

A systematic review and meta-analysis of the use of plant essential oils and extracts in the development of antimicrobial edible films for dairy application

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Abstract

The purpose of this review was to assess the use of plant essential oils and extracts (PEOE) in the development of antimicrobial edible films for dairy application through a systematic review and meta-analysis. All studies published in multiple databases were explored via PRISMA protocol on November 1, 2022. According to the results, the interquartile range of pathogen reduction potential of essential oil (EO) in dairy products, irrespective of EO, film and product type, was 0.10 - 4.70 log CFU g⁻¹ per % concentration. The findings from 38 articles indicate that among all EOs or their compounds, *Zataria multiflora* Boiss in protein film, thyme in protein film, *Z. multiflora* Boiss EO in protein film, *Trans*-cinnamaldehyde in carbohydrate film and lemongrass EO in protein film had extraordinary pathogen reduction potential on important foodborne pathogens. In the case of plant extract, fish gelatin film with *Lepidium sativum* extract, whey protein isolate film loaded with oregano EO and carboxymethyl cellulose film with clove EOs had the highest antimicrobial effect on mesophilic bacteria (9.50 log CFU g⁻¹ per % concentration), yeast-mold (2.63 log CFU g⁻¹ per % concentration) and mesophilic/psychrophilic counts (> 9.06 log CFU g⁻¹ per % concentration), respectively. *Listeria monocytogenes* is the primary species of interest; whereas, mesophiles and mold-yeast populations were the most investigated microbiota/mycobiota in cheese with PEOE-incorporated film. In light of these findings, the choice of PEOE at appropriate concentrations with the selection of appropriate edible film may improve the safety, sensory, and shelf life of dairy products.

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Introduction

Milk and dairy products have high nutritional value due to their lipids, proteins, carbohydrates (lactose), amino acids, vitamins, and minerals.¹ However, these products are an appropriate environment for the growth of microorganisms (e.g. foodborne pathogens and spoilers) which can lead to food spoilage and safety consequences.² According to the type of microorganism, the spoilage mechanisms of dairy products are different. Generally, when microorganisms break down carbohydrates, proteins, and milk fats by their extra-cellular enzymes, spoilage occurs and all or part of the product becomes unusable and dairy food waste is created.³ It has been proven that packaging plays a vital role in the protection of dairy products post-processing and can be used as an effective method to improve shelf life.

Active packaging is one of the latest methods of smart

food packaging being used in a variety of ways. Antimicrobial packaging is one of the most important approaches which is of great interest to producers, consumers, and researchers around the world. In this method, antimicrobials are added to the self-standing matrix and used to enhance the quality and microbial safety of food.⁴ One of the most important techniques in manufacturing this type of packaging is to include antimicrobials in polymer substrates (e.g. synthetic and natural) in order to develop antimicrobial films.⁵ Edible films are thin layers less than 0.30 mm in thickness being used to protect food from the environment (i.e. ultraviolet rays, water/organic vapors and gases), mechanical damage, microbial growth, and contamination.^{6,7} Polysaccharides (e.g. pectin, starch, alginate, carrageenan and xanthan gum), proteins (e.g. collagen, soy protein, milk protein and zein) and lipids (e.g. waxes and fatty acids) have been widely used as biopolymers to develop edible

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antimicrobial films.⁸⁻¹⁰ Among different natural antimicrobial compounds originating from plants, animals, microorganisms, and mushrooms, plant-based antimicrobials are attracting increasing interest. The addition of plant essential oils and extracts (PEOE) in the film formulation will increase the antimicrobial activity and ensure microbial safety and quality of food.^{11,12}

The PEOE are anti-microbial compounds being extracted from different parts of the plant, including roots, bark, leaves, seeds and fruits^{7,13} by steam distillation, mechanical processes, and dry distillation in the case of essential oil (EO) and solvent extraction in the case of plant extracts.¹⁴ The PEOE have numerous advantages compared to chemical preservatives in terms of safety and consumer desires and are approved by the food and drugs administration as "generally recognized as safe". The PEOE represent strong or low anti-bacterial, anti-viral, and anti-fungal performance; however, they have antioxidant activity and food flavoring properties.¹⁵⁻¹⁷ A major proportion of the antimicrobial activity of PEOE is attributed to their phenolic content mainly flavonoids and their derivatives.^{18,19} The antimicrobial activity mechanisms of phenolic compounds are related to the destruction of the cell wall and cytoplasmic membranes, preventing the synthesis of DNA, RNA, proteins and polysaccharides in bacteria and fungi.²⁰

There have been various original types of research on the use of PEOE in developing of anti-microbial edible films for dairy applications worldwide. However, there are a few comprehensive reviews on the use of PEOE in edible films for dairy applications.^{14,21} Then, to reach an overall and concise conclusion, it is necessary to conduct a systematic review of published studies. To our knowledge, no systematic review or meta-analysis has been conducted on this topic. The purpose of this review was to critically examine information obtained from all published studies on the use of PEOE in the development of antimicrobial edible films for dairy application.

Materials and Methods

Search strategy. The PRISMA guide was used to perform a literature search and article extraction, as shown in Figure 1.²² To examine and extract the required results from published articles and reports related to the subject, a systematic search was carried out in national databases including Magiran, IranDoc and SID as well as international databases including ScienceDirect, Scopus, PubMed, and Google Scholar up to November 1, 2022. References from all articles related to the subject were also reviewed. To maximize the exhaustiveness of the search, both general and specific keywords include the words "food packaging", "active packaging", "biodegradable packaging", "antimicrobial agent", "dairy products such as cheese, butter, yogurt, cream, etc.", "essential oil or

essence", "extract", "plant extract", "edible film", "edible packaging", and all possible combinations of words have been used in combination with "AND" and "OR" operators for English language databases. In addition, a manual search was performed by verifying the list of identified article references to additional articles. This systematic review has included articles dealing with the use of PEOE in the development of only edible films (i.e. biopolymers) for dairy applications. Subsequently, duplicate reports or articles having enough information were removed.

Data extraction. Data extracted from each study include the type of film, first author's name and the year of publication, type of cheese/product, source of EOs (or their compounds) and extract, type of edible film, film preparation method, the function of film, form of using EOs and type of pathogenic and spoiler microorganisms and their reduction rate. From the paired data, the pathogen and/or other microbial counts in the treatment group (i.e. extract or EO) were subtracted from the control group (without extract or EO) to calculate the reduction rate. The pathogen/spoiler reduction was then divided by the concentration of extract or EO (in %) to arrive at microbial reduction potential (expressed as log CFU g⁻¹ per % concentration).²³

Inclusion and exclusion criteria. The inclusion criteria were edible films loaded with EO and/or extract and their use in dairy products, full-text availability, being original research, and availability of pathogen and/or spoiler reduction rate. Studies failed to meet the criteria were excluded.

