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REVIEWS

IMPACT OF ESSENTIAL OILS ON THE CHARACTERISTICS OF WHITE BRINED CHEESE DURING PRODUCTION AND STORAGE: ADVANCES IN DAIRY SCIENCE

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ABSTRACT

The shift in consumer dietary habits has increased the demand for foods free of artificial preservatives, necessitating the exploration of various herbal extracts with antimicrobial activity and their application in the production and storage of brined cheeses. Essential oils (EOs), natural plant-derived compounds, are widely used in medicine, flavoring, and food preservation due to their proven antioxidant, antibacterial, and antifungal properties. This review investigates the role of EOs in the technological process and microbiological control during white brined cheese production, as well as their impact on cheese characteristics and starter cultures.

Keywords: *essential oils, natural preservatives, brined cheese*

INTRODUCTION

Traditional Bulgarian white brined cheese is produced from cow, sheep, goat, buffalo, or mixed milk. The physicochemical composition and physical properties of cow's milk, commonly used as a raw material for white brined cheese, must comply with the requirements outlined in Regulation (EU) No 1151/2012 of the European Parliament and the Council, dated November 21, 2012. Parameters such as protein content, fat percentage, non-fat dry matter, and lactose levels, as well as physical characteristics like hydration, density, and freezing point, are measured using ultrasonic milk analyzers. Active acidity (pH) is determined with a pH meter, while titratable acidity is assessed using the Thorner method. The titratable acidity, expressed as the volume of 0.1 M NaOH required for neutralization, is a key criterion reflecting both the primary composition and the secondary fermentation processes. These processes involve lactic acid bacteria converting lactose to lactic acid, depending on the storage temperature of the milk. Microbiological parameters of raw milk are evaluated according to Bulgarian State Standards (1).

The dynamics of lactic acid fermentation and the resulting acidification during cheese maturation can vary significantly between the 24th and 45th days of ripening. This process involves an increase in titratable acidity due to the metabolic activity of starter cultures. These cultures are optimized complexes of selected strains of lactic acid bacteria. In white brined cheese production, the starter cultures commonly include *Lactococcus lactis subsp. lactis* and *Lactobacillus casei*, along with symbiotic blends of *Lactobacillus delbrueckii subsp. bulgaricus* and *Streptococcus thermophilus*. The resulting cheese

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is subjected to microbiological, physicochemical, and organoleptic evaluations, with the acidification process monitored through titratable acidity measurements. Organoleptic assessments are conducted following BDS 15612:1983.

Dairy products are particularly prone to contamination due to their rich nutritional composition and origin as animal products. In dairy production, particularly in small-scale farm settings, numerous contamination sources have been identified, including raw milk, environmental factors, and, in some cases, the use of vegetable coagulants (2,3). Yeasts and molds play a significant role in dairy spoilage, often encouraged by excess moisture during ripening (4). Among bacterial species, *Listeria monocytogenes, Salmonella spp., Staphylococcus aureus,* and enteropathogenic *Escherichia coli* present the greatest risks to cheese safety.

According to European Community Regulation (EC) No 1333/2008 on food additives, preservatives are defined as "substances that extend the shelf life of foods by protecting them from deterioration caused by microorganisms and/or preventing the growth of pathogenic microorganisms"(5). Commonly used additives in food products include butylated hydroxyanisole, butylated hydroxytoluene, sulfites, nitrates, sorbates, benzoates, and other organic acids(6). However, consumer concerns regarding the safety of chemical preservatives and the negative perception of synthetic additives have spurred growing interest in natural alternatives, particularly plant-derived compounds (7). Recent studies (8,9,10,11) have highlighted the potential use and benefits of herbal extracts and essential oils (EOs) in the dairy sector, demonstrating the applicability of a wide range of natural compounds in various formulations.

This article reviews current research on the application of EOs in cheese production, focusing on their antimicrobial properties, mechanisms of action, and potential for cheese preservation. It also explores the challenges and limitations of using EOs as natural preservatives and identifies opportunities for future research.

MATERIALS AND METHODS

A systematic literature review was conducted using databases such as PubMed, Scopus, and Web of Science. Keywords included "essential oils,""natural preservatives," and "brined cheese."

RESULTS AND DISCUSSION

The growing demand for food products free from artificial preservatives has driven the use of EOs in the production and storage of brined cheeses. Essential oils, lipophilic liquids extracted from various plants, contain natural bioactive components with various properties. Known for their antimicrobial, antioxidant (12,13,14), and flavor-enhancing effects, EOs have shown promise as sustainable and effective agents for preserving cheese quality, extending shelf life, and improving safety (15,16).

