

Antimicrobial Effect of *Cuminum Cyminum* Essential Oil on Iranian White Cheese in Air and

Running title: Antimicrobial effect of Cuminum cyminum oil on White soft cheese

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Abstract

BACKGROUND: As one of the most crucial food categories, nutritionists commonly recommend dairy products. Since these products are highly perishable, it is important to find a method to increase the shelf life of them and conserve their freshness for a long time.

OBJECTIVES: The present study aims to assess the antibacterial properties of *Cuminum cyminum* essential oil (CCE) on the quality of refrigerated white cheese preserved through packing under gas mixtures (air, modified atmosphere packaged (MAP): 70% O₂ – 30% CO₂).

METHODS: The mesophilic bacteria (TMC), psychrotrophic bacteria (PTC), lactic acid bacteria (LAB), Enterobacteriaceae, *Listeria monocytogenes*, and mold and yeast counts were determined using PCA, DCRB, PCA, MRS agar, violet red bile agar, and PALCAM agar, respectively, during 35 days of storage period

RESULTS: The results revealed that the growth rate of TMC and LAB, Enterobacteriaceae, mold and yeast, PTC, and *Listeria monocytogenes* considerably decreased in white cheese samples as a result of the integration of CCE and MAP. The lowest number was observed in a case with samples packed in MAP+ 0.06 % CCE after 35 days of storage.

CONCLUSIONS: Given the microbial characterization improvements, CCE was determined to be an optimal alternative, along with MAP, for applications in white cheese.

Keywords: Antibacterial, *Cuminum cyminum*, Essential oil, Cheese, Shelf life.

Introduction

Cheese is one of the most commonly-used food products worldwide since it can be produced from a wide range of milk types and with various technologies, people can achieve considerable product varieties. People have shown an increasing inclination to consume this product due to its scrumptious, great protein level, and being perceived as a healthy food (Gouvea *et al.*, 2017).

However, this food product easily contaminates and spoils by pathogenic microorganisms, which has adverse effects on shelf life, and jeopardizes the consumer's health. It was documented *Listeria monocytogenes*, *Staphylococcus aureus*, and *Salmonella* spp. are often associated with food-borne diseases due to the cheese consumption (Gouvea *et al.*, 2017).

White cheese is manufactured by using different kinds of milk. This product has constantly been present in human diet, as it is rich in protein, calcium, minerals, and vitamins. Cheese consumption has experienced a dramatic rise worldwide during the past several years. Cheese's physical, sensory, color, and chemical properties have been proven to be the result of biological and biochemical reactions taking place throughout storage.

White cheese is rather susceptible to contamination and spoilage by pathogenic microorganism-induced contamination, which is likely to lead to decreased shelf life and a serious threat to the health of human beings. The bacteria, molds, and yeasts are usually responsible for this type of contamination, leading to a decrease in flavor and the quality of cheese. This usually occurs when the product is stored without packing (H.S. El-Sayed; S.M. El-Sayed, 2021).

Using modified atmosphere packaging (MAP) has gained more popularity to increase the shelf life of various foods, as a result of increased demands on the part of consumers for preservative-free, "clean label" foods. Modifying the gas content surrounding a food product during storage, including N₂ and CO₂, will result in comparatively lower physiological deterioration, oxidation reactions, and microbial growth rate (Brown *et al.*, 2017).

Presently, various preservation methods have promoted the shelf storage of food products, the most promising of which is packaging. Certain important functions are attributed to the packaging process, including prevention of deterioration by microbial and chemical changes and development in handling and marketing of packaged goods. Presently, the purpose of food packaging is not only for convenience and protection attributes but also for many other applications (Khoshgozaran *et al.*, 2012). Modified atmosphere packaging has gained a significant position in research areas as a pragmatic method to preserve the quality of various food products and satisfy customers' growing demands for fresh and preservative-free food (Khoshgozaran *et al.*, 2012). Moreover, this technique is characterized by several crucial outcomes, including retaining the quality of fresh products, promoting the visual and appearance

properties of the product, extending the shelf life, and minimizing the application of additives and preservatives (Khoshgozaran *et al.*, 2012).

In recent years, the application of natural antimicrobial agents for food preservation has gained wide acceptance due to the public unpopularity of synthetic additives, which were of frequent use to inhibit microbial proliferation in food products. Essential oils (EOs) are extracted from medicinal plants known to have significant antimicrobial activity against various pathogenic and spoilage microorganisms. (M. Artiga-Artigas *et al.*, 2017).