Statistical analysis. In this study, the two-way ANOVA test was used to compare the mean reduction in microbial load in dairy products by PEOE according to the type of film and type of dairy product. All analyses were performed with GraphPad Prism (version 5.0; GraphPad Software Inc., San Diego, USA).

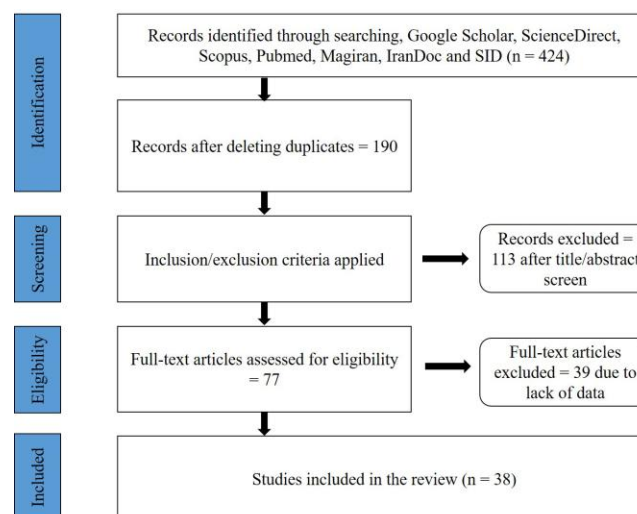


Fig. 1. Flow chart of retrieved, screened, included, and excluded articles.

Results and Discussion

During the initial search, 424 articles were found; but, after deleting the duplicate articles and reviewing the titles and abstracts articles, 77 articles were examined. Thirty-nine articles were subsequently left out due to inadequate data mentioned in the section “inclusion and exclusion criteria”. Finally, a total of 38 articles were included in the study (Fig. 1).

Effect of EO-incorporated films on dairy pathogens.

The EOs are among the most studied natural antimicrobials in food. Although EOs are commonly used as flavoring agents in the food industry, most of them exhibit different antimicrobial performances on both food pathogens and spoilers. A key mechanism for the antimicrobial effect of EOs has been demonstrated to be linked to the dissolution of EO components in the lipids of bacterial cell membranes and mitochondria due to their lipophilic properties, leading to the leakage of proteins and nucleic acids and finally cell death.²⁴ In addition, EOs can affect cell metabolic activity and cause microbial death by changing bacterial membrane potential.²⁵ The interquartile range (IR) of pathogen reduction potential of EOs in dairy products, irrespective of the type EO, film and dairy product, was 0.10 - 4.70 log CFU g⁻¹ per % concentration. Therefore, a fairly high variability in the antimicrobial efficacy of EO is evident implying that not all EOs incorporated into edible films can give promising results. The type of cheese is a key factor influencing the pathogen reduction potential, since certain intrinsic factors such as moisture, water activity, pH and type of process can affect the growth and death of the pathogen. Cheeses can be categorized according to the moisture content as hard (< 25.00% moisture), semi-hard (25.00 - 36.00% moisture), semi-soft (36.00 - 40.00% moisture), and soft cheeses (> 40.00% moisture). From a microbiology point of view, cheese with high moisture content is an appropriate medium for the rapid growth of bacterial strains.²⁶ According to the Figure 2, most of the studies conducted on the use of EO-containing films have focused on semi-hard, soft, and hard cheeses, respectively. *Listeria monocytogenes*, *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella enteritidis* are among the major pathogens studied in the published literature (Fig. 2). The *L. monocytogenes* is the major species followed by *S. aureus*. The growth/survival behavior of these bacteria is different depending on the cheese. The growth/survival behaviors of *L. monocytogenes* and *S. aureus* have been reported in some soft and semi-soft cheeses including Feta cheese and Iranian white cheese.^{27,28} Among the EOs, the most antibacterial performance was found on the use of *Zataria multiflora* Boiss (*Z. multiflora*) on *S. aureus* (4.70 log CFU g⁻¹ per % concentration; Fig. 2A), *E. coli* (2.88 log CFU g⁻¹ per % concentration; Fig. 2B), *S. enteritidis* (4.70 log CFU g⁻¹ per % concentration; Fig. 2C), and *L. monocytogenes*

(3.24 log CFU g⁻¹ per % concentration; Fig. 2D) in soft cheese. In addition, the highest effect was reported on *Bacillus cereus*;⁵ while, the lowest effect was reported on *S. aureus*.²⁹ A high antimicrobial activity is linked to the high levels of phenolic monoterpenes such as thymol, carvacrol, and *p*-cymene and their synergy.

As noted, the choice of EO and edible film are important variables needing to be considered in antimicrobial dairy packaging. Edible protein films have excellent mechanical strength compared to polysaccharides.⁸ According to the published papers (Table 1), protein-based films, in particular, zein, are one of the most widely used polymer matrices for dairy applications followed by polysaccharides. Out of the 16 EOs or EO individual chemicals, *Z. multiflora* in zein film (Fig. 3A), thyme in whey protein isolate film (Fig. 3B), *Z. multiflora* in zein film (Fig. 3C), *Trans*-cinnamaldehyde in chitosan film (Fig. 3D), and lemongrass in alginate film (Table 1) had extraordinary pathogen reduction potential on *S. aureus* (4.70 log CFU g⁻¹ per % concentration), *E. coli* (1.82 log CFU g⁻¹ per % concentration), *S. enteritidis* (4.70 log CFU g⁻¹ per % concentration), *L. monocytogenes* (3.87 log CFU g⁻¹ per % concentration), and *B. cereus* (12.40 log CFU g⁻¹ per % concentration), respectively. The tendency towards composite films was less pronounced. For example, chitosan-whey protein film containing *Z. multiflora* EO³⁰ and sodium caseinate-chitosan film with oregano EO³¹ were developed for Feta and Panella cheeses, respectively.

