

## The Effect of Lemongrass as a Natural Antioxidant and Antimicrobial Agent on the Physicochemical Properties and Shelf Life of Mayonnaise

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**ABSTRACT:** This study aimed to explore the impact of lemongrass as a natural antioxidant and antimicrobial agent on the physicochemical properties and shelf life of mayonnaise. In the present study, mayonnaise samples were formulated with the incorporation of lemongrass essential oil at concentrations of 0%, 0.5%, 1%, and 1.5%. These samples were subsequently stored for a duration of two months. The investigation encompassed several characteristics of the produced samples, including pH, acidity, peroxide value, anisidine value, total oxidation (tox), thiobarbituric acid content, viscosity, stability, biphasicity, as well as a comprehensive sensory evaluation. The sensory assessment included parameters such as flavor, odor, texture, color, and overall acceptance. Additionally, microbiological analyses were conducted to determine the total bacterial count and the total mold and yeast count. The findings indicated that the pH levels of all samples ranged from 3.79 to 3.98, while the acidity levels varied from 0.51 to 0.96. Throughout the storage period, a decline in pH levels was observed, accompanied by an increase in acidity. The increase in the concentration of essential oil within the samples corresponded to a significant reduction in the intensity of lipid oxidation in mayonnaise. This change notably inhibited the upward trend of all associated indices, encompassing peroxide, anisidine, thiobarbituric acid, and tototox levels, with the most pronounced effect observed in the T<sub>3</sub> treatment group. The viscosity, biphasic formation, and stability observed in the T<sub>3</sub> treatment exhibited a lesser degree of reduction in comparison to the other samples that incorporated lemongrass essential oil, as well as the control sample. The mayonnaise sample containing 1.5% lemongrass essential oil (T<sub>3</sub>) exhibited a significantly greater inhibitory effect on the increase of total bacterial count as well as total mold and yeast count compared to the other samples analyzed. The sensory evaluations assessing taste, color, texture, and overall acceptance revealed that the samples containing 0.5% and 1% concentrations of lemongrass essential oil, along with the control sample, received the lowest scores. Conversely, the highest ratings were attributed to the sample containing a 1.5% concentration of lemongrass essential oil. The evaluators assessed the aroma and olfactory scores of samples containing 1% and 1.5% lemongrass essential oil (T<sub>3</sub>, T<sub>4</sub>) and in conclusion reached the decision that, the incorporation of lemongrass essential oil at a concentration of 1.5% (T<sub>3</sub>) has been shown to enhance the physicochemical, sensory, microbial, and shelf-life properties of mayonnaise.

**Keywords:** Antioxidant, Lemongrass Essential Oil, Mayonnaise, Physicochemical Properties.

### Introduction

Mayonnaise is among the most extensively consumed condiments globally and is esteemed for its distinctive and creamy flavor profile. Numerous strategies have been proposed to enhance the

nutritional profile of mayonnaise. Fat reduction represents the most widely employed approach within this domain. A contemporary methodology entails the fortification of emulsion sauces with bioactive compounds, specifically antioxidants, aimed at addressing the health-related requirements of individuals

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(Flamminii *et al.*, 2020). There is an increasing trend within the food industry to utilize plant extracts as antimicrobial agents and as alternatives to synthetic antioxidants. This practice is aimed at preventing lipid oxidation, combating foodborne pathogens, and extending the shelf life of food products. Medicinal plants serve as a vital source of bioactive natural products, as endorsed by the World Health Organization. Natural antioxidants possess the capacity to mitigate or avert oxidative damage to lipids. Thus, the integration of these compounds into fat-based food products serves as an effective strategy for mitigating the deterioration of product quality associated with lipid oxidation (Tometri *et al.*, 2020). Since ancient times, humans have extensively utilized a variety of herbs and spices to enhance the flavor and aroma of food, as well as to leverage their medicinal properties and preservative qualities. The advantageous effects of plants on health can be predominantly attributed to the presence of polyphenolic compounds within their chemical compositions. Other volatile compounds found in plants, particularly essential oils, may also play a significant role in the biological activity of these organisms, thereby exerting a health-promoting effect on human populations (Ranjah *et al.*, 2022). Mayonnaise, similar to other foods that contain fats, is prone to spoilage due to the autoxidation processes that affect unsaturated and polyunsaturated fatty acids. During the process of autoxidation, external energy sources, including light, interact with unsaturated fatty acids in the presence of catalytic agents, such as iron, resulting in the generation of free radicals. The structural composition of mayonnaise is characterized by a significant proportion of oil being dispersed within the aqueous phase, which may also encompass

dissolved oxygen. Furthermore, during the mixing process, air bubbles are generated and subsequently entrapped within the structure of the emulsion. The presence of air bubbles and oxygen can serve as catalysts for the process of autoxidation (Balali & Rahman, 2023).

Citratum, a member of the Gramineae family, is a medicinal plant that is recognized internationally under the common name lemongrass. The prefix "lemon" denotes a scent reminiscent of lemon, which can be attributed to the presence of citral within the compound. Citral is classified as a monoterpene, a type of organic compound characterized by a molecular structure consisting of two isoprene units. Lemongrass is comprised of various phytochemical constituents, notably including tannins, flavonoids, and alkaloids. This species is a perennial herbaceous plant, capable of reaching heights of up to one meter. It exhibits numerous leafy stems that originate from a compact rhizomatic root system (Sehajpal *et al.*, 2023).

Lemongrass possesses notable antioxidant and antimicrobial properties, rendering it beneficial in the management of a variety of diseases. The essential oil derived from the lemongrass plant predominantly consists of citral isomers, specifically alpha-citral and beta-citral, which collectively account for approximately 61% of the oil's composition. The antimicrobial properties of lemongrass are attributed to the presence of citral within its chemical composition (Melo *et al.*, 2020).

## Materials and Methods

### - *Preparation and Formulation of Mayonnaise*

In the present study, the preparation of each treatment involved the production of mayonnaise following a set of specific

protocols. Initially, water, powdered ingredients, and eggs were introduced into a mixing vessel. Subsequently, the residual oil was incorporated following an exhaustive mixing of the dry ingredients for a duration of two minutes. Subsequently, the oil was introduced gradually in a dropwise manner, followed by a continuous application in a thin layer over the course of seven minutes. Subsequent to the formation of the emulsion and the achievement of an appropriate texture, vinegar was incrementally incorporated into the mixture. Lemongrass essential oil was integrated into the sauce formulation at varying concentrations of 0.5%, 1%, and 1.5% and finally, the sample was subjected to homogenization utilizing a high-speed homogenizer set at 1000 revolutions per minute for a duration of five minutes. It is important to highlight that a total of one kilogram of the sample was prepared for each treatment condition. The final product underwent testing on days one, thirty, and sixty. The formulation of treatments examined in this study are presented in Table 1.