Mechanisms of Action of Essential Oils

The antimicrobial properties of EOs against a broad spectrum of microorganisms have been demonstrated in numerous studies (17,18). Due to their hydrophobic components, EOs easily penetrate bacterial cell membranes through molecular transport mechanisms, causing membrane disruption and subsequent cell inactivation. Essential oils typically exhibit stronger inhibitory effects against Gram-positive bacteria compared to Gram-negative bacteria, due to the lipopolysaccharide barrier in the outer membrane of Gram-negative bacteria (19,20). The antimicrobial activity of EOs is thought to occur through several mechanisms:

Membrane disruption: The lipophilic nature of EOs allows them to integrate into microbial cell membranes, disrupting their structure and integrity. This leads to increased membrane permeability, leakage of cellular contents, and eventual cell death (21).

Enzyme inhibition: Some EOs inhibit key metabolic enzymes in microorganisms, including those involved in energy production, protein synthesis, or cell wall formation (22). This disruption significantly impairs microbial growth and reproduction.

Genetic and protein interference: Essential oils have been shown to affect the synthesis of nucleic acids and proteins in microorganisms, further contributing to their antimicrobial effects.

Impact of Essential Oils on Organoleptic and Physicochemical Properties of Cheese

The use of plants in cheese production is not a new practice, as it has traditionally been associated

with enhancing flavor or extending shelf life. Studies have shown that EOs can improve sensory properties and maintain cheese quality. For example, Ehsani A et al. reported that black cumin oil enhanced sensory attributes (texture, color, odor, taste, and overall acceptability) in white cheese, with treated samples outperforming the control(23). Similarly, Hassanein MFR et al. observed higher sensory scores for cheese supplemented with black cumin oil (24).

Asensio CM et al. (2015) found that oregano oil did not affect cheese ripening but prolonged shelf life by reducing the formation of organic acids such as lactic, formic, and acetic acids, as well as by slowing lipid oxidation and the breakdown of certain unsaturated fatty acids(25).

Mint oil was reported to increase protein content in white cheese and exert a mild beneficial effect on fat and moisture content during storage at low temperatures (26). Additionally, basil oil enhanced the antioxidant activity of cheese, reduced moisture levels, and preserved the content of unsaturated fatty acids and proteins (10,27).

Conversely, high concentrations of clove and tea tree oils required for significant antimicrobial activity resulted in strong undesirable flavors in cheese (28). Similarly, some EOs, including oregano oil, may impart a pronounced bitter taste to cheese at certain concentrations(29, 30).

Antimicrobial Activity of Essential Oils in Cheese

The antibacterial activity of natural compounds is typically evaluated against key pathogenic microorganisms frequently associated with cheese, such as *Listeria monocytogenes, Staphylococcus aureus, Escherichia coli, and Salmonella spp.*

Among the most effective plant-derived oils, black cumin seed oil, when incorporated into soft cheese, has demonstrated broad-spectrum antibacterial activity against all major pathogenic bacteria found in cheese (24).

Thyme and oregano EOs are among the most widely studied in this context. The primary active components of thyme EO, thymol and carvacrol, are responsible for its antimicrobial effects. In cheese, thyme oil has been shown to inhibit the growth of *Listeria monocytogenes*, *E. coli*, and molds contributing to spoilage (31). Oregano oil, which also contains high concentrations of carvacrol and thymol, is highly effective against various foodborne pathogens. Research indicates that oregano EO can extend the shelf life of cheese by preventing mold growth and a broad range of bacterial contamination (21).

Studies on the antibacterial efficacy of oregano and thyme EOs have established that a concentration of 0.2 mL/100 g is the minimum inhibitory concentration against *L. monocytogenes* and *E. coli* O157:H7 in cheese. Additionally, both oils exhibit a stronger inhibitory effect on *L. monocytogenes* compared to *E. coli* O157:H7 (12).

Similar investigations have been conducted with other EOs, such as rosemary and peppermint. Rosemary EO, which contains compounds like rosmarinic acid and eucalyptol, demonstrates both antioxidant and antimicrobial properties. It has been found to reduce the growth of spoilage bacteria, particularly in soft cheeses (32). The antimicrobial activity of peppermint oil is attributed to its main terpenoid compounds, carvone and limonene (33).