Effect of EO-incorporated films on dairy microbiota/mycobiota. The effect of films containing EOs on the microbiota/mycobiota of soft and semi-hard cheeses has also been studied. Most studies have focused on the effect of antimicrobial film on mesophiles and mold-yeast populations (Table 1). Mesophiles have high proteolytic and lipolytic potential activities, changing the smell, taste, and physicochemical characteristics of cheese.³² The IR of mesophiles reduction potential of EOs in dairy products, irrespective of EO, film, and product type, was 0.25 - 9.06 log CFU g⁻¹ per % concentration (Fig. 4). Out of the seven EOs, only carbohydrate film with clove EO (Fig. 4A) had extraordinary “mesophiles reduction potential” of 9.06 log CFU g⁻¹ per % concentration in soft cheese (Fig. 4B). Cheese spoilage can be caused by fungal growth. The growth of yeast-mold is associated with unpleasant odors, curd liquefaction, and in some cases, mycotoxin production.³³ The most important of these are *Aspergillus*, *Penicillium*, *Fusarium*, *Botrytis*, *Cladosporium*, *Mucor* spp. etc. *Aspergillus* and *Penicillium* are frequently involved in cheese spoilage.³⁴ In cheese, it appears that the effects of antimicrobial films with EO on yeast-mold are greater than mesophilic bacteria, which may be due to the direct contact of EO with molds tending to grow on the surface of the cheese.³⁵ The initial impact of EO is on fungal hyphae, resulting in loss of membrane integrity and reducing the amount of ergosterol.

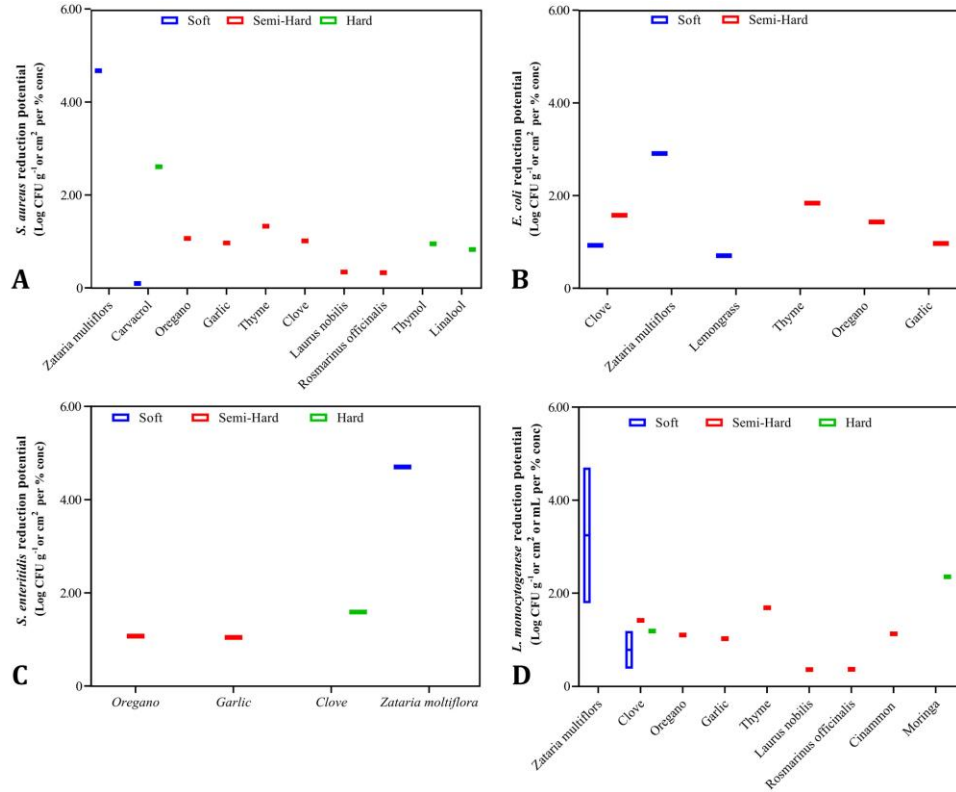


Fig. 2. Antibacterial effect of essential oils against A) *Staphylococcus aureus*, B) *Escherichia coli*, C) *Salmonella enteritidis* and D) *Listeria monocytogenes* based on the type of cheese.

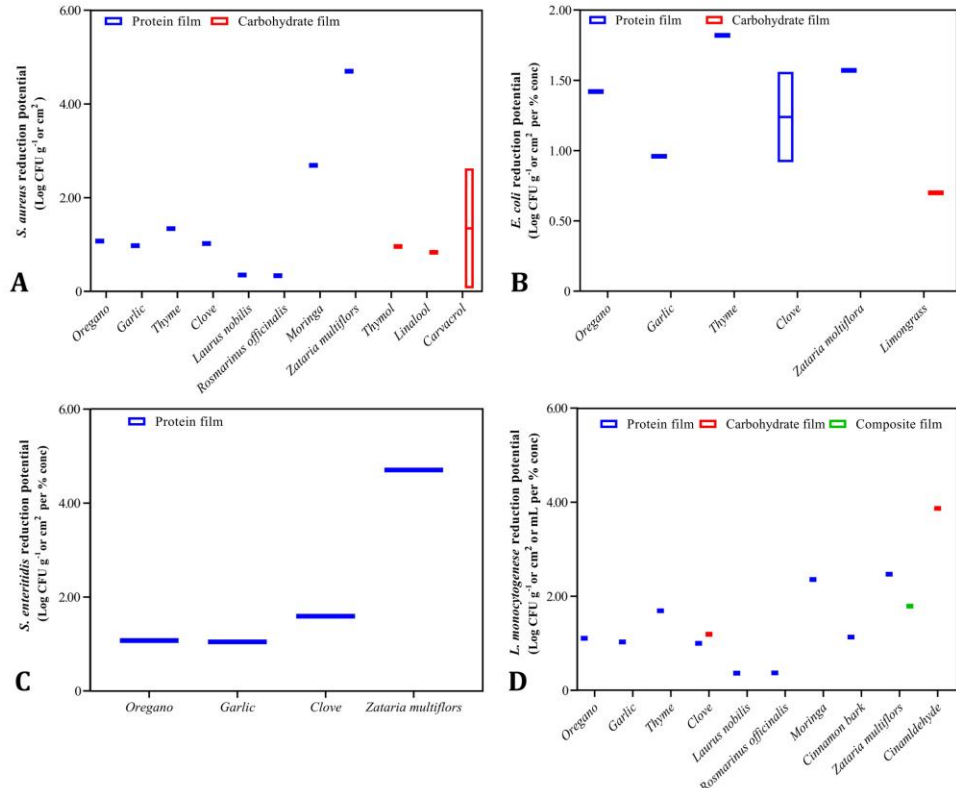


Fig. 3. Antibacterial effect of essential oils against A) *Staphylococcus aureus*, B) *Escherichia coli*, C) *Salmonella enteritidis* and D) *Listeria monocytogenes* based on the type of film.