**Table 1.** Treatments Studied

Treatment Number	Treatment Conditions
	Percentage of extracted lemongrass extract (w/w%)
T <sub>0</sub>	0
T <sub>1</sub>	0.5
T <sub>2</sub>	1
T <sub>3</sub>	1.5

- **Mayonnaise Tests During Storage**
- **Peroxide Value Measurement**

Mayonnaise oil was extracted utilizing normal hexane as the solvent. A quantity of five grams of oil was accurately measured and placed into a 250 ml Erlenmeyer flask. Subsequently, 30 ml of a solution comprised of acetic acid and chloroform, in a ratio of 3:2, was added to

the flask and the resulting mixture was thoroughly homogenized. Subsequently, 0.05 milliliters of a saturated potassium iodide solution was administered and allowed to remain in darkness for a duration of one minute. Upon removal from the dark environment, 30 mL of distilled water was introduced to the sample. Subsequently, the solution was titrated using 0.1 N sodium thiosulfate until the cessation of the yellow coloration was observed. During the titration process, the mixture was subjected to vigorous stirring to facilitate the separation of iodine from the chloroform layer. Subsequently, 0.5 milliliters of the starch indicator reagent was introduced, and the titration process was carried on until the disappearance of the blue coloration was observed. In conjunction with the sample analysis, a control titration was conducted utilizing a mixture of acetic acid and chloroform devoid of oil. Ultimately, the peroxide value was determined utilizing the subsequent formula and was expressed in milliequivalents of peroxide per 1000 grams of oil (Shahidi & Zhong, 2005).

$$\text{Peroxide Value} = \frac{(V_2 - V_1) \times N \times 1000}{m}$$

where,

V<sub>2</sub> = volume of sodium thiosulfate utilized for the sample

V<sub>1</sub> = volume of sodium thiosulfate employed for the control

N = normality of the thiosulfate

M = mass of the oil (gr)

- **Anisidine Index**

The anisidine index was determined using the AOSC (18-90 CD) method. The p-anisidine number is defined as the absorbance measured of a solution prepared with 1 gram of oil or fat dissolved in 100 milliliters of isooctane, which is supplemented with p-anisidine. In

order to prepare a 25% p-anisidine solution, 25 grams of p-anisidine were dissolved in 100 milliliters of glacial acetic acid. Subsequently, 1 gram of the sample was precisely weighed and introduced into a volumetric flask. The sample was then dissolved in isooctane, and the solution was brought to the desired volume. Isooctane was employed as a control to establish a baseline for absorbance measurements. The absorbance of the solution containing the sample was quantified using a spectrophotometer at a wavelength of 350 nm. Subsequently, 5 mL of the prepared solution containing the sample was transferred into a test tube, followed by the addition of 1 mL of the prepared p-anisidine reagent. Five milliliters of isooctane and one milliliter of p-anisidine were subsequently introduced into the second test tube. After a duration of 10 minutes, the absorbance at 350 nm was measured, utilizing the solution in the second test tube as a baseline (set to zero) and comparing it to the absorbance of the solution in the first test tube.

$$(\text{Anisidine value}) AV = 25(1.2A_s - A_b) / M$$

where,

$A_s$  = light absorption by the fat solution after reacting with anisidine reagent.

$A_b$  = light absorption in fat solution.

$M$  = weight of fat sample (g).

#### - **Thiobarbituric Acid**

A sample weighing 10 grams was combined with 25 milliliters of distilled water, followed by the addition of 25 cubic centimeters of chloroacetic acid. Subsequent to the filtration of the resultant mixture, 4 milliliters of the filtered solution were combined with 1 milliliter of a TBA solution (concentration: 0.06 M) and subjected to a boiling water bath for a duration of 10 minutes. Subsequently, the

test tubes containing the sample were allowed to equilibrate at room temperature for a duration of five minutes. The results were quantified and reported as milligrams of malondialdehyde equivalents per kilogram. ("Journal of the American Oil Chemist's Society," 1994)

#### - **Totox Index**

The Totox index serves as an indicator of total oxidation, encompassing both primary and secondary oxidation products. The calculation of this index is conducted in accordance with the Iranian National Standard Number (2007) and is guided by the following formula: (Institute of Standards and Industrial Research of Iran (ISIRI), 2017)

$$\text{Totox index} = (\text{Peroxide value} \times 2) + \text{Anisidine value}$$

#### - **Total Mold and Yeast Counts**

Total mold and yeast count was performed according to the Iranian national standard. For this purpose, ten grams of the mayonnaise mixture were mixed under sterile conditions with 0.1% peptone water broth, and dilutions of  $10^{-1}$  and  $10^{-2}$  of the desired suspension were prepared. Fifteen to twenty milliliters of Rose Bengal Chloramphenicol DRBC agar culture medium were inoculated with 1 ml of the desired dilution in a sterile plate and incubated for 3 to 5 days at 25°C. After this time, the number of colonies was counted and reported (Institute of Standards and Industrial Research of Iran (ISIRI), 2013).

#### - **Total Bacterial Count**

For the purposes of microbiological testing, a sample weighing 1 gram was introduced under sterile conditions into a tube containing 9 milliliters of sterile peptone water (0.1%), followed by

thorough homogenization of the mixture. Various dilutions of  $10^{-2}$ ,  $10^{-3}$ , and  $10^{-4}$  were systematically prepared and subsequently inoculated onto King agar medium. The plates were incubated for a duration of 48 hours at a temperature of 37°C. The quantified bacterial counts were expressed as logarithmic values of colony-forming units per gram (log CFU/g). (Institute of Standards and Industrial Research of Iran (ISIRI), 2022)

#### - pH

It was carried out in accordance with the Iranian National Standard No. 2454.(Iran., 2023)

#### - Acidity

Fifteen grams of the sample were thoroughly mixed in 200 mL of distilled water that had been neutralized against phenol until a homogeneous solution was obtained. Subsequently, the mixture was titrated with 0.1 M sodium hydroxide in the presence of phenol reagent. The National Standard of Iran was published in 2014.(Iran., 2023)

Acidity in terms of percentage by weight of acetic acid =  $(100 \times 0.006 \times A)/S$

where,

A = volume of NaOH used

S = sample weight in grams

#### - Viscosity

In order to ascertain the rheological properties of mayonnaise samples, a Brookfield II-DV RV viscometer (USA) was employed. Consequently, shaft number 7 was employed for this purpose. To evaluate the degree of thickness, a specified volume of the test solution (500 mL) was transferred into a 600 mL container. Subsequently, the shaft was immersed into the solution until it reached the designated check line. At that juncture,

the consistency of the tests was systematically assessed at a temperature of 25 °C across a range of shaft rotational speeds, specifically at 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 105, 120, 135, 140, 160, 180, and 200 revolutions per minute (rpm) (Beigi *et al.*, 2022a).