Cuminum cyminum is a small, herbaceous, annual plant that belongs to the Umbelliferae family (Petretto *et al.*, 2018). This plant is found in Asia, North Africa, Europe, and America and has also been cultivated in Middle East countries, India, China, and the Mediterranean countries (Petretto *et al.*, 2018; Akrami *et al.*, 2015). The seeds of this plant are constantly used as a flavoring spice in various recipes belonging to different cultures (Petretto *et al.*, 2018), particularly in cooking and making salads (Karimirad *et al.*, 2019). Moreover, different varieties of this plant are of extensive use in both traditional and veterinary medicine as stimulant, carminative, astringent, and as a treatment for indigestion, flatulence, and diarrhea (Akrami *et al.*, 2015; Derakhshan *et al.*, 2008). CCE has high levels of γ -terpinene, p-cymene, pinene, cuminaldehyde, safranal, and cuminal with antimicrobial and antioxidant properties (Karimirada *et al.*, 2019). *Cuminum cyminum* essential oil has an appropriate antimicrobial and antioxidants activity that can be applied as a suitable food preservative agent (Petretto *et al.*, 2018).

The aim of the current study was to assess (i) the combined effect of CCE and MAP for the control of *L. monocytogenes* inoculated to white Iranian white cheese and (ii) the possible shelf life extension of white Iranian white cheese using the mentioned combination.

Materials and Methods

Plant Material

Cuminum cyminum seeds were collected from Kerman, Iran, in the summer of 2020. Taxonomic identification of plant material was carried out by the Institute of Medicinal Plants, Medical University of Tehran, Iran.

Essential oil Extraction and analysis

In the preparation phase, 100 g of powdered seeds were mixed with 1000 mL of distilled water. The CCE was obtained via the 'Clevenger apparatus', for 3 hours. Dehydration of CCE was done by the addition of sodium sulfate. The collected CCE was stored in dark glass at 4°C for further analysis (karimirad *et al.*, 2019; Akrami *et al.*, 2015). The Gas chromatography–mass spectrometry (GC–MS) analysis was performed according to the method described by Akrami *et al.*, 2015.

Preparation of test microorganisms

Listeria monocytogenes (ATCC1918) was inoculated in Brain Heart Infusion (BHI) broth. After 24h incubation at 35°C, the second subculture was prepared and incubated for 24 h at 35°C. The *L. monocytogenes* broth culture was placed in a sterile cuvette and optical density (OD) was adjusted an absorbance of 0.1, using a spectrophotometer (Jenway, UK). Then the number of cells in the suspension was estimated by duplicate plating from 10-fold serial dilutions on BHI agar and counting the colonies after 24h incubation at 35°C and then the suspension was diluted to 4 logs CFU/ml using 0.1% peptone water Preparation of white cheese.

In order to produce of Iranian white cheese, fresh and whole cow's milk was applied, which was pasteurized at a temperature of $72 \pm 2^\circ\text{C}$ for 15 seconds. Before starting the different stages of cheese- making, the temperature of the milk was raised to 35 °C, and 10 liters of milk was added to the cheese making container. After that, the starter in the amount of 0.5% (V/V) was added to the milk samples at the same time. After half an hour, the amount of 0.02% (weight by volume)

of calcium chloride (2CaCl) (weight by volume) in 20 ml of sterile distilled water at a temperature of 40 °C was added to the milk. Finally, the amount of 0.001% (W/V) rennet was added and the temperature of the milk is maintained at around 35°C during the time of clot formation. After the passage of one hour, the formed clot was cut into 1-2 cubic cm pieces and according to the instructions for making Iranian white cheeses, it was put under 10 kg pressure for six hours to absorb water. Then, the clot was cut into pieces with dimensions of 6 x 8 x 12 cm and placed in 2% sterile salt water for 42 days at a temperature of 4 °C (Abasifar *et al.*, 2016). Immediately after spraying, all blocks were packaged in plastic trays and sealed by a sealing machine. Finally, the samples were air packaged and gas-flushed with 30% CO₂ + 70% N₂, sealed using a MAP machine, and stored at 4 °C, so the tests would be carried out on day 35 (Govaris *et al.*, 2011).

Microbiological characteristics of white cheese throughout storage

25 g of cheese samples were homogenized with 225 ml of sterile tri-sodium citrate (2.00% w/v) for 1 min. after that, decimal dilution with 9 ml sterile NaCl (0.85%) was performed.

The microbiological properties of white cheese samples were determined as follows:

LAB count was enumerated by using MRS agar, Yeasts, and mold counts were determined on Rose Bengal Chloramphenicol agar, TMC, and PSB was estimated on plate count agar, *L. monocytogenes* bacteria counts were determined using PALCAM Listeria Selective Agar (H. S. El-Sayed and El-Sayed, 2021).

Statistical analysis

The experimental data were analyzed by Variance (ANOVA) and the significant differences between mean values in different sampling days were evaluated by Duncan's Multiple Range Test/least significant difference. Data analysis was performed using the SPSS version 14.0 for Windows, SPSS Inc, Chicago, IL, USA.

Results

Chemical Composition of *Cuminum cyminum* essential oil

The analysis of CCE revealed EO yield is 4% (v/w). Among all individual constituents identified by GC/MS, 1, 4-p-Menthadien-7-al and cumin aldehyde stand as the two major compounds with 32.20% and 29.57% percentages, respectively (Table 1).