The EOs also prevent the formation of the wall, leading to the denaturation of cellular components. They are able to penetrate and interfere with the fungal cell wall and cytoplasmic membrane, inhibit membrane ATPases and cytokines, and finally, express a certain number of genes involved in cell adhesion, growth, and sporulation.^{36,37} According to the literature, molds, particularly saprophyte molds with a wide range of degrading enzymes, are less sensitive to EOs.³⁸ The IR of yeast-mold reduction potential of EOs in dairy products, irrespective of EO, film, and product type, was 0.16 - 2.63 log CFU g⁻¹ per % concentration (Fig. 4). The *A. niger* and *Saccharomyces cerevisiae* are among the most investigated mycobiota. Based on the type of film and out of the 4 EOs, oregano EO in protein film, clove EO in the carbohydrate film, and garlic EO in protein film had extraordinary “yeast-mold reduction potential” of 2.06, 2.54 and 2.25 log CFU g⁻¹ per % concentration, respectively (Fig. 4C). However, the type of spoilage varies depending on the type of cheese. Based on the type of cheese, whey protein isolate film containing oregano EOs in semi-hard cheese and carboxymethyl cellulose film with clove EOs in soft cheese had the highest “yeast-mold reduction potential” of 2.54 and 2.25 log CFU g⁻¹ per % concentration, respectively (Fig. 4D). The antimicrobial effect of other EOs on other different microbial groups has been investigated; but inadequate information was identified in the papers (Table 1). The IR of microorganism reduction potential of EOs in dairy products, irrespective of EO, film, and product type, was

0.32 - 13.60 log CFU g⁻¹ per % concentration (Fig. 5A). Out of 9 EOs, only clove and lemongrass EOs had extra-ordinary reduction potential of > 9.06 log CFU g⁻¹ per % concentration on *B. cereus* and total psychrophilic counts.

Based on the published papers, direct incorporation of EO into edible film through solvent casting is the most common method in the included articles (Table 1). In a simple and low-cost casting technique (so-called wet process), EO is initially dissolved in a previously prepared polymer through the emulsification process and poured on a flat surface made from Teflon or glass templates.³⁹⁻⁴¹ Restriction in molding, uneven thickness, the low amount of film formation, and the long drying time are the most important drawbacks of the casting technique.⁴² Recently, researchers have attempted to investigate new methods of film development such as lamination (layer-by-layer film) and electrospinning techniques (Table 1). Electrospinning has been successfully used in the fabrication of EO-based antimicrobial film for dairy applications due to a large surface-to-volume ratio, and the processing conditions of electrospun mats which address the sustained-release specification of antimicrobial films.^{4,14,27,43} The EOs are also added to the polymer matrix in the form of macro-emulsion, microemulsion, nanoemulsion, and Pickering emulsion. The main interest in this area is the EO nanoemulsion, which is more being used in recent years for the development of EO-based antimicrobial films due to the specific shape, size, and stability over macro-emulsion and microemulsion⁴⁴ (Table 1).

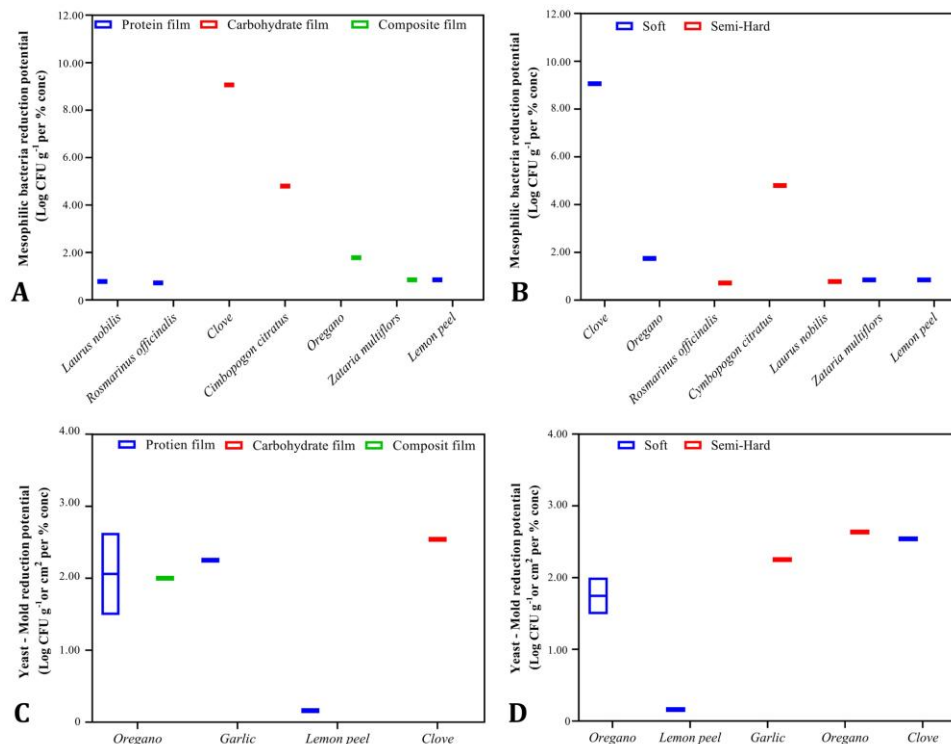


Fig. 4. Antimicrobial effect of essential oils on mesophilic microorganisms in cheese based on the **A)** type of film and **B)** type of cheese, and also against yeast-mold in cheese based on the **C)** type of film and **D)** type of cheese.

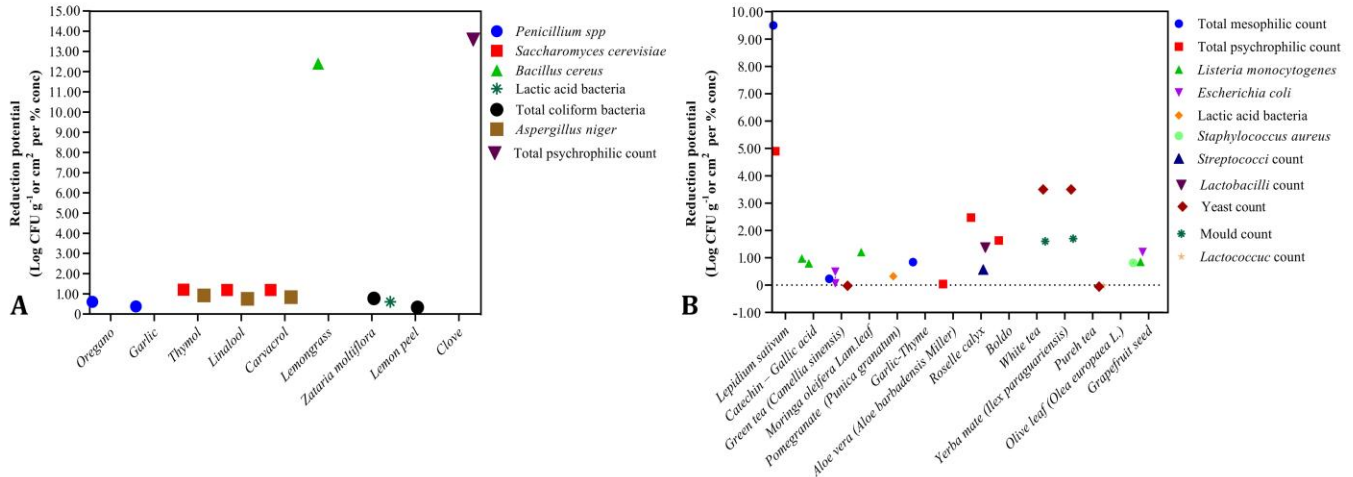


Fig. 5. A) Antimicrobial effect of edible films incorporated with essential oil on different microbial groups in dairy products. **B)** Antimicrobial effect of edible films incorporated with extract on different microbial groups in dairy products.