#### - Stability and Biphasic Assessment

The assessment of mayonnaise soundness was conducted on the primary day, as well as on the 30th and 60th days. A sample weighing 25 grams was introduced into a centrifuge tube and subjected to centrifugation at 3000 revolutions per minute (rpm) for a duration of 10 minutes. At this stage, the centrifuged samples were subjected to an incubation process at a temperature of 50°C for a duration of 48 hours. Subsequently, the stability of the emulsion was assessed and expressed as a percentage, utilizing the following equation.(Beigi *et al.*, 2022b)

$$\text{Emulsion stability} = (\text{Initial sample weight} / \text{Centrifuge weight}) \times 100$$

#### - Sensory Evaluation

The sensory characteristics, including color, taste, odor, and texture, were evaluated employing a 5-point hedonic scale by a cohort of ten untrained evaluators. Each evaluator completed an assessment survey for each treatment on the day of production, as well as 30 days and 60 days post-production, all conducted at a controlled temperature of 20°C. The assessment criteria were organized on a scale ranging from one to five, with the following corresponding descriptions: 1 = Strongly Dislike (extremely weak), 2 = Dislike Slightly (somewhat ineffective), 3 = Neutral (moderately effective), 4 = Like Moderately (considerably effective), and 5 = Strongly Like (extremely effective). The final evaluation list constituted the

overarching assessment criteria. In the current investigation, sampling was performed at each predetermined testing interval and for each treatment condition, with all experiments conducted in triplicate to ensure methodological rigor. Sampling was conducted on the first day subsequent to the completion of the production process, as well as on the 30th and 60th days after production (Khalkhali & Noveir, 2018).

#### - *Statistical Analysis*

The means of each parameter were analyzed using a factorial arrangement within a completely randomized design, utilizing the Statistical Package for the Social Sciences (SPSS) version 22. 0 for data analysis. Differences among treatment groups were analyzed utilizing Duncan's multiple range test, with a confidence level of 95%. The resultant data were subsequently plotted using Microsoft Excel 2013.

### **Results and Discussion**

Figure 1 illustrates a notable increase in the peroxide value across all treatments throughout the storage period. The peroxide value serves as an indicator of the concentration of hydroperoxides and the early products generated during the process of lipid oxidation. During the process of oxidation, the concentration of hydroperoxides initially experiences an increase followed by a subsequent decline. This pattern is indicative of their sequential formation and subsequent degradation. Mayonnaise is classified as an oil-in-water emulsion, characterized by the presence of an oil phase interacting with a significant surface area of water. This structural composition renders it particularly vulnerable to oxidative degradation. Consequently, the duration of storage correlates with an upward trend in

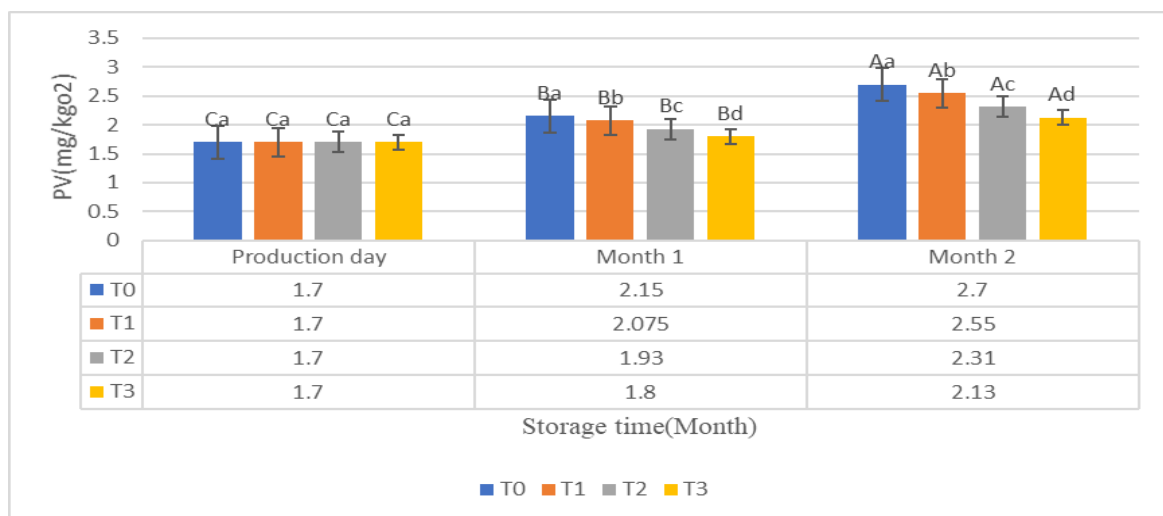
the peroxide value of mayonnaise samples, indicative of the progressive enhancement of oxidative processes (Gorjian *et al.*, 2021). Furthermore, the application of lemongrass essential oil substantially curtailed the progression of peroxide value. Notably, a concentration of 1. 5% lemongrass essential oil exhibited the most minimal increase in peroxide value at the conclusion of the second month of storage. The mayonnaise formulation incorporating 0. 5% lemongrass essential oil did not exhibit a statistically significant difference when compared to the control treatment. The findings indicate that a concentration of 0. 5% lemongrass essential oil does not possess the capacity to inhibit the oxidative processes occurring in mayonnaise. The findings indicated that the incorporation of lemongrass essential oil into the mayonnaise formulations resulted in a reduction of peroxide value. This phenomenon can be elucidated by the high concentrations of essential oil exhibiting antioxidant properties. As the concentration of the essential oil escalated, there was a corresponding increase in the levels of phenolic compounds, which subsequently led to an enhancement of pro-oxidant effects (Qasemzadeh *et al.*, 2023).

In oxidized oils, peroxides function as unstable intermediates that undergo decomposition, resulting in the formation of various carbonyl compounds. The principal oxidation products exhibit characteristics of being both colorless and tasteless. In contrast, the secondary oxidation products are responsible for the development of off-odors in food products. In order to assess the final stage of the oxidation process in mayonnaise, it is advisable to measure the anisidine index (Balali & Rahman, 2023).