Microbiological characteristics of white cheese during the storage period

In general, compared to control samples, all coatings in this experiment exhibited significant antibacterial activity against TMC, PSB, LAB, *Enterobacteriaceae*, and *L. monocytogenes* in white cheese packed in air and MAP during refrigerated storage (Figs 1-5).

Comparatively, higher bacterial counts were present in groups packed in air compared to those packed in MAP ($P < 0.05$). It can be seen from Figs. 1 and 2 that the initial TMC and LAB of white cheese samples were found to be 4.5 and 4.32 log CFU/g, respectively. The TMC and LAB of control samples constantly increased and reached 7.53 and 7.54 log CFU/g, after 35 days of storage in air packaging, respectively. Figs (1, 2). The TMC and LAB of control samples continuously increased and reached 7.22 and 6.1 log CFU/g after 35 days of storage in MAP packaging, respectively. Packing conditions determined the growth of TMC and LAB. The lowest TMC and LAB belonged samples packed in MAP+ 0.06 % CCE was 6.26 and 5.51 log CFU/g, respectively.

In the present study, PSB count in the control group was found to increase from an initial count of 5.43 log CFU/g to 7.9 log CFU/g at the end of chilled storage in air packaging and an initial count of 5.43 log CFU/g to 6.09 log CFU/g at the end of chilled storage in MAP packaging. The smallest PSB count belonged to samples packed in air +0.06 % CCE (6.72) after 35 days of storage. The lowest PSB count was obtained for samples packed in MAP+0.06 % CCE (5.72) after 35 day of storage. Fig (3).

In the present study, mold and yeast counts in the control group were increased from an initial count of 6.77 log CFU/g to 8.37 log CFU/g at the end of chilled storage in air packaging and an initial count of 6.77 log CFU/g to 7.73 log CFU/g at the end of chilled storage in MAP

packaging. The lowest mold and yeast count belonged to samples packed in air +0.06 % CCE after 35 days of storage. The lowest mold and yeast count was achieved in the case with samples packed in MAP+0.06% CCE after 35 days of storage. Fig (4).

In the present study, *Listeria monocytogenes* count in the control group was found to increase from an initial count of 4.22 log CFU/g to 8.23 log CFU/g at the end of chilled storage in air packaging and an initial count of 4.22 log CFU/g to 6.98 log CFU/g at the end of chilled storage in MAP packaging. The lowest *Listeria monocytogenes* count was determined to belong to samples packed in air +0.06 % CCE (6.87 log CFU/g) after 35 days of storage. The lowest *Listeria monocytogenes* count was achieved in the case with samples packed in MAP+0.06 % CCE after 35 days of storage. Fig (5).

No *coliforms* were detected in any batch of cheese produced for the shelf life experiments.

Discussion

The analysis of used CCE revealed that 1, 4-p-Menthadien-7-al and cumin aldehyde stand as the two major compounds of this essential oil, respectively (Table 1). This finding is in agreement with those reported by Karimirad *et al.*(2019) who reported cumin aldehyde with 23.6 % and γ -Terpinen-7-al with 22.23% are the major components of CCE; Petretto *et al.*, 2018 noted twenty-five compounds were identified with γ -Terpinen-7-al being the major component in CCE; Derakhshan *et al.*, 2008 who declacumin aldehyde with 25.2%, p -mentha-1,3-dien-7-al with 13% and p -mentha-1,4- dien-7-al with 16.6%); and H.S. El-Sayed and S.M. El-Sayed., 2021 noted Cumin aldehyde (30.9%), sabinene (14.3%), p-cymene (13.3%), γ -terpinene (12.6%), cuminyl Alcohol (11.5%), p-cymen-7-ol (8.8%).

According to certain studies, the quality and quantity of certain essential oils can be affected by factors such as harvesting season, geographic location, soil conditions, and essential oil extraction technique (Kalemba and Kunicka, 2003; Kizil *et al.*, 2010).

Furthermore, as reported by Eikani *et al.* (1999), cumin aldehyde and cuminyl alcohol have potent antimicrobial and antioxidant properties with an extensive range, and as a result, may be

considered an appropriate candidates for preserving agents, and are promising to the food industry.

Monoterpenes compounds, such as cumin aldehyde, were reported to be responsible for the antimicrobial activity of *Cuminum cyminum* EO. The antibacterial effect of EO plants may be associated with the hydrophobicity nature of constituent's EO especially oxygenated monoterpenes and their capability of disrupting the lipid layer of the cell membrane and interacting with membrane proteins and intracellular targets of microorganisms. This phenomenon is believed to be able to change the bacterial phospholipid membrane and, as a result, reduce cellular uptake of ethidium bromide, while increasing leakage of potassium ions, ATP, and carboxyfluorescein (Kakaei and Shahbazi , 2016).