Effect of extract-incorporated films on dairy pathogens. The majority of studies focused on the use of composite films, followed by carbohydrate and protein films. However, most of the studies dealt with the use of extract-incorporated films in soft, semi-hard, and hard cheeses, respectively. Casting was the only method used for developing extract-containing films (Table 2). The IR of pathogen reduction potential of extract in dairy products, irrespective of extract, film, and dairy product, was 0.06 - 1.21 log CFU g⁻¹ per % concentration. Out of the eight extracts or extract individual chemicals, grapefruit seed extract in red alga film in sliced cheese and green tea extract in whey protein film in soft (mixture fresh goat-sheep) cheese had the highest and lowest pathogen reduction potential on *E. coli*, respectively (1.21 and 0.064 log CFU g⁻¹ per % concentration).

Effect of extract-incorporated films on dairy microbiota/mycobiota. The IR of total bacteria count reduction potential of plant extracts, regardless of extract, film, and product type, was -0.015 - 1.20 log CFU g⁻¹ per % concentration (Fig. 6), revealing the fact that the antimicrobial effect of the extract is different and not all extracts give acceptable outcomes. Out of the 9 extracts, *Yerba Mate Ilex paraguariensis* loaded on furcellaran-whey protein film and pomegranate peel loaded on zein film (Fig. 6A) had the highest antimicrobial effect (0.90 and 1.20 log CFU g⁻¹ per % concentration, respectively) in the soft cheese (Fig. 6B); meanwhile, pu-erh tea extract had a negative antimicrobial effect (-0.015 log CFU g⁻¹ per % concentration). It appears that some of these negative findings may be related to the intrinsic factors of certain cheeses which may support the growth of certain microbial strains. The results from other studies are presented in Figure 5B.

The most promising effect was observed on mesophilic bacteria in the use of fish gelatin film incorporated with *Lepidium sativum* extract in Ricotta cheese;⁴⁵ while, the yeast population was interestingly increased in the presence of furcellaran-whey protein film incorporated with green tea extract in Quark cheese.⁴⁶ Out of 14 extracts, the greatest antimicrobial efficacy was found with the use of *L. sativum* extract on mesophilic and psychrophilic bacteria (4.90 and 9.50 log CFU g⁻¹ per % concentration, respectively), and white tea and yerba mate extracts on yeast count (both 3.50 log CFU g⁻¹ per % concentration; Fig. 5B). Plant extracts significantly affect the cell membrane of Gram-positive and Gram-negative bacteria, lowering the intracellular pH, hyperpolarizing the cell membrane, and eventually leading to cell death.⁴⁷ The sensitivity of Gram-positive and Gram-negative bacteria depends on different factors mainly type of PEOE. Gram-negatives are slightly less susceptible than Gram-positive bacteria. In addition, fungal strains are generally more susceptible to PEOE.

The IR of yeast-mold reduction potential of extracts in dairy products, irrespective of extract, film, and product type, was -1.17 - 2.49 log CFU g⁻¹ per % concentration (Fig. 6). Two (sage and rosemary) out of the 7 extracts loaded in protein film (Fig. 6C) in soft cheese (Fig. 6D) showed a high yeast-mold reduction potential of 2.49 log CFU g⁻¹ per % concentration. However, the negative antimicrobial effect of the garlic-thyme extract on yeast-mold is significant (-1.17 log CFU g⁻¹ per % concentration). The diversity of the chemical components of plant extracts, even those obtained from the same species, results in different inhibitory effects.⁴⁸ In general, the antimicrobial effect of plant extracts is associated with the inhibition of bacterial protein biosynthesis, nucleic acid synthesis, cell wall biosynthesis, and destruction of the bacterial cell wall.⁴⁹

Table 1. Different types of edible films incorporated with essential oils (EOs) applied in various dairy products.