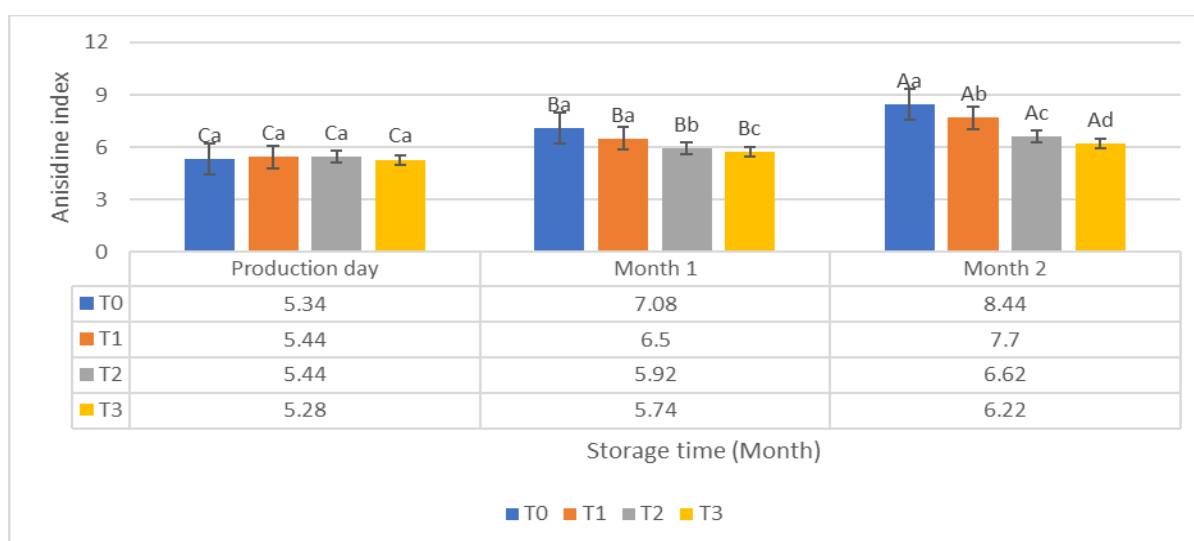
The findings of the current investigation, as depicted in Figure 2,

indicate a significant elevation in the anisidine index across all treatment conditions throughout the storage duration. The significant increase in the anisidine index observed in all mayonnaise samples during the storage period can be attributed to the degradation of hydroperoxides and the resultant rise in the production of secondary oxidation compounds. This phenomenon can be attributed to the inherent instability of the primary products resulting from lipid oxidation. These primary products undergo decomposition, which ultimately results in the formation of secondary oxidation products, including aldehydes and ketones. As the storage period progresses and the rate of hydroperoxide decomposition accelerates, there is a corresponding incremental increase in the formation of secondary products. The anisidine index serves as a quantifiable indicator of secondary products resulting from lipid oxidation. Consequently, an escalation in the formation of secondary oxidation products correlates with a corresponding rise in the anisidine index (Balali & Rahman, 2023). Furthermore, the application of lemongrass essential oil demonstrated a significant reduction in the increase of

anisidine number. Specifically, a concentration of 1.5% lemongrass essential oil exhibited the most pronounced inhibition of anisidine number rise at the conclusion of the second month of storage. The findings suggest that a concentration of 0.5% lemongrass essential oil does not effectively inhibit the oxidation process in mayonnaise. During the storage period, a significant increase in the anisidine index was observed across all mayonnaise samples. This elevation can be attributed to the degradation of hydroperoxides and the subsequent rise in the generation of secondary oxidation compounds. The findings indicate that the incorporation of lemongrass essential oil into the formulation of mayonnaise treatments resulted in a reduction of the anisidine index. This phenomenon can be elucidated by the presence of antioxidant properties inherent in the essential oil at elevated concentrations. As the concentration of the essential oil increased, the rate of increase in the peroxide value diminished. This observation suggests that the initial stages of oxidation and the formation of hydroperoxides were somewhat inhibited.



**Fig. 1.** Comparison of the average peroxide index during two months of storage at a significance level of 0.05.



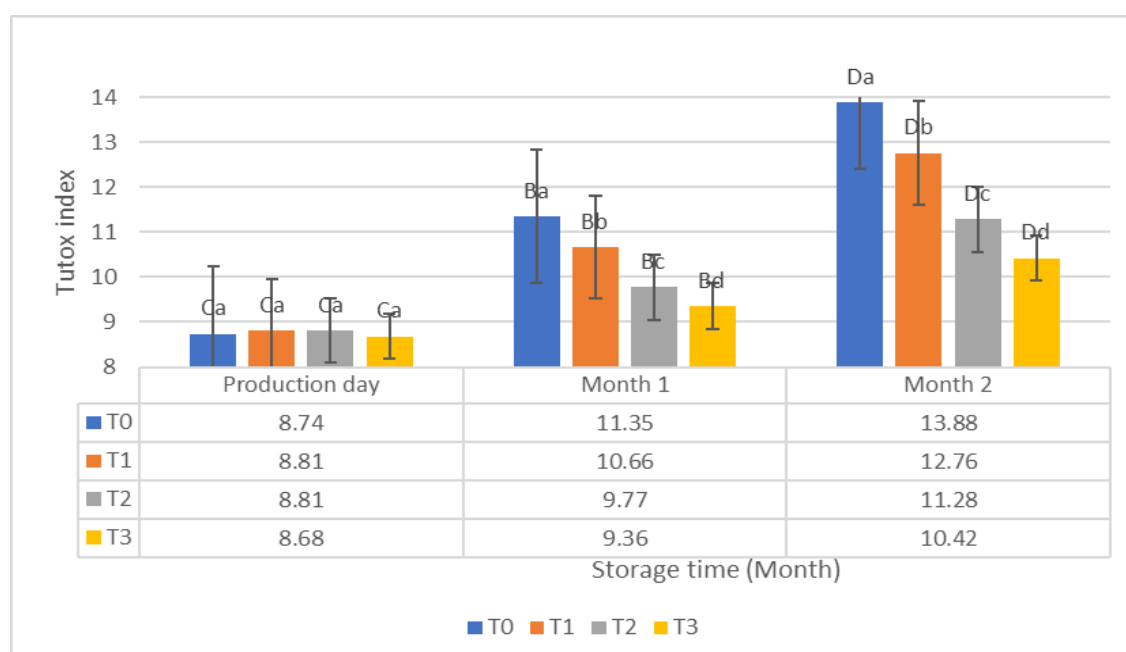
**Fig. 2.** Comparison of the average Anisidine index during two months of storage at a significance level of 0.05.

Figure 3 illustrates that throughout the storage period, a noteworthy increase in the Totox index was observed across all treatments. Furthermore, the application of lemongrass essential oil exhibited a substantial inhibitory effect on the increase of the Totox number. Notably, the formulation containing 1.5% lemongrass essential oil displayed the lowest increment in the Totox index by the conclusion of the second month of storage. The findings suggest that a concentration of 0.5% lemongrass essential oil does not exhibit efficacy in inhibiting the oxidation process of mayonnaise. The elevated Totox index observed in the control sample can be attributed to the increased levels of peroxide and Totox indices relative to those recorded in the other treatments (Abarchai *et al.*, 2022). The research findings indicate that the incorporation of sumac extract and phosphatidylamine has a direct and significant correlation with the Totox index, as evidenced by its relationship with both the anisidine index and the peroxide index. These findings are consistent with the results obtained in the present study (Abarchai *et al.*, 2022).