Hydrocarbon derivatives have been reported to have a low antimicrobial function when used alone. It is evident that their low water solubility and limited hydrogen- bound capacity are responsible for this phenomenon. However, the compounds mentioned before can potentiate the activity of terpenoids such as cumin aldehyde, γ -terpinen7-al and γ -terpinene (Monoterpene aldehydes) which exhibit a higher antimicrobial potential owing to their functional groups. For instance, aldehyde moiety by amino groups can link with DNA and proteins and interfere with their normal function. Furthermore, it has been stated that hydrocarbon monoterpenes like p-cymene promote the entrance of other compounds into the cell wall via swelling of the cell membrane (karimirad, *et al.*, 2019).

Regarding crucial foods and pathogens, only a limited number of studies have been conducted on this essential oil.

De *et al.* (2003) examined the antimicrobial properties of some Indian spices and investigated the antimicrobial properties of *Cuminum cyminum* against the microbes tested (*Bacillus subtilis*, *Escherichia coli*), resulting in the approval of its usage as a disinfectant food preservative.

In another study, Chaudhry Ahmed *et al.* (2008) examined the antibacterial properties of extracts of a few plants such as Shab-kur, *Cuminum cyminum*, and Poppy against 188 species of bacteria such as *Bacillus subtilis*, *Saccharomyces cerevisiae*, and *E. coli*, via disk diffusion method (DDM). They concluded that Green Cumin extract had the greatest inhibitory effect (73%) on bacteria.

According to Sadeghi *et al.* (2008), CCE was reported to contain 29.02% Cumin Aldehyde, 20.70% Alpha-Terpinene, and 12.94% Gamma-Terpinene. It was also observed that the growth rate of *Staphylococcus aureus* can significantly be reduced in cheese during 75 days of storage compared to the control group, by the addition of 30 ml/ 100µl of CCE.

In another study, the antimicrobial effects of CCE against 10 bacterial strains, belonging to 8 different species, and 6 yeast strains from 4 species, were examined by Petretto *et al.* (2018). LAB were found to exhibit great resistance to all EO tested while the CCE showed a strong antifungal activity that affected both maximum specific growth rate and lag time.

Dairy products are categorized as the most popular food which are highly suggested by nutritionists. This food category is critically perishable; therefore, it is crucial to increase its shelf life for being fresher. As a result of consumers' increased knowledge about the threats of preservatives, technologists, and researchers have attempted to introduce novel preservative-free methods, one of which is modified atmosphere packaging (MAP), which modifies the natural gas surrounding the product in the package to delay deteriorative changes.

Since CO₂ inhibits a variety of spoilage and pathogenic microorganisms, especially gram-negative bacteria and molds, it is usually employed in MAP. Inhibition of microbial growth by CO₂ has been provided in MAP of perishable foods which improves the shelf life. However, CO₂ inhibition activity against microbial growth increases under chilled conditions, since CO₂ is more soluble in food at lower temperatures (Lee *et al.*, 2008). In addition to controlling and to retard microbial growth, the presence of CO₂ in the package's headspace also leads to the change in the microbial content to bacteria with lower spoilage capacity (McMillin, 2008). Low temperatures during storage help this capacity.

Several studies have confirmed the effect of MAP in the development of the shelf life of dairy products, especially cheese, and a variety of gas compositions has been suggested for MAP of cheese. The storage stability of 24-month-old portioned-packed *Parmigiano Reggiano* cheese, packed in nylon/polyethylene bags and stored for 3 months at 4 °C was examined by Romani *et al.* (1999). No particular change occurred in the quality of different packed products, though samples packed in a 100 % N₂ atmosphere exhibited flavor profiles quite distant from freshly cut, unpacked cheese. The fungal growth and mycotoxin production on commercial sliced cheddar cheese under modified atmospheres was examined by Taniwaki *et al.* (2001). Eight

fungal species were incubated under conditions of decreasing levels of O₂ (5% to packed in aluminum foil and modified atmospheres (100% N₂, 30% CO₂/70% N₂, 50% CO₂/50% N₂, 70% CO₂/30% N₂, 100% CO₂, 30% CO₂/60% N₂/10% O₂, 70% CO₂/20% N₂/10% O₂ using oriented polyethylene polyamide as the packaging material) as well as in vacuum. The proliferation of coli bacteria was observed in the experimented cheese in aluminum foil, while the population of the coli group remained unchanged in other samples during the storage period, irrespective of the applied gas mixture.

In another relevant study, the shelf life of Mozzarella cheese was examined by Alam and Goyal (2007) in different atmospheres (air, vacuum, 100% CO₂, 100% N₂, and 50% N₂/50% CO₂) packed in high-barrier bags and stored at -10 to -15 °C. According to their observations, Mozzarella cheese under MAP showed a significant increase in its shelf life compared to that of kept in a conventional air package (14–16, 90, 75, and 65 days under air, 100% CO₂, 50% N₂/50% CO₂, and 100% N₂, respectively).