Film source	Product category/ Name	EOs source	Film preparation method	Aim of use in food	Pathogen and reduction rate	Spoiler and reduction rate	Ref.
Film type: Protein							
Whey protein isolate	Semi-hard (Kashar cheese)	Oregano ^a	Casting	Safety	<i>E. coli</i> OEO 2.84 log CFU cm ⁻² (2.00% w/v)	TYM: 5.27 log CFU cm ⁻² (2.00% w/v)	50
					<i>S. enteritidis</i> 2.15 log CFU cm ⁻² (2.00% w/v) <i>L. monocytogenes</i> 2.21 log CFU cm ⁻² (2.00% w/v) <i>S. aureus</i> 2.15 log CFU cm ⁻² (2.00% w/v)	<i>Penicillium</i> spp.: 1.20 log CFU cm ⁻² (2.00% w/v)	
Whey protein isolate	Semi-hard (Kashar cheese)	Garlic ^a	Casting	Safety	1.92 log CFU cm ⁻² (2.00% w/v)	TYM: 4.50 log CFU cm ⁻² (2.00%)	50
					<i>S. enteritidis</i> 2.09 log CFU cm ⁻² (2.00% w/v) <i>L. monocytogenes</i> 2.05 log CFU cm ⁻² (2.00% w/v) <i>S. aureus</i> 1.95 log CFU cm ⁻² (2.00% w/v)	<i>Penicillium</i> spp.: 0.76 log CFU cm ⁻² (2.00% w/v)	
Sodium caseinate	Semi-hard (Pategrás cheese)	Oregano ^a	Casting	Shelf life	NR	TMC: 1.71 log CFU g ⁻¹ (1.00% w/v)	51
						TMC: 0.011 log CFU g ⁻¹ (1.00% w/v)	
Sodium caseinate	Semi-hard (Pategrás cheese)	Laurel ^a	Casting	Shelf life	NR	TMC: 0.011 log CFU g ⁻¹ (1.00% w/v)	51
						TYM: 4.49 log CFU g ⁻¹ (3.00% w/v)	
Whey protein isolate	Soft (Queso blanco cheese)	Oregano ^a	Casting	Shelf life	NR		52
Whey protein isolate	Semi-hard (Kashar cheese)	Thyme ^a	Casting	Safety	<i>E. coli</i> O157:H7 2.74 log CFU g ⁻¹	NR	53
					<i>L. monocytogenes</i> 2.54 log CFU g ⁻¹ <i>S. aureus</i> 2.01 log CFU g ⁻¹ (1.50% v/v) <i>E. coli</i> O157:H7 2.32 log CFU g ⁻¹		
Whey protein isolate	Semi-hard (Kashar cheese)	Clove ^a	Casting	Safety	<i>L. monocytogenes</i> 2.13 log CFU g ⁻¹ <i>S. aureus</i> 1.54 log CFU g ⁻¹ (1.50% v/v)	NR	53
Zein	Semi-hard (Queso Guoda cheese)	<i>Laurus nobilis</i> ^a	Electrospinning	Safety	1.85 log CFU g ⁻¹ (5.00% w/w) <i>S. aureus</i> 1.75 log CFU g ⁻¹ (5.00% w/w)	TMC: 3.90 log CFU g ⁻¹ (5.00% w/w)	4
					<i>L. monocytogenes</i> 1.84 log CFU g ⁻¹ (5.00% w/w) <i>S. aureus</i> 1.72 log CFU g ⁻¹ (5.00% w/w)		
Zein	Semi-hard (Queso Guoda cheese)	<i>Rosmarinus officinalis</i> ^a	Electrospinning	Safety	<i>L. monocytogenes</i> 6.28 log CFU g ⁻¹ (10.00%) <i>E. coli</i> O157:H7	TMC: 3.60 log CFU g ⁻¹ (5.00% w/w)	4
Zein	Soft (Iranian white cheese)	Clove ^a	Electrospinning	Shelf life	6.25 log CFU g ⁻¹ (10.00% w/w)	NR	27

Table 1 (continued).

Zein	Soft (Iranian white cheese)	Clove ^a	Electrospinning	Shelf life	<i>E. coli</i> O157:H7 6.28 log CFU g ⁻¹ (10.00%) <i>L. monocytogenes</i> 6.25 log CFU g ⁻¹ (10.00% w/w) <i>S. aureus</i>	NR	27
Gelatin	Hard (Cheshire cheddar cheese)	Moringa ^b	Electrospinning	Safety	5.38 log CFU g ⁻¹ (2.00% w/v) <i>L. monocytogenes</i> 4.71 log CFU g ⁻¹ (2.00% w/v) <i>L. monocytogenes</i> 1.13 log CFU g ⁻¹ (1.00% w/v) <i>L. monocytogenes</i>	NR	54
Chicken bone gelatin	Semi-hard (Mozzarella cheese)	Cinnamon bark ^c	Casting	Safety	1.19 log CFU g ⁻¹ (1.00% v/v) <i>S. Enteritidis</i> 1.59 log CFU g ⁻¹ (1.00% v/v) <i>E. coli</i> O157:H7	NR	55
Chicken bone gelatin	Hard (Cheddar cheese)	Clove bud ^c	Casting	Safety	2.88 log CFU g ⁻¹ (1.00% w/v) <i>L. monocytogenes</i> 4.70 log CFU g ⁻¹ (1.00% w/v) <i>S. aureus</i>	NR	56
Zein	Soft (Feta cheese)	<i>Z. multiflora</i> ^a	Casting	Safety	4.70 log CFU g ⁻¹ (1.00% w/v) <i>S. enteritidis</i> 4.70 log CFU g ⁻¹ (1.00% w/v)	NR	28
Soy protein	Soft (Iraqi white cheese)	Lemon peel ^a	Casting	Shelf life	NR	TMC: 2.25 log CFU g ⁻¹ (3.00% v/w) TCC: 0.98 log CFU g ⁻¹ (3.00% v/w) <i>Staphylococci</i> count: 1.55 log CFU g ⁻¹ (3.00% v/w) TYM: 0.48 log CFU g ⁻¹ (3.00% v/w)	57
Zein	Pasteurized cow's milk	<i>Z. multiflora</i> ^a	Casting	Safety	<i>E. coli</i> O157:H7 2.64 log CFU mL ⁻¹ (10.00% w/w) <i>L. monocytogenes</i> 1.18 log CFU mL ⁻¹ (5.00% w/w)	NR	58
Film type: Carbohydrate							
Starch	Hard (Cheddar cheese)	Linalool ^a	Casting	Shelf life	NR	<i>A. niger</i> 1.80 log CFU g ⁻¹ (2.38% w/w)	59
Starch	Hard (Cheddar cheese)	Carvacrol ^a	Casting	Shelf life	NR	<i>A. niger</i> 2.00 log CFU g ⁻¹ (2.38% w/w)	59
Starch	Hard (Cheddar cheese)	Thymol ^a	Casting	Shelf life	NR	<i>A. niger</i> 2.20 log CFU g ⁻¹ (2.38% w/w)	59
Starch	Hard (Cheddar cheese)	Thymol ^a	Lamination	Safety	<i>S. cerevisiae</i> 1.20 log CFU g ⁻¹ (1.00% w/w)	NR	60

Table 1 (continued).