Clove EO, known for its high eugenol content, exhibits potent antimicrobial activity against both bacteria and fungi, making it a promising candidate for cheese preservation (34). Studies reveal that the addition of 1% clove and cinnamon EOs results in significant antimicrobial activity against *L. monocytogenes*. Bin et al. (2011) further evaluated the antibacterial efficacy of cinnamon, oregano, and clove oils against *Staphylococcus aureus, Listeria monocytogenes,* and *Salmonella enterica* in cheese at room temperature (35). Their findings show that all three EOs effectively inhibit the growth of these pathogens in cheese. Additionally, treatment with these oils improves cheese stability by protecting against lipid oxidation.

Among the tested oils, clove demonstrated the highest antibacterial and antioxidant activity. The researchers concluded that clove, cinnamon, and oregano EOs have significant potential as natural food preservatives.Table 1 summarizes the reported antimicrobial activities of EOs used in cheese formulations as documented in the literature.

Influence of Essential Oils on Starter Cultures and Cheese Acidification

One of the main concerns regarding the use of plant-based compounds as natural preservatives

Table 1. Essential oils and their antimicrobial activity in cheese preservation.

Essential Oil	Pathogenic Bacteria Inhibited	Main Active Compounds
Thyme	*Staphylococcus aureus*, *Escherichia coli*, *Salmonellaspp.*	Thymol, carvacrol
Oregano	*Escherichia coli*, *Salmonellaspp.*, *Listeria monocytogenes*	Carvacrol, thymol
Cinnamon	*Escherichia coli*, *Salmonellaspp.*, *Listeriaspp.*, *Campylobacterspp.*	Cinnamaldehyde, eugenol
Clove	*Escherichia coli*, *Salmonellaspp.*, *Staphylococcus aureus*	Eugenol, acetyleugenol
Lemon	*Escherichia coli*, *Listeria monocytogenes*, *Salmonellaspp.*	Limonene, citral
Peppermint	*Escherichia coli*, *Salmonellaspp.*, *Listeriaspp.* , *Campylobacterspp.*	Menthol, menthone
Basil	*Escherichia coli*, *Salmonella*, *Listeriaspp.*	Eugenol, methyl chavicol
Rosemary	*Escherichia coli*, *Salmonellaspp.*, *Listeria monocytogenes*	Carnosic acid, rosmarinic acid
Garlic	*Escherichia coli*, *Salmonellaspp.*, *Listeriaspp.* , *Staphylococcus aureus*	Allicin, diallyl disulfide

is their potential to inhibit the activity of the natural microbial population or artificially added starter microorganisms in cheese. Reduced activity of lactic acid bacteria during the ripening period can lead to alterations in flavor, slow acidification, and excessive growth of undesirable microorganisms (11).

However, numerous studies suggest that lactic acid bacteria are relatively resistant to the antimicrobial effects of EOs, with their growth typically not being significantly affected (36). Research evaluating the impact of oregano (37), thyme, and basil EOs(38) on cheese production reports that EOs have little to no effect or exert only a mild to moderate influence on the growth and acidification activity of lactic acid bacteria in milk. These studies even suggest improvements in the microbiological quality of cheese during ripening.

In Bulgaria, studies have also been conducted on fruits and EO derived from caraway (*Carum carvi*), which possess proven antimicrobial properties. The primary components of caraway oil are carvone and limonene, with smaller amounts of dihydrocarvone, dihydrocarveol, and perilla aldehyde. These antimicrobial properties explain the traditional use of caraway fruits in folk medicine (39). In the food industry, caraway fruits are widely used as a seasoning for sauces, sausages, meat preserves, baked goods, various cheeses, and curds.

Experimental data indicate slower acidification in brined cheese when caraway oil or fruits are added. This trend persists throughout the observation period, with acidity levels remaining within the technological requirements for white brined cheese production (200–270°T).

These findings highlight that while caraway oil and fruits exhibit antimicrobial properties, they do not significantly disrupt the development of lactic acid bacteria populations. The active fermentation process is maintained, resulting in a product with excellent organoleptic characteristics and compliance with technological standards.

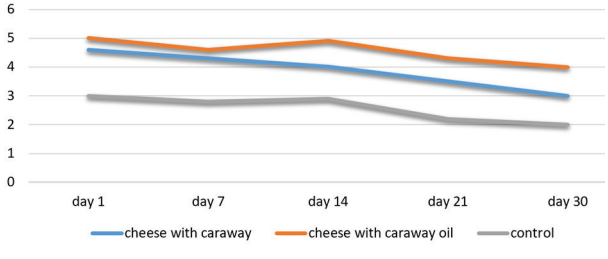


Fig. 1. Dynamics of lactic acid bacteria development in cheese with caraway fruits, caraway essential oil, and control sample.