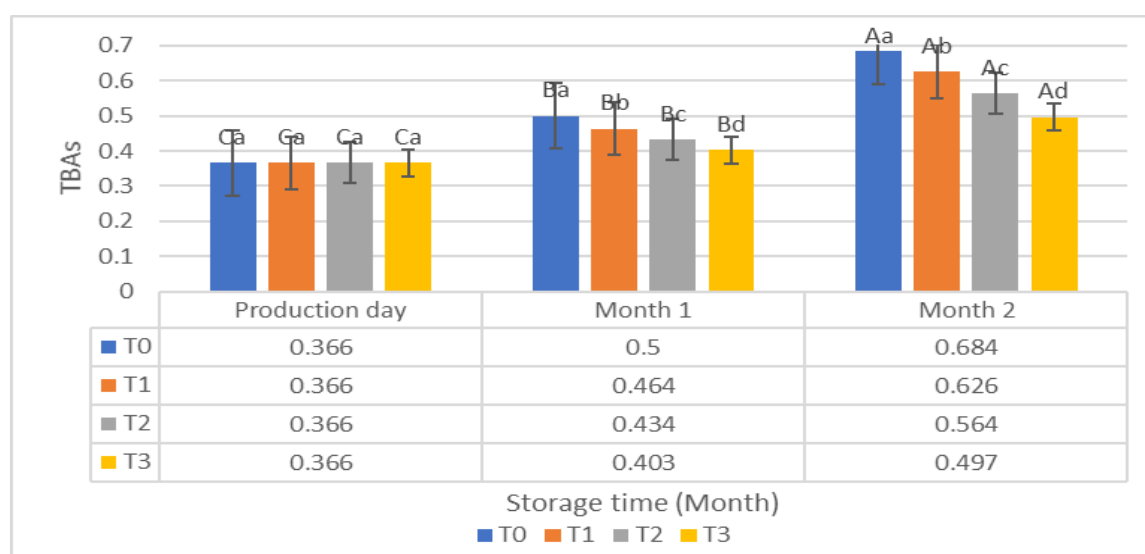
Figure 4 illustrates the oxidation outcomes of mayonnaise treatments, as assessed by the absorption of thiobarbituric acid, which serves as an indicator of secondary oxidation compounds. The most significant inhibitory activity of thiobarbituric acid is associated with its minimal absorption. On the initial day of storage, the absorption of thiobarbituric acid across all treatments containing the sauce was observed to be comparable to that of the control treatment. After a storage period of two months, the treatment infused with 1.5% lemongrass essential oil exhibited the lowest rate of absorption. This phenomenon can be attributed to the observed increase in the concentration of lemongrass essential oil, which correlates with a rise in phenolic compound levels. This elevation appears to inhibit oxidative reactions, thereby hindering the formation of secondary compounds and malonaldehydes (Gorjian *et al.*, 2021).

In a recent study, researchers found that chicken burgers formulated with 1.5% lemongrass extract exhibited a significant decrease in thiobarbituric acid levels following a 90-day period of frozen storage (Zaki, 2018).





**Fig. 3.** Comparison of the average Totox index during two months of storage at a significance level of 0.05.



**Fig. 4.** Comparison of the average TBA index during two months of storage at a significance level of 0.05.

Figure 5 illustrates a notable reduction in pH levels observed during the storage period across all treatment conditions. Furthermore, the application of lemongrass essential oil demonstrated a significant inhibitory effect on the reduction of pH levels. Notably, a concentration of 1.5% lemongrass essential oil resulted in the most minimal decrease in pH after two months of

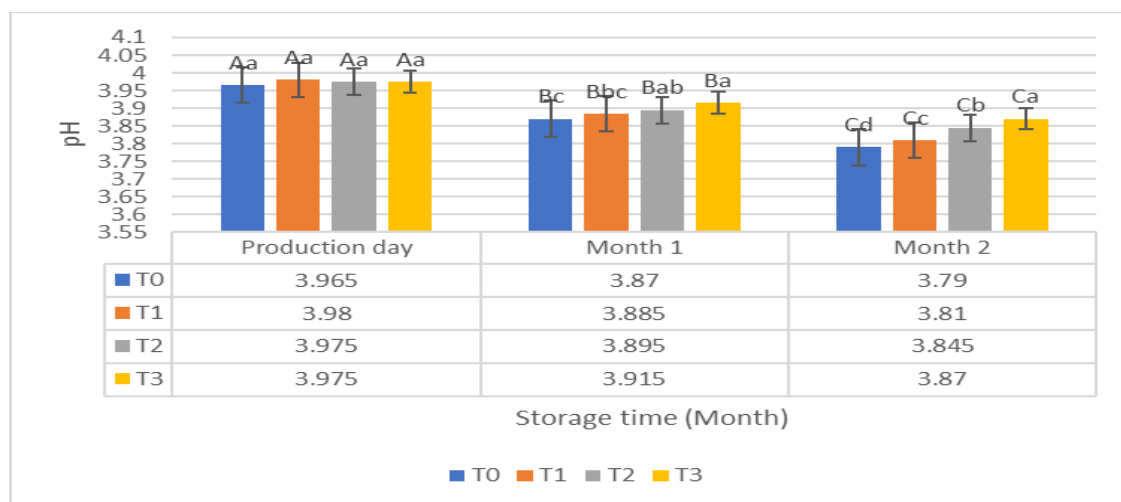
storage. The observed reduction in pH is attributed to an increase in acidity within the mayonnaise, which can be linked to fat oxidation and sugar fermentation resulting from the proliferation of microbial flora. As the concentration of lemongrass essential oil in the experimental treatments was elevated, a corresponding reduction in the extent of pH decrease observed during storage became less pronounced. This

phenomenon can be attributed to the inhibitory effects of lemongrass essential oil on lipid oxidation, as well as its inherent antimicrobial properties.

Figure 6 shows that there was a significant increase in acidity during storage in all treatments. If the acidity of the sauce is more than 1.5%, the mayonnaise will have an unpleasant taste, and if the acidity is less than 0.6%, the conditions for the growth of microorganisms and spoilage will be suitable. The standard acidity range is 0.7% to 1.2% (Rahbari *et al.*, 2013). On the initial day of observation, all treatment conditions exhibited an equivalent level of acidity, measured at 0.6, which is below the established standard range. The application of lemongrass essential oil notably suppressed the rise in acidity levels during storage. Specifically, the treatment involving 1.5% lemongrass essential oil (T3) demonstrated the lowest acidity measurement at the conclusion of the second month of storage, yielding a value of 0.7, which falls within the established standard range. Upon the conclusion of the two-month storage period, all treatments exhibited acidity levels that remained within the established

standard range. In addition to utilizing lemongrass essential oil as an inhibitory agent, the constituents of the mayonnaise emulsion also exert influence on the pH levels and acidity (Taslikh *et al.*, 2022).