Starch	Hard (Cheddar cheese)	Linalool ^a	Lamination	Safety	<i>S. cerevisiae</i> 1.19 log CFU g ⁻¹ (1.00% w/w)	NR	60
Starch	Hard (Cheddar cheese)	Carvacrol ^a	Lamination	Safety	<i>S. cerevisiae</i> 1.19 log CFU g ⁻¹ (1.00% w/w)	NR	60
Starch	Hard (Cheddar cheese)	Thymol ^a	Lamination	Safety	<i>S. aureus</i> 0.46 log CFU g ⁻¹ (0.48% w/w)	NR	61
Starch	Hard (Cheddar cheese)	Linalool ^a	Lamination	Safety	<i>S. aureus</i> 0.40 log CFU g ⁻¹ (0.48% w/w)	NR	61
Starch	Hard (Cheddar cheese)	Carvacrol ^a	Lamination	Safety	<i>S. aureus</i> 0.99 log CFU g ⁻¹ (0.48% w/w)	NR	61
Alginate	Soft (Telemea cheese)	Lemongrass (citronella) ^a	Casting	Safety	<i>E. coli</i> O157:H7 0.70 log CFU g ⁻¹ (1.00% w/w)	NR	5
Starch	Soft (Commercial processed-filled cheeses)	Carvacrol ^c	Casting	Safety	<i>B. cereus</i> 12.40 log CFU g ⁻¹ (1.00% w/w)	NR	29
Foxtail millet starch	Soft (Queso blanco cheese)	Clove ^a	Casting	Safety	<i>L. monocytogenes</i> 1.19 log CFU g ⁻¹ (1.00% w/v)	NR	62
Chitosan	Pasteurized cow's milk	Cinnamaldehyde ^a	Casting	Safety	<i>L. monocytogenes</i> 3.87 log CFU mL ⁻¹ (1.00% w/v)	NR	63
Carboxymethyl cellulose	Soft (Paneer)	Clove ^a	Casting	Shelf life	NR	TMC: 4.53 log CFU g ⁻¹ (0.50% w/w) TPC: 6.80 log CFU g ⁻¹ (0.50% w/w) TYM: 1.27 log CFU g ⁻¹ (0.50% w/w)	64
Galactomannan	Semi-hard (Coalho cheese)	<i>Cymbopogon citratus</i> ^a	NR	Shelf Life	NR	TMC: 2.4 log CFU g ⁻¹ (0.50% v/v) TMC: 1.75 log CFU g ⁻¹ (1.00% w/v) TYM: 2.00 log CFU g ⁻¹ (1.00% w/v)	65
Sodium Caseinate-Chitosan	Soft (Panela cheese)	Oregano ^a	Casting	Shelf life	NR	TMC: 0.85 log CFU g ⁻¹ (1.00% w/w) TCC: 0.77 log CFU g ⁻¹ (1.00% w/w) LAB: 0.60 log CFU g ⁻¹ (1.00% w/w)	31
Film type: Composite							
Chitosan-whey protein	Soft (Feta cheese)	<i>Z. multiflora</i> ^a	Casting	Shelf life	TMC: 0.85 log CFU g ⁻¹ (1.00% w/w) TCC: 0.77 log CFU g ⁻¹ (1.00% w/w) LAB: 0.60 log CFU g ⁻¹ (1.00% w/w)	TMC: 0.85 log CFU g ⁻¹ (1.00% w/w) TCC: 0.77 log CFU g ⁻¹ (1.00% w/w) LAB: 0.60 log CFU g ⁻¹ (1.00% w/w)	30

NR: Not reported, TYM: Total yeast-mold, TVC: Total viable count, TMC: Total mesophilic bacteria, TPC: Total coliform bacteria, TCC: Total psychrophilic count, LAB: Lactic acid bacteria.
^{abc} indicate the form of EOs usage as free, nanoemulsion, and microemulsion, respectively.

Table 2. Different types of edible films incorporated with plant extracts applied in various dairy products.

Extract source (Type)	Film type	Product category/ Name	Film preparation method	Aim of use in food	Pathogen and reduction rate	Spoiler and reduction rate	Ref.
Film type: Protein							
<i>Lepidium sativum</i> (water/ethanol)	Fish gelatin	Soft (Ricotta)	Casting	Shelf life	NR	TMC: 1.90 log CFU g ⁻¹ (0.20% w/v) TPC: 0.98 log CFU g ⁻¹ (0.20% w/v)	45
Catechin – Gallic acid	Zein	Semi-hard (Kashar cheese)	Casting	Safety	<i>L. monocytogenes</i> 2.38 log CFU g ⁻¹ (3.00 mg cm ⁻²)	NR	66
Green Tea (<i>Camellia sinensis</i> L.) (ethanol)	Whey Protein	Soft (fresh goat cheese)	Casting	Shelf life	<i>E. coli</i> 1.22 log CFU g ⁻¹ (2.50% w/w)	TMC: 0.58 log CFU g ⁻¹ (2.50% w/w)	67
Green Tea (<i>Camellia sinensis</i> L.) (ethanol)	Whey Protein	Soft (mixture of fresh goat-sheep cheese)	Casting	Shelf life	<i>E. coli</i> 0.16 log CFU g ⁻¹ (2.50% w/w)	NR	67
<i>Moringa oleifera</i> Lam.leaf (ethanol)	Fish skin gelatin	Semi-hard cheese (Gouda)	Casting	Safety	<i>L. monocytogenes</i> 1.21 log CFU g ⁻¹ (1.00% w/v)	NR	68
Pomegranate peel (<i>Punica ganatum</i>) (water)	Zein	Soft (Fresh Himalayan cheese; Kalari/Kradi)	Casting	Shelf Life	NR	TBC: 6.00 log CFU g ⁻¹ (5.00% w/w) LAB: 1.60 log CFU g ⁻¹ (5.00% w/w) TYM:	69
Garlic-Thyme	Zein	Soft (mozzarella cheese)	Casting	Shelf Life	NR	4.25 log CFU g ⁻¹ (5.00% w/w) TMC: 2.52 log CFU g ⁻¹ (3.00% v/v) TYM: -3.50 log CFU g ⁻¹ (3.00% v/v)	70
Film type: Carbohydrate							
Rosemary (water)	Whey Protein	Soft cheese	Casting	Shelf Life	NR	TYM: 5.00 log CFU g ⁻¹ (2.00% w/w)	71
Sage (water)	Whey Protein	Soft cheese	Casting	Shelf Life	NR	TYM: 5.00 log CFU g ⁻¹ (2.00% w/w)	71
<i>Aloe vera</i> (<i>Aloe barbadensis</i> Miller) (water)	Carrageenan	Kulfi (Frozen dairy dessert)	Casting	Shelf Life	NR	TBC: 1.95 log CFU g ⁻¹ (15.00% v/v) TPC: 0.60 log CFU g ⁻¹ (15.00% v/v) TYM: 0.70 log CFU g ⁻¹ (15.00% v/v)	72
Purslane (<i>Portulaca oleracea</i>) (ethanol)	Chitosan	Soft (Mozzarella cheese)	Casting	Shelf life	NR	TBC: 0.39 log CFU g ⁻¹ (10.00% v/v) TYM: 1.65 log CFU g ⁻¹ (10.00% v/v)	73

Table 2 (continued).