In the current study, the PSB count in the control group increased from 5.43 log CFU/g to 7.9 log CFU/g at the end of chilled storage in air packaging and an initial count of 5.43 log CFU/g to 6.09 log CFU/g at the end of storage period in MAP. It was demonstrated by Alves *et al.* (1996) that MAP (100% CO₂) was able to decrease just the beginning of the PSB growth in Mozzarella cheese, which was similar to other studies (Pintado and Malcata 2000 on Requeijão cheese; Gammariello *et al.* 2009a on Apulian fresh cheeses). Moreover, Eliot *et al.* (1998) reported the presence of PSB during the first weeks of storage for the products with the modified atmospheres (10%, 25%, 50%, 75%, 100% N₂) since PSB is a complicated population and species in Mozzarella cheeses are resistant to CO₂ inhibitory effect. In addition, various storage temperatures (10 °C by Moir *et al.* 1993 on Cottage cheese; 7 °C by Alves *et al.* 1996 on Mozzarella cheese; 10 °C by Eliot *et al.* 1998 on Cameros cheese; 4 °C by Gonzalez-Fandos *et al.* 2000 on Cameros cheese) have been evaluated as a possible contributor means for MAP in controlling PSB growth, and applying low temperatures were found as an effective way in combination with MAP. Due to lower temperatures, a higher inhibitory effect of CO₂ can be achieved by higher CO₂ solubility.

These findings are in line with the studies noted for whey cheese (Dermiki *et al.*, 2008), fresh goat cheese (Gonzalez-Fandos *et al.*, 2000; Olarte *et al.*, 2002), and Mozzarella (Alam and

Goyal, 2011) cheese, where psychrotolerant growth was lower when the CO₂ concentration increased, with 100% CO₂ conditions was the most effective in the growth inhibition. These findings are in agreement with the article which showed that most psychrotolerant bacteria in dairy products are aerobic, gram-negative bacteria, that are usually more sensitive to CO₂ than gram-positive ones (Rosenthal *et al.*, 1991).

In the present study, mold and yeast counts in the control group were found to increase from 6.77 log CFU/g at the beginning of the study to 8.37 log CFU/g after 35 days of storage at chilled storage in air packaging and an initial count of 6.77 logs CFU/g to 7.73 logs CFU/g at the end of refrigerated storage in MAP packaging. The lowest mold and yeast count belonged to samples packed in air +0.06 % CCE (7.7) after 35 days of storage. The inhibition and decrease in yeast and mold population under modified atmospheres compared with growth when packaged under air were confirmed by the previous reports for Mozzarella kept under the same conditions (Alam and Goyal, 2011). The controlling effect of CO₂ on the count of bacteria and yeast is also in line with the previous reports (Alves *et al.*, 1996; Eliot *et al.*, 1998; Dermiki *et al.*, 2008).

Since the 1970s, *Listeria monocytogenes* has been considered an important food-borne pathogen. Foods such as vegetables, meat, and even certain kinds of cheeses are ideal for *Listeria* growth (Genigeorgis *et al.*, 1991; Mossel *et al.*, 1995). Outbreaks of listeriosis following the consumption of Mexican-style cheese from California (James *et al.* 1985), Mexican soft cheeses (Linnan *et al.* 1988), and Vacherin Mont D'Or (Bille, 1990) have led the concerns toward *L. monocytogenes*. The inhibitory effect of CO₂ on *L. monocytogenes* was reported by Chen and Hotchkiss (1991), especially in a hurdle technology with cold-temperature storage (4 °C) and pH in Cottage cheese. In the present study, *Listeria monocytogenes* count in the control group increased from 4.22 log CFU/g to 8.23 log CFU/g at the end of the study period in air packaging and an initial count of 4.22 log CFU/g to 6.98 log CFU/g at the end of refrigerated storage in MAP. According to the study of Brown *et al.*, 2018 which evaluated the effect of modified atmosphere packaging on *Listeria monocytogenes* on fresh cheese, the mean of *L. monocytogenes* counts increased to levels significantly higher than inoculation on cheeses stored under MAP conditions.

Enterobacteriaceae are suggested as an indicator of fecal contamination within the analysis of food, including zoonotic bacteria such as *Salmonella* spp., *Yersinia* spp., and *Escherichia coli*.

Enterobacteriaceae can cause serious infections in humans, while many of the major important members of the genus of this family are resistant to many of the available antimicrobials (Paterson, 2006). Coliforms had not isolated in any batch of produced Irania white cheese in this study.

Conclusion

The result of the current investigation indicated that the combination of CCE and MAP condition had a strong antimicrobial effect against the *Listeria monocytogenes* and extended the shelf life. Accordingly, MAP+0.06% CCE exhibited the best inhibitory effects on the microbial population.

Ethical Considerations

Compliance with ethical guidelines

There were no ethical considerations to be considered in this research.