In a study conducted by researchers, the impact of savory and tarragon extracts on the physicochemical properties, shelf life, and sensory characteristics of mayonnaise was examined. The findings of this investigation were found to be consistent with those reported in the present study. The investigation revealed that, following a storage period of three months, both the pH and acidity levels in all samples remained within the established parameters set by the Iranian national standard for mayonnaise (Amiri Aghdaei *et al.*, 2023). In a separate study, researchers observed analogous findings when tarragon essential oil was incorporated into mayonnaise. The findings of this study indicate that increased storage duration correlates with elevated acidity levels across all samples examined. Furthermore, the rate of this increase was found to be more pronounced in the control sample (Maldonado-Alvarado *et al.*, 2023).



**Fig. 5.** Comparison of the average pH over two months of storage at a significance level of 0.05.

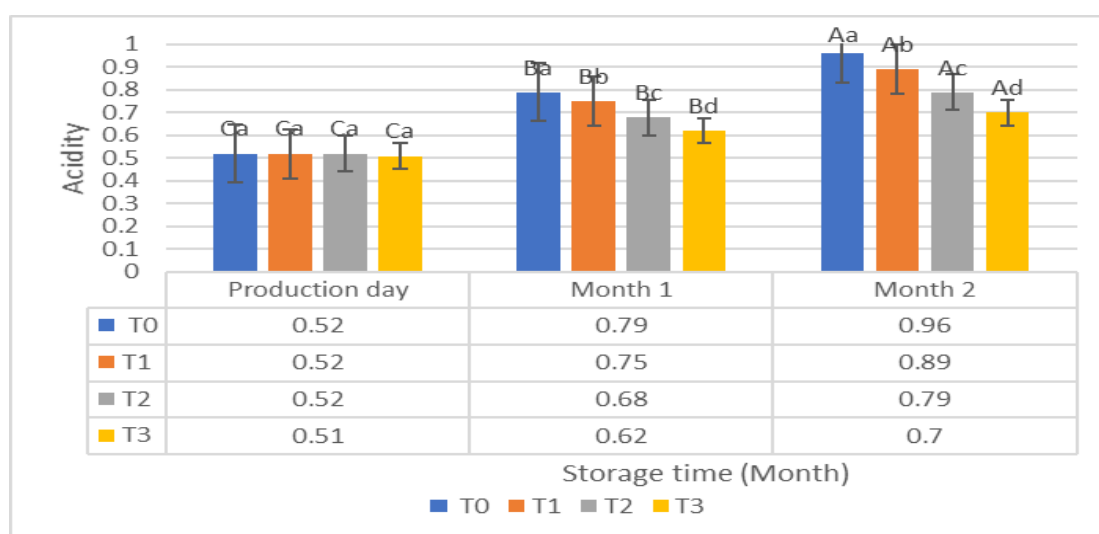


Fig. 6. Comparison of the average acidity index during two months of storage at a significance level of 0.05.

Figure 7 illustrates a notable reduction in viscosity observed during the storage period across all treatment conditions. Furthermore, the application of lemongrass essential oil demonstrated a significant effect in mitigating the reduction of viscosity. Notably, a concentration of 1.5% lemongrass essential oil resulted in the minimal decline in viscosity observed at the conclusion of the second month of storage. The observed decline in viscosity among various mayonnaise samples during storage may be closely correlated with a decrease in pH. This reduction in pH is associated with a diminished stability of the emulsion, which subsequently contributes to the reduction in viscosity.

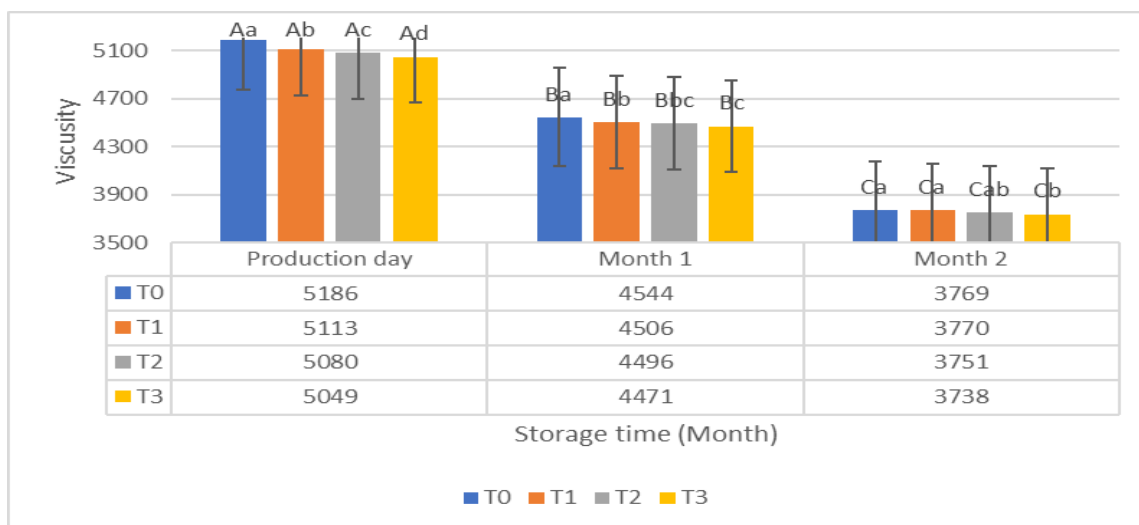
The observed reduction in viscosity across various mayonnaise samples during storage is likely correlated with a decline in pH levels. A lower pH is associated with diminished emulsion stability, which subsequently leads to a decrease in viscosity (Balali & Rahman, 2023). In a separate investigation, it was observed that the viscosity of the solution diminished with the incorporation of clove essential oil. This observation can be attributed to the presence of phenolic compounds in

clove essential oil, which aligns with the findings of the current study (Qasemzadeh *et al.*, 2023).