Influence of Essential Oils on Starter Cultures and Cheese Acidification

According to researchers, the slower lactic acid fermentation observed in white brined cheese with added caraway fruits or caraway EO is attributed to their antimicrobial properties. The impact of caraway fruits and EO on the development of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* revealed a higher tolerance in the *Streptococcus* population compared to *Lactobacillus* throughout the study. Despite this, the addition of caraway EO and fruits did not exert a significant negative effect on the activity of lactic acid bacteria. The fermentation process proceeded effectively, resulting in a product with excellent organoleptic characteristics (40,41). Essential oils demonstrate significant potential as natural antimicrobial and antioxidant agents. Their study as bioactive compounds, supported by scientific evidence, enhances their potential for the development of powerful, eco-friendly, and safer food preservatives, offering an alternative to synthetic chemicals.

Application of Essential Oils in Cheese Production

The use of EOs in cheese preservation can take several forms, including incorporation into the cheese matrix, surface coating, or as an additive to milk during the production process:

Incorporation into the cheese matrix: Essential oilscan be added directly to the cheese or introduced into the milk before coagulation. This ensures that the antimicrobial properties of the EOs are evenly distributed throughout the cheese.

Surface coating: Another approach involves spraying or brushing the cheese with EOs after shaping and before packaging. This creates a protective layer on the cheese surface, preventing microbial contamination and spoilage (3).

Encapsulation: To enhance stability and control the release of EOs, encapsulation technologies such as nanoemulsions and liposomes have been explored (22,42). Encapsulation can help prevent the evaporation of volatile compounds and ensure prolonged antimicrobial activity throughout the cheese's shelf life (43,44).

Challenges and Limitations

While EOs offer promising antimicrobial properties, their use as cheese preservatives presents several challenges:

Sensory Impact: Essential oils are highly aromatic and can significantly alter the taste and aroma of cheese. Strong flavors, such as those of clove or cinnamon oils, may not be desirable in all cheese types.

Regulatory issues: The use of EOs in food products is subject to regulatory approval in many countries. Ensuring that EOs are safe for consumption at the required concentrations is essential for their acceptance in the food industry.

Essential oils are recognized as Generally Regarded as Safe (GRAS) by the FDA . Their use in food, including cheese, is legally approved in the United States (45,46). In Europe, however, a different legal framework applies. Essential oils are not classified as additives, and their use in food is not permitted, although many EO components (e.g., limonene) are approved as flavorings (47,48). Consequently, under current regulations, EOs cannot be included in cheese and cheese products in the European Union, limiting the range of new products that could be commercialized(11).

Effectiveness in different cheese types:The antimicrobial efficacy of EOs can vary depending on the type of cheese, its moisture content, and overall composition. Some cheese varieties may require higher concentrations of EOs to achieve the desired preservative effect.

Future Research Directions

Future studies on the use of EOs as cheese preservatives should focus on optimizing their application methods and concentrations to minimize sensory impact while maximizing antimicrobial efficacy. Additionally, the development of novel delivery systems, such as microencapsulation, could improve the stability and controlled release of EOs. Exploring the combination of EOs with other natural preservatives, such as bacteriocins or plant extracts, may also provide synergistic effects to enhance cheese preservation.

Further research is needed to assess the long-term effects of EO usage on the nutritional quality of cheese, including their impact on beneficial microbi-

al communities, such as probiotics, which are often present in fermented cheeses.

CONCLUSION

The application of EOs as natural preservatives in cheese production offers a promising alternative to synthetic chemicals. Essential oils demonstrate significant antimicrobial properties, helping to prevent spoilage and extend shelf life without compromising the safety or nutritional quality of the cheese. However, challenges related to sensory impact, regulatory approval, and effective application methods must be addressed before EOs can gain widespread acceptance in cheese production.

Ongoing research into the use of EOs, combined with innovative delivery systems and potential synergistic effects with other natural preservatives, will play a critical role in developing sustainable cheese preservation techniques. The use of EOs such as thyme, oregano, rosemary, clove, caraway, and others in the formulation of white brined cheese or in brine for its preservation has demonstrated normal lactic acid fermentation processes and resulted in products with excellent organoleptic characteristics. Applying diverse technologies and combined strategies for incorporating EOs in various suitable forms facilitates their use across a broader range of food products.

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