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بررسی اثر ضد میکروبی اسانس زیره سبز در پنیرسفید ایرانی بسته بندی شده به صورت معمولی و
اتمسفیر اصلاح شده طی مدت زمان نگهداری در یخچال

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چکیده:

زمینه مطالعه: متخصصان تغذیه به عنوان یکی از مهم ترین دسته های غذایی معمولاً محصولات لبنی را توصیه می کنند. از آنجایی که این محصولات بسیار فاسد شدنی هستند، یافتن راهی برای افزایش ماندگاری و تازه نگه داشتن آنها برای مدت طولانی بسیار حیاتی است.

هدف: مطالعه حاضر با هدف بررسی خواص ضد باکتریایی اسانس زیره سبز (CCE) بر کیفیت پنیر سفید در شرایط بسته بندی معمولی و در شرایط اتمسفیر اصلاح شده (70 درصد CO₂ و 30 درصد O₂) در دمای یخچالی است.

روش کار: تعداد باکتری های مزوفیل، کپک و مخمر، باکتری های سرماگرا، باکتری های اسیدلاکتیک، انتروباکتریاسه و لیستریا مونوسیٹوژنز به ترتیب با استفاده از پلیت کانت آگار، دی کلران رزبنگال آگار، پلیت کانت آگار، MRS، VRBA و پالکام آگار تعیین شد.

نتایج: نشان داد که سرعت رشد تعداد باکتری های مزوفیل و تعداد باکتری های اسید لاکتیک، انتروباکتریاسه، کپک و مخمر، باکتری های سرماگرا و لیستریا مونوسیٹوژنز در نمونه های پنیر سفید در نتیجه مصرف همزمان اسانس زیره سبز و اتمسفر اصلاح شده به میزان قابل توجهی کاهش یافت. کمترین تعداد در مورد نمونه های بسته بندی شده در MAP+ 0.06 % CCE پس از 35 روز نگهداری مشاهده شد.

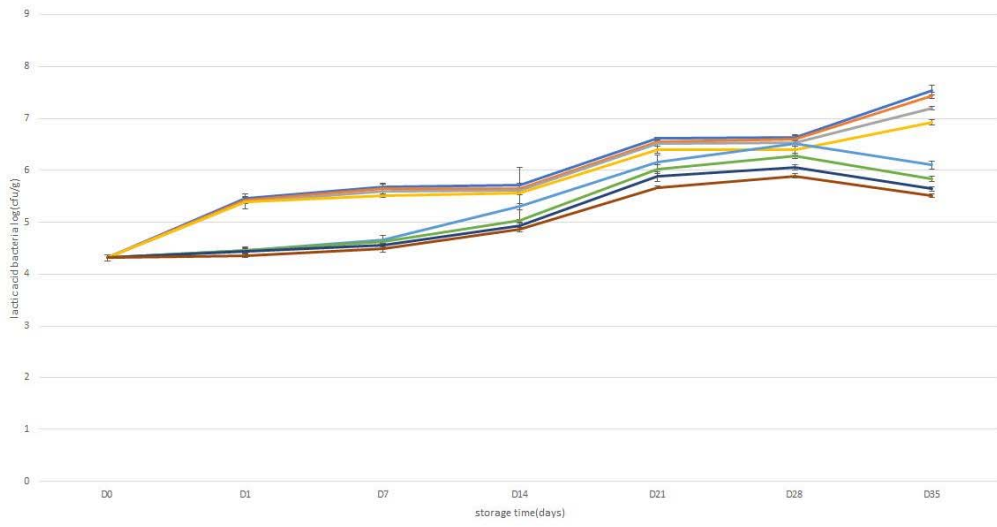
نتیجه گیری نهایی: با توجه به بهبود خصوصیات میکروبی، اسانس زیره سبز به عنوان یک جایگزین بهینه به همراه اتمسفر اصلاح شده برای کاربرد در پنیر سفید تعیین شد.

کلمات کلیدی: ضد میکروبی، اسانس، زیره سبز، پنیر، زمان نگهداری

Table 1. Composition of *cuminum cyminum* l. essential oil identified by gas chromatography-mass spectrometry