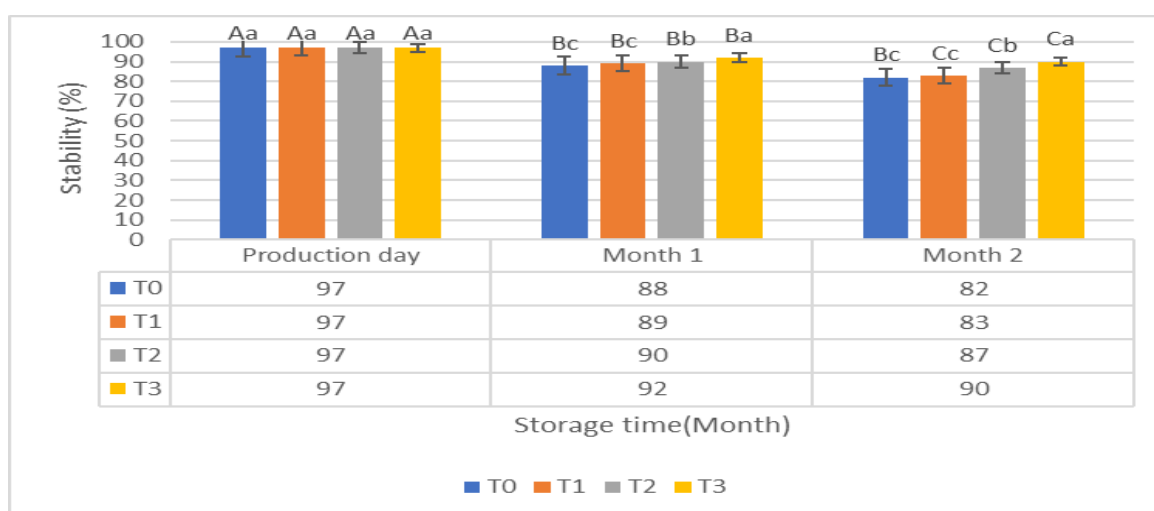
Figure 8 illustrates a marked reduction in stability across all treatments during the storage period. Furthermore, the application of lemongrass essential oil markedly attenuated the decline in stability, with a concentration of 1.5% lemongrass essential oil exhibiting the minimal reduction in stability at the conclusion of the second month of storage. The stability of mayonnaise incorporated with 0.5% lemongrass essential oil, following a storage duration of two months, demonstrated no statistically significant differences when compared to the control treatment. As the concentration of lemongrass essential oil in the treatments increased, the stability of the mayonnaise exhibited a marked improvement in comparison to the control treatment following a storage period of two months. Researchers have documented that the incorporation of phenolic compounds influences the stability and viscosity of emulsions, including mayonnaise. Their findings indicate that the addition of phenolic extracts derived from olive oil results in a

reduction of both viscosity and stability of

these emulsions (Kasapis *et al.*, 1999).



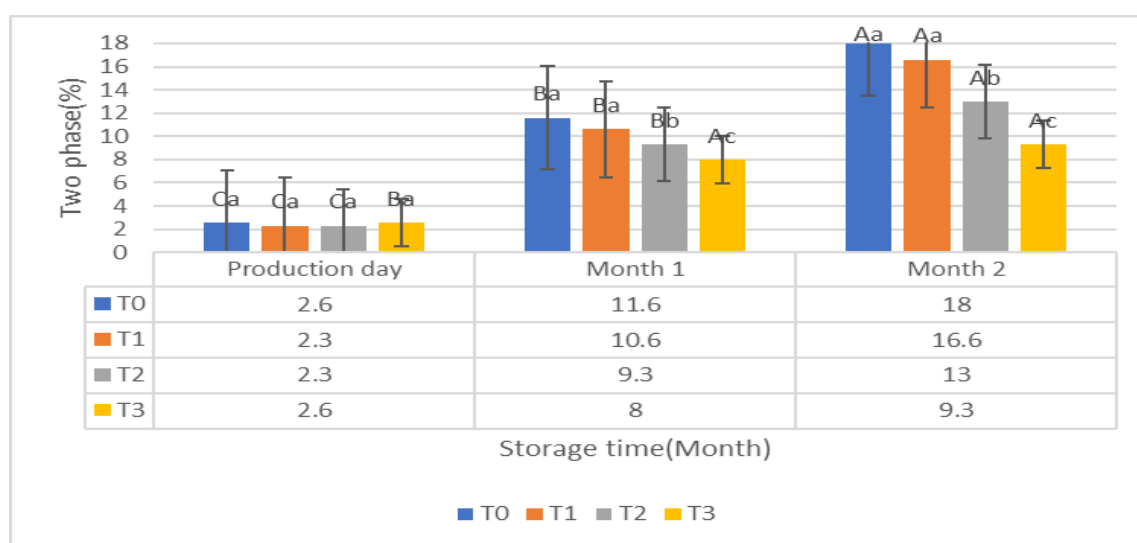
**Fig. 7.** presents a comparative analysis of the average viscosity index over a two-month storage period, evaluated at a significance level of 0. 05



**Fig. 8.** Comparison of average stability during two months of storage at a significance level of 0.05.

As illustrated in Figure 9, all mayonnaise treatments demonstrated a notable upward trajectory in the biphasic index throughout the storage period. On the initial day of the study, all treatments were standardized to a uniform level. However, following a two-month storage period, an observed increase in the concentration of lemongrass essential oil within the mayonnaise treatments corresponded with a suppression of the upward trend in the biphasic index. The

application of mayonnaise infused with 1.5% lemongrass essential oil exhibited the lowest biphasic index following the storage period. This finding demonstrated a statistically significant difference when compared to both the control treatment and the other experimental treatments. Lemongrass essential oil exerts a prophylactic effect on the biphasic index by inhibiting the decline in pH levels (Giacintucci *et al.*, 2016).



**Fig. 9.** Comparison of the average two-phase values during two months of storage at a significance level of 0.05

As illustrated in Figure 10, there was a significant increase in the total bacterial count across all treatments throughout the storage period. Notably, the control treatment exhibited the most pronounced increase, with the highest total bacterial count recorded in the control sample following two months of storage. The findings indicated that the application of lemongrass essential oil resulted in a significant reduction in the total bacterial count. Notably, the highest concentration of lemongrass essential oil demonstrated the most pronounced inhibitory effect. One contributing factor to these alterations can be ascribed to the antimicrobial compounds found in lemongrass essential oil, notably eugenol. The increased concentration of lemongrass essential oil in the treatments subsequently led to a higher quantity of eugenol. The observed inhibitory effect can be attributed to the presence of antimicrobial compounds, particularly eugenol, which are found within the structural composition of lemongrass. Ghasemzadeh *et al.* investigated the impact of clove essential oil on the microbial dynamics within mayonnaise, revealing that the application of clove essential oil substantially

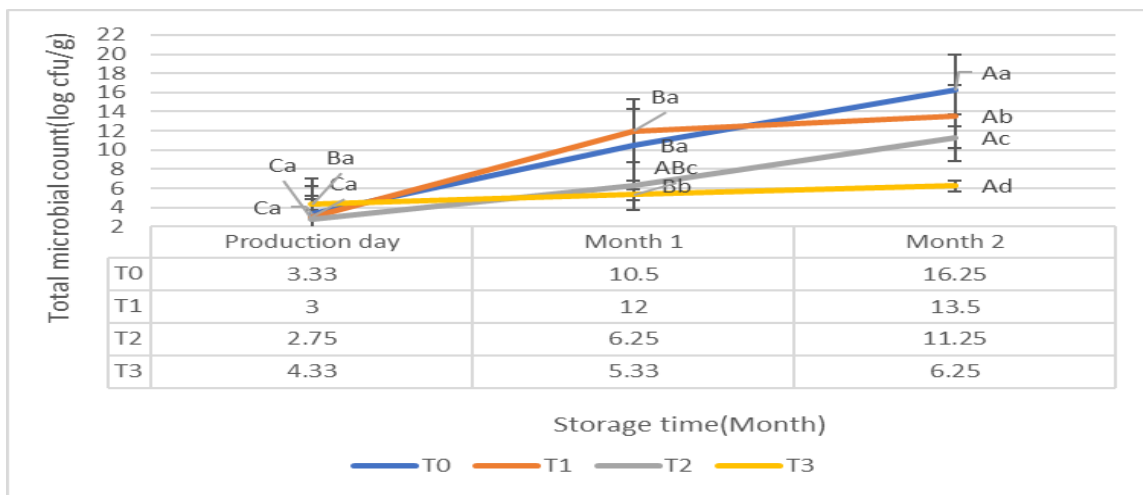
suppressed the populations of *Lactobacillus*, *Salmonella*, molds, and yeasts. Their findings suggest that an increased concentration of essential oil correlates with enhanced antimicrobial effects. One contributing factor to the observed changes can be attributed to the presence of antimicrobial compounds in clove essential oil, particularly eugenol (Qasemzadeh *et al.*, 2023).

Researchers conducted an investigation into the antimicrobial properties of oregano essential oil against *Salmonella* Enteritidis in mayonnaise. The findings indicated that a higher concentration of oregano essential oil significantly diminished the population of *Salmonella* Enteritidis in the analyzed sauce samples (Ghorbani *et al.*, 2015). In a separate investigation, researchers noted that with an increase in the concentration of yellow mustard, there was a corresponding decline in microbial populations (Adeli milani *et al.*, 2010). Another investigation demonstrated that elevating the concentration of marjoram essential oil to 250 parts per million (ppm) significantly contributed to the inhibition of mold and yeast proliferation, thereby effectively preventing microbial spoilage to a degree

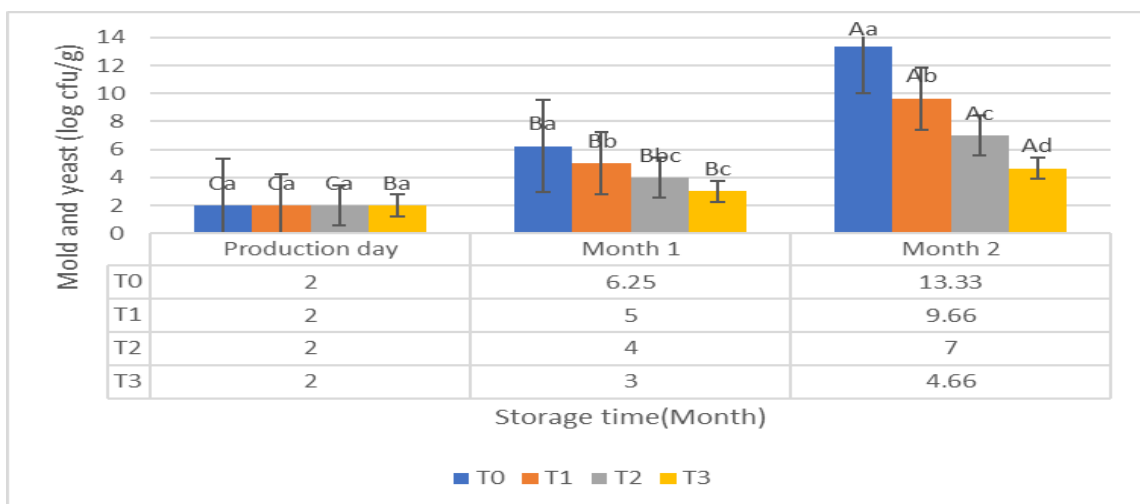
consistent with desired outcomes. The observed effects can be ascribed to the presence of compounds such as thymol and carvacrol within the structural composition of the essential oil (Ghorbani *et al.*, 2015).

The results pertaining to the total counts of mold and yeast in various samples of mayonnaise incorporating lemongrass essential oil are presented in Figure 11. Throughout the storage period, a significant increase in total mold and yeast counts was observed across all treatments, with particularly pronounced

levels in the control treatment. Notably, the highest total mold and yeast counts were recorded in the control sample at the two-month mark of storage. The findings of this study demonstrated that the application of lemongrass essential oil significantly suppressed the proliferation of total mold and yeast populations. The observed inhibitory effect can be ascribed to the presence of antimicrobial compounds, specifically eugenol, within the composition of lemongrass (Qasemzadeh *et al.*, 2023).



**Fig. 10.** Comparison of the average total microbial count during two months of storage at a significance level of 0.05.

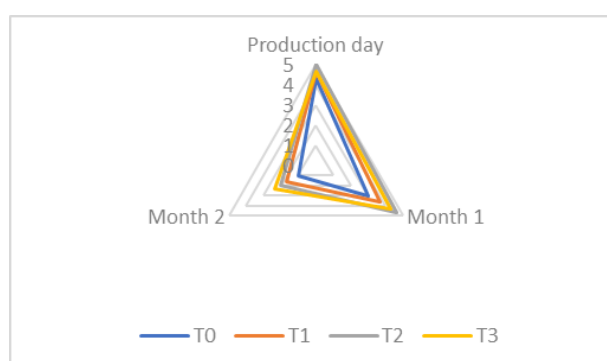


**Fig. 11.** Comparison of the average mold and yeast counts during two months of storage at a significance level of 0.05.

The findings from the sensory evaluation pertaining to aroma, taste, color, texture, and overall acceptance are presented in Figures 12 through 16, respectively. The findings indicate that, prior to the commencement of the storage period, all parameters under investigation exhibited optimal values. Over time, a reduction in sensory scores was noted across all parameters examined in this study. This phenomenon can be ascribed to the oxidation of lipids during storage, which negatively influences the sensory attributes of mayonnaise. The application of lemongrass essential oil demonstrated a notable capacity to decelerate the degradation process, with the observed