Olive leaf (Olea europaea L.) (water)	Methylcellulose	Semi-hard (Kashar cheese)	Casting	Safety	<i>S. aureus</i> 1.22 log CFU cm ⁻² (1.50% w/v) <i>E. coli</i> O157:H7 1.21 log CFU g ⁻¹ (1.00% w/w) <i>L. monocytogenes</i> 0.85 log CFU g ⁻¹ (1.00% w/w)	NR	74
Grapefruit Seed (unknown)	Red algae	Sliced cheese	Casting	Safety		NR	75
Roselle calyx (ethanol)	Chitosan-guar gum	Hard (Ras cheese)	Casting	Shelf Life		TBC: 0.46 log CFU g ⁻¹ (1.00% v/v) TPC: 2.47 log CFU g ⁻¹ (1.00% v/v) TYM: 2.36 log CFU g ⁻¹ (1.00% v/v) <i>Streptococci</i> counts: 0.56 log CFU g ⁻¹ (1.00% v/v) <i>Lactobacilli</i> counts: 1.38 log CFU g ⁻¹ (1.00% v/v)	76
Film type: Composite							
Boldo (ethanol)	Gelatin-chitosan	Soft (Prato)	Casting	Shelf life		NR	77
Catechin - Gallic acid	Zein- wax	Semi-hard (Kashar cheese)	Casting	Safety	<i>L. monocytogenes</i> 2.54 log CFU g ⁻¹ (3.00 mg cm ⁻²)	NR	66
White tea (water)	Furcellaran- whey protein	Soft (Rennet- curd cheese)	Casting	Shelf life		TBC: 0.80 log CFU g ⁻¹ Yeasts count 3.50 log CFU g ⁻¹ Mold count: 1.60 log CFU g ⁻¹ (1.00% v/w) TBC: 0.90 log CFU g ⁻¹	78
Yerba Mate <i>Ilex paraguariensis</i> (water)	Furcellaran- whey protein	Soft (Rennet- curd cheese)	Casting	Shelf life		NR	78
Puerh tea (water)	Furcellaran- whey protein	Soft (Quark)	Casting	Shelf life		TBC: -0.30 Log CFU g ⁻¹ <i>Lactococcus</i> count -0.70 log CFU g ⁻¹ Yeast count: -1.10 log CFU g ⁻¹ (20.00% w/w)	46
Green tea (water)	Furcellaran- whey protein	Soft (Quark)	Casting	Shelf life		TBC: 0.60 log CFU g ⁻¹ <i>Lactococcus</i> count 0.00 log CFU g ⁻¹ Yeast count: -0.50 log CFU g ⁻¹ (20.00% w/w)	46

NR: Not reported, TYM: Total yeast-mold, TVC: Total viable count, TMC: Total mesophilic count, TCC: Total coliform bacteria, TPC: Total psychrophilic count, LAB: Lactic acid bacteria.

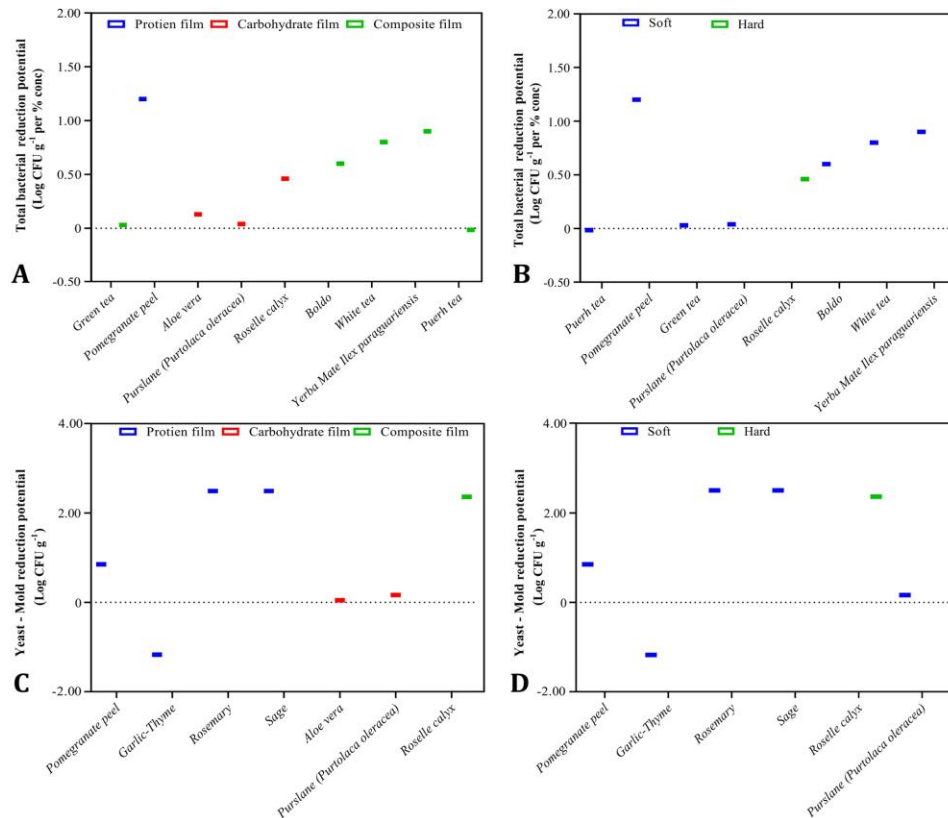


Fig. 6. Antimicrobial effect of extract against total bacterial count in cheese based on the **A)** type of film and **B)** type of cheese, and also against yeast-mold in cheese based on the **C)** type of film and **D)** type of cheese.

In conclusion, out of 77, 39 papers had inadequate information to be included in this review. Different findings were obtained on the use of PEOE in dairy products with respect to the type and concentration of PEOE, type of product, type of investigated pathogen and microbiota/mycobiota, type of film, and method of film development. Except for three cases, the remaining studies related to the topic of this article were in the cheese matrix. The main findings of this work are as follows:

- Zein was the major protein polymer used in the development of edible film containing PEOE for food application followed by whey protein; whereas, starch was among the most studied carbohydrate polymer.
- Most of studies carried out on the use of films containing PEOE concentrated on the semi-hard cheese.
- Concerning the film preparation method, solvent casting was the most common method for developing PEOE-based antimicrobial films, followed by electrospinning and lamination.
- Plant EOs were more effective than extract on food-borne pathogens and spoilers, and the effects of antimicrobial films with EO on yeast-mold were greater than mesophilic bacteria.
- The *L. monocytogenes*, *S. aureus*, *E. coli*, and *S. enteritidis* are among the major pathogens studied in the published literature.

- It appears that EOs containing thymol and carvacrol successfully inhibit the growth of bacterial species within the cheese matrix.
- The type and concentration are key parameters of the antimicrobial effectiveness of PEOE, followed by microbial strains and cheese type.

Therefore, it is extremely important to choose an appropriate film matrix for each type of dairy product and to select an effective concentration of PEOE without producing adverse effects on the sensory property of dairy products. The use of PEOE can be a good choice to increase the quality and safety of dairy products; but, due to the strong EOs aroma, sensory evaluation should always be considered. This problem can be resolved by using EOs with other antimicrobials having a synergistic effect and thus, requiring less EO for food preservation.

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Conflict of interest

Nothing declared.

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