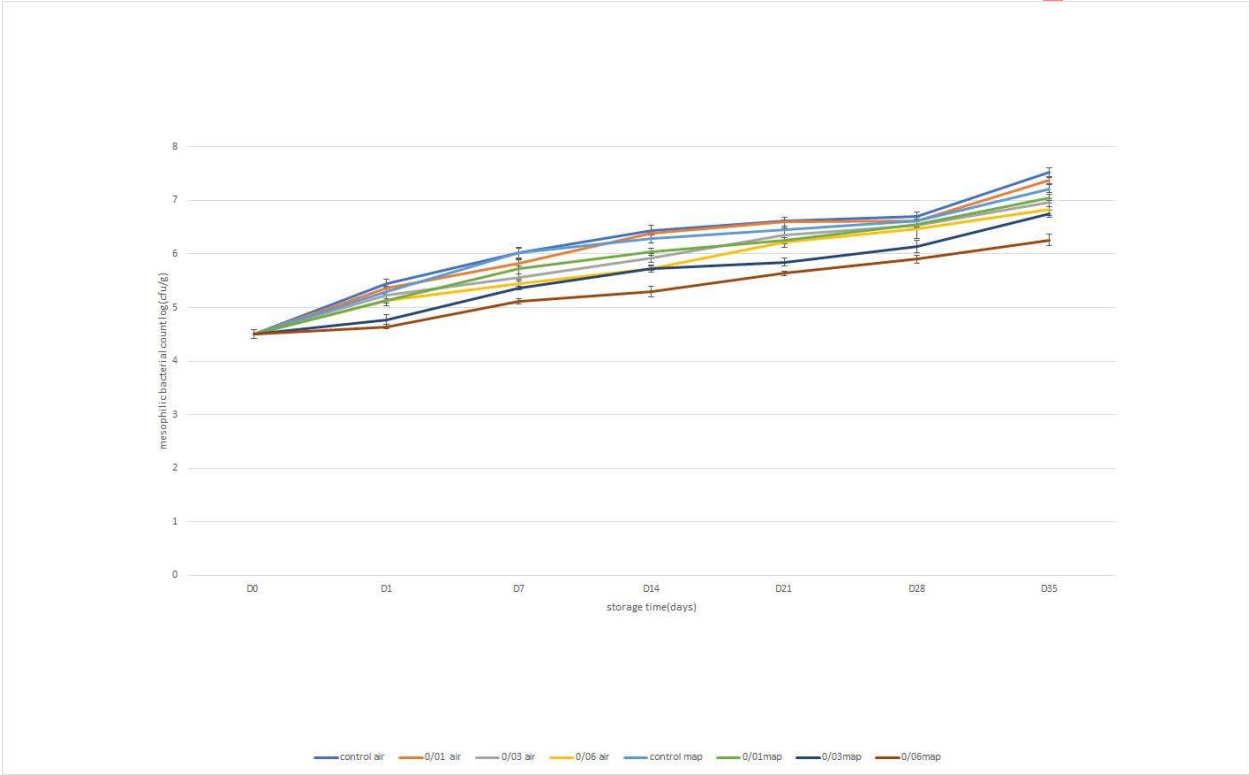
No	RT (min)	Area%	Name	Quality
1	553.6	14.0	alpha-thujene	91
2	771.6	26.0	2-Pinene	97
3	042.8	20.0	Sabinene	96
4	224.8	16.5	beta-Pinene	97
5	748.8	31.0	beta-Myrcene	95
6	204.9	20.0	1-Phellandrene	97
7	915.9	46.5	Cymene	95
8	128.10	13.0	1,8-Cineole	92
9	581.11	11.10	gamma-Terpinene	97
10	153.13	02.0	delta,3-Carene	91
11	684.14	04.0	3-Methyl-2,4-hexadiene	76
12	349.16	19.0	4-Terpineol	96
13	738.16	88.0	3-Cyclopentylcyclopentan-1-one	83
14	935.16	02.0	Isoterpinolene	72

15	25.19	57.29	Cuminaldehyde	98
16	185.21	50.14	2-Caren-10-al	72
17	067.22	20.32	1,4-p-Menthadien-7-al	78
18	084.23	03.0	Myrtenal	76
19	249.26	03.0	Alloocimene	76
20	02.29	05.0	Trans-.beta.-Farnesene	55
21	466.29	13.0	UNKOWN FROM LIMEN OIL	93
22	255.30	03.0	cis-2-Methylenehexahydroindan-7-one	38
23	636.32	02.0	2-Methoxybenzyl alchohol	64
24	238.33	06.0	Carotol	90

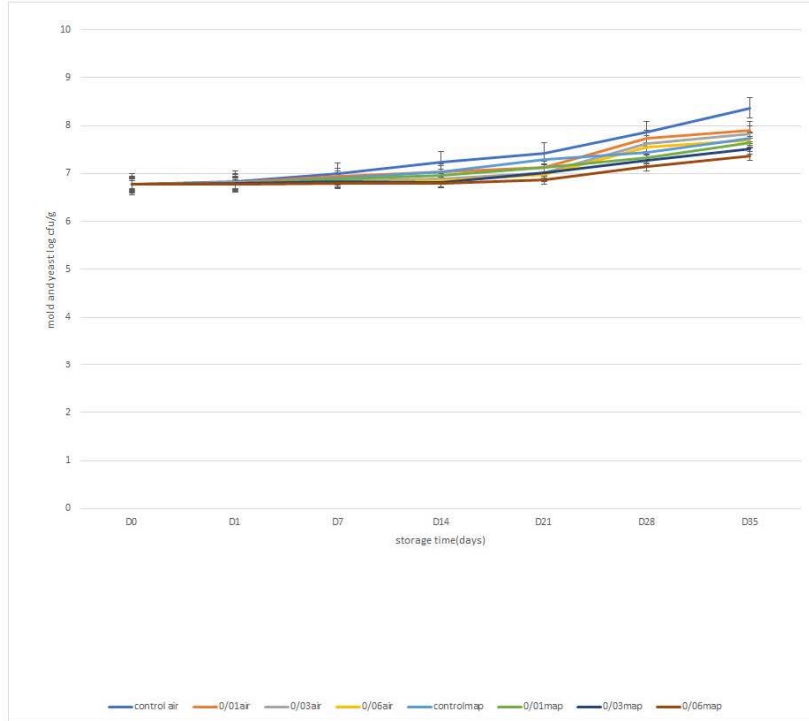


control air 0/01 air 0/03 air 0/06 air control map 0/01 map 0/03 map 0/06 map

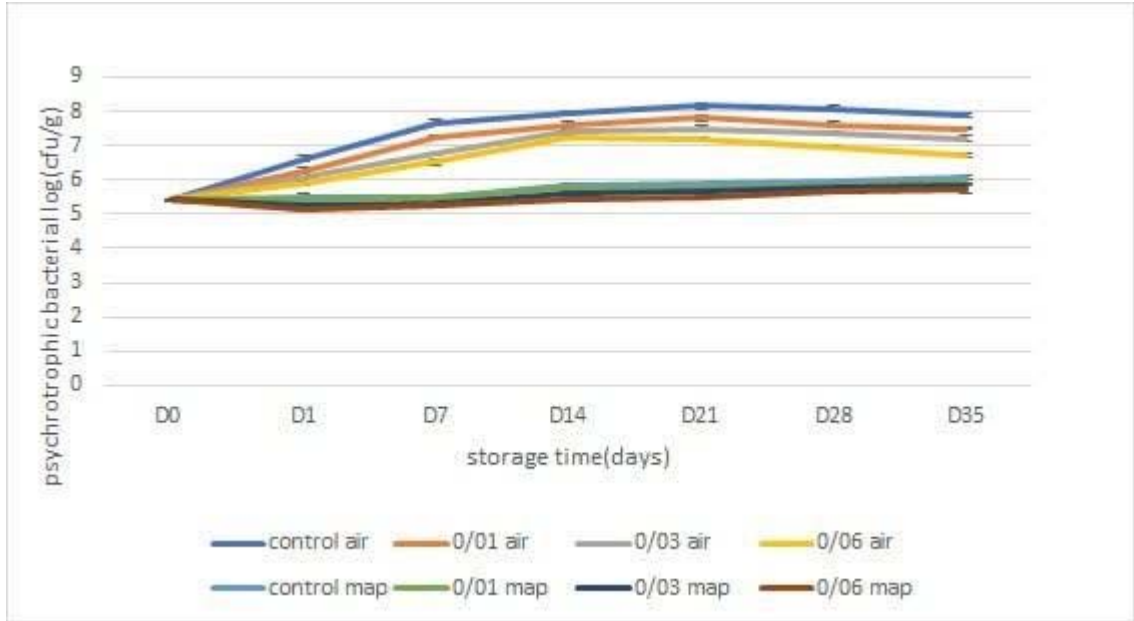
Uncorrected



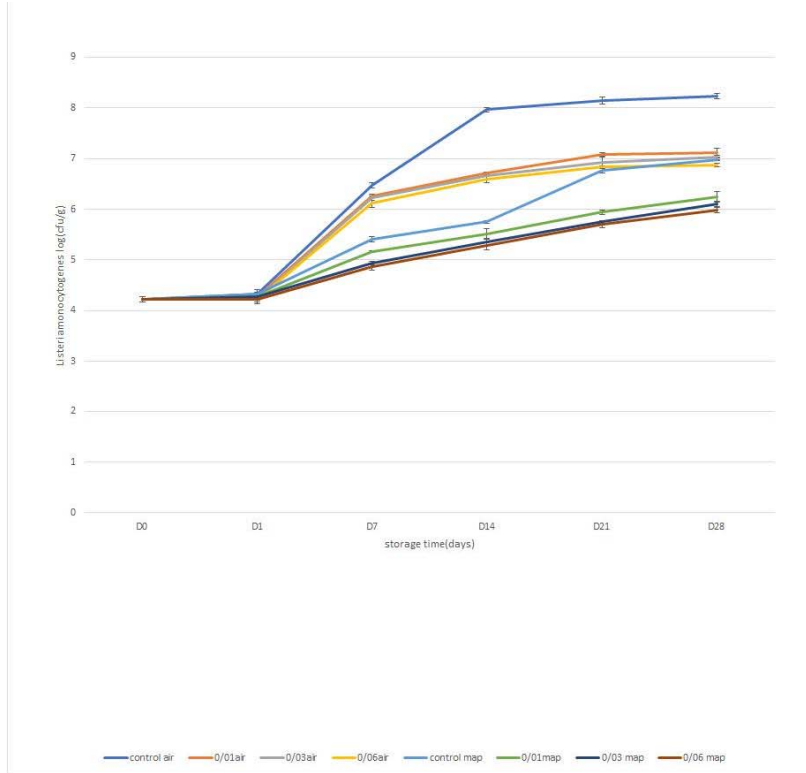
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