al. 2022). Likewise, tomatoes use xanthan gum coating, whey protein, and clove EO that prolonged shelf life and maintenance of quality attributes, such as firmness, after 15 days of storage at 20 °C (Kumar and Saini 2021).

Research using clove, oregano, and thyme EO is also found in the literature. For example, Cui et al. (2020) observed a change in the morphology of biofilms when using clove EO, resulting in a 61.48% reduction in the metabolic activity of *E.coli*. This change may be associated with cell collapse and exudation of intracellular content caused by components present in clove EO. In another study, clove essential oil reduced 32.7% of *B. gladioli* cells, and this effect was potentiated by combining essential oil nanoemulsions in chitosan coating, achieving a 52% of reduction. The reduction was associated with the lipid structures of cell membranes disruption and mitochondrial membranes. Furthermore, this study observed better conservation of fresh *Tremella fuciformis* treated with clove oil and chitosan (Wang et al. 2022).

In vitro studies, the microencapsulated oregano essential oil could inhibit 100% of microorganisms on avocados. In vivo, a reduction in anthracnose lesions in the fruits was observed, and critical sensorial characteristics, such as color and firmness, were maintained. Another important finding in this research was the increase in the content of flavonoids and phenolic compounds in treated avocados (Colín-Chávez et al. 2022). Similar results in maintaining quality and delaying postharvest effects were also observed by Lee et al. (2022) when studying the EO effect of oregano on chitosan and cellulose coating in strawberries.

Sazvar et al. (2022) evaluated the effect of the essential oils (anise, chamomile, marjoram, black caraway, and thyme) on inhibiting the growth of *Alternaria*. These authors tested five concentrations (0, 200, 400, 600, and 800 µL/L) and observed that in vitro results revealed that by increasing the concentration of essential oils, their antifungal activity improved. However, the greatest inhibitory effect was associated to the use of thyme essential oil. When evaluating the application of the oil in vivo (fruit), the better result is related to lowest percentage of fruit weight loss, and this was observed with thyme oil at 600 µL/L and a 0.02% concentration.

Thyme essential oil can also be used as a repellent against pests that affect cereals during storage without changing sensory and quality parameters and can be used to replace chemical agents and reduce the environmental impact (Barros et al. 2022). Other essential oils used in food are presented in Table 5.2.

5.4 Application of Essential Oils against Fruit Spoilage Fungi

In the above section, we discussed about the application of different types of EOs. Essential oils have been used in the industry as a flavoring. Thus, in recent years, new research has shown that it can also be used for reducing microorganisms in food (Angane et al. 2022; Osimani et al. 2022).

Table 5.2 Examples of studies using essential oils in fruits

Matrix used	Type of essential oil	Application	Reference
Dragon fruit	Cinnamon oil and lemon grass oil and eucalyptus oil and clove oil, and rosemary oil	In vivo assay	Castro et al. (2017)
Tomato	<i>Adansonia digitata</i>	Sanitizer	Kayode et al. (2018)
Mangosteen	Peppermint oil and lime oil	Closed system	Owolabi et al. (2021)
<i>Piper nigrum</i> L. fruits	<i>Boswellia carterii</i>	Sanitizer	Prakash et al. (2014)
Avocado	<i>Lippia scaberrima</i> /spearmint oil	Commercial coatings	Regnier et al. (2010)
Papaya	Mint oil	Chitosan coating	Ayón Reyna et al. (2022)
Strawberry and cherry	<i>Zanthoxylum bungeanum</i>	Active package film	Zhang et al. (2022)

When it comes to vegetable food, there is a big concern about pathogen food-borne diseases since they are used to produce ready-to-eat food (Nikkhah et al. 2017; Nikkhah and Hashemi 2020; Shen et al. 2023). So, these oils can be applied to food in different ways; for example, sanitizers that can be sprayed on the food surface or essential oils can be used in edible coatings and films (Atif et al. 2020b; Shen et al. 2023). EO_s have already been used in different vegetable foods, such as raw vegetables and fruits or cereal-based foods and juices (Osimani et al. 2022).

According to Ni et al. (2021), these applications could be restricted due to the characteristics of EO such as high volatility, **hydrophobicity**, and ease of **oxidation**. However, there is a potential use when micro and nanotechnology were applied.

In this section, we will be present the application of EOs in food preservation and agricultural products.

5.4.1 Food Packaging: Edible Coatings and Films

Consumers demand healthy, fresh food that is as close to natural as possible, especially without the use of synthetic food additives. Food additives are purposely added to obtain technology and sensorial functions. Some of them could control microbial growth, chemical reactions, and promote an increase of shelf life of food products (Mesías et al. 2021). However, at same time, synthetic food additives could be a risk to human health and environment (Ni et al. 2021).

Currently, active food packaging could be a good option that unites an innovative method and possible ecological solution. This packaging is commonly applied with biodegradable materials and natural compounds. So, EOs could be used in films and coatings (Ni et al. 2021).

When it comes to edible coatings and films, there are differences between them, and it will influence industries' choices when applying them to food. Protein, starch,

and polysaccharide are the most used ingredients in edible coatings. However, it is crucial that in the final product, the coat must have a particular texture to cover the fruits. In this case, the coats are thick liquids into which the fruit is dipped (Shen et al. 2023; Tripathi et al. 2021).

Edible films, on the other hand, are prepared as thin sheets. Therefore, a drawback is that it can be broken and let some portion of the food exposed, not conferring the protective goal (Pandey et al. 2022).

Edible coatings are often used in food. Usually, chitosan is one of the most popular ingredients in this material, mainly because it has antibacterial and antifungal properties. However, in recent studies, essential oils have been shown that it can improve the chitosan's effect on food (Anjum et al. 2020; Pandey et al. 2022; Shen et al. 2023).

Nanoemulsions are used for adding essential oils to edible coatings. The reason is that this process guarantees a stable emulsion that will not suffer during the storage period and break its connection (Anjum et al. 2020).

In a study with carboxymethyl cellulose and cardamom oil in a nanoemulsion edible coating in tomatoes, the edible coat was efficient to prolongate shelf life due to oxidative damage and bacterial growth (Das et al. 2022).

Long et al. (2022) analyzed chitosan/fennel seed essential oil/starch sodium octenyl succinate composite films in different formulations applied to apples. They found that the film in special one of the formulations had a powerful fungicidal effect against *B. cinerea*, *T. roseum*, and *P. expansum*. However, all six formulations used in the study showed antifungal activity in vitro.

Guo et al. (2022) produced a film material to preserve apricots by inhibiting the growth of *Alternaria alternata*. EO Oregano essential oil (OEO) (2%, 4%, 6% (w/v)) was added to chitosan-based films in the form of nanoemulsions. The 4% OEO nanoemulsion with 20 min of ultrasound treatment demonstrated the best antifungal action. The chitosan film combined with 1% (v/v) of 4% (w/v) OEO nanoemulsion significantly increased the antifungal properties. These authors observed also that CS-1%OEO films control the disease resistance of apricots from the black spot caused by *Alternaria alternata*.

According to Ni et al. (2021), EO affects the microstructure of the final packaging material and could contribute to increase the tensile, barrier, and optical properties. Furthermore, it is important to consider that composition of food could influence the migration rate of compounds.

5.4.2 Sanitizers

Sanitization is an important step for the microbiological control of fruits. The most common way to sanitize fruits and vegetables is by immersion in chlorinated compounds (de São José et al. 2018; de Moraes Motta Machado et al. 2022; Moncioso et al. 2021; Pelissari et al. 2021). However, since these chemicals are used mainly in food industries due to their efficiency and low costs, other additional treatments come to improve further functionality (Atif et al. 2020b; Kanetis et al. 2007). Furthermore, the chlorinated compounds are intended to be substituted due to

potential harmful consequences on the environment and health (Monciosio et al. 2021; Pelissari et al. 2021).

Recently, many studies have shown, according to their methodology, possible use of essential oils as sanitizers, tested directly on the fruit or vegetable surface, or even as a possible compound to be used in a blend of other antimicrobial components (Antonioli et al. 2020; Atif et al. 2020b; Nikkhah et al. 2017; Nikkhah and Hashemi 2020; Seshadri et al. 2020b; Chen et al. 2022).

Studies proposing the use of essential oil to control fungi are scarce. However, we present this study to indicate that there is a trend toward the application of EOs as sanitizers. Soraggi Battagin et al. (2021) evaluated the clove essential oil as an alternative sanitizer for the disinfection of citrus fruit in packinghouses to control *Xanthomonas citri* subsp. *citri*. Clove EO was able to inhibit *X. citri* when used at 0.75% (v/v). In simulate packinghouse conditions, the sanitization process with 5% of the EO promoted an effect similar to sodium hypochlorite.

Atif et al. (2020a, b) have studied two essential oils from medicinal plants and protective properties in jack fruits. They sprayed the essential oils onto the jackfruit peel and the damage designed to the jackfruit peel. *Basilicum* oil has shown promising activity against *Rhizopus microsporus* MTCC383; mycelial growth was affected by *Basilicum* and *V. zizanioides* indicating that essential oils affected the developmental stages of fungi.

In a study with essential oils of *Cinnamomum loureirii* and *Evolvulus alsinoides*, researchers found that after applying essential oil emulsions to the guava peeled skin, after eight days, the oils were capable of inhibiting food spoilage fungus growth. (Seshadri et al. 2020b).

Monciosio et al. (2021) evaluated citric acid and clove essential oil as alternatives to chlorine compounds on sanitization of apples. The authors observed that the treatment with clove essential oil reduced the amount of the fungi group to 1.64 log CFU/g.

5.4.3 Agricultural Application

EOs have potential to control pests and diseases of crops as substitute of synthetic pesticides, and they could be safer to the humans and the environment (Torre et al. 2021).

Hashem et al. (2019) aimed to apply Thieves oil blend (six oils in one mixture) to protect peach fruit from spoilage during cold storage. The authors evaluated the effects on *Alternaria alternata*, *Fusarium oxysporum*, *Geotrichum candidum*, and *Monilinia laxa*. Thieves oil blend (2.0 mL/L) inhibited fungal growth in vitro and reduced the disease rate to 12.0% and the disease severity index to 1.2 after 7 days at 27 °C in peach fruit.

Arasu et al. (2019) evaluated essential oil of four medicinal plants and protective properties in plum fruits against the spoilage bacteria and fungi. The minimum inhibitory concentration of the essential oils against fungi ranged from 1.1 ± 0.4 to 292 ± 3.2 µg/mL. *Allium sativum* presented the highest antifungal activity against

Penicillium notatum and inhibited the growth of *Aspergillus niger*, *Aspergillus flavus*, and *Rhizopus microsporus*. Authors concluded that *Allium sativum* EO could inhibit deterioration of plum fruit.

Torre et al. (2021) evaluated that basil aromatic oil was against three common storage fungi (*Fusarium oxysporum*, *Penicillium* spp., and *Colletotrichum gloeosporioides*), and also the effect of basil aromatic oil on the germination of commercial seeds (lettuce and tomatoes). The basil essential oil presented toxicity against the fungi *F. oxysporum*, *Penicillium* spp., and *C. gloeosporioides*. The authors observed also that tomato and lettuce seeds' germination were not significantly affected by the essential oils.

Xu et al. (2021) investigated the effectiveness of tea tree oil, thyme oil, rosemary oil, and lemon oil for controlling rot in post-harvest peaches (*Monilinia fructicola*). Tea tree oil had the antifungal action against in vitro and in inoculated peach fruit. According to these authors, tea tree oil impacts on the composition of cell membrane, causing alterations in mycelial morphology, membrane permeability, and concentrations of intracellular reactive oxygen species. Finally, the authors indicated this oil as a possible alternative for conventional fungicides applied to control rot in peach.

5.5 Limitations of the Use of EO

The structure and composition of a food can make it difficult and restrict the activity of essential oils since some foods require higher concentrations due to their matrix. In addition, many studies evaluate the use of essential oils in vitro. However, higher levels of essential oils are fundamental for the same effect to be performed on food (Speranza and Corbo 2010), which can have consequences for sensory aspects. Furthermore, the essential oil obtained depends on some aspects such as climate and maturation stage, which can interfere with the quality of the product to alter its constitution and, therefore, the activity of this compound.

5.6 Final Considerations and Future Perspectives

Essential oils presented potential in application against fruit spoilage fungi. The advances in studies in this area contribute to the issue of food safety from various aspects. The advantages include environmental and consumer health issues since they are compounds of natural origin. In addition, the antimicrobial potential of these oils contributes to reducing waste and fighting fruit spoilage, which has economic benefits. However, there are some challenges. For example, the use of essential oils on an industrial scale is hampered due to the low solubility and stability of these substances. More studies are required to unravel, elucidate, and improve the knowledge about essential oils and their mechanisms. Some applications, such as nanoemulsions, deserve more attention to expand their use in edible food coatings.

Furthermore, combining essential oils with other preservation techniques, for example, should be evaluated and used on an industrial scale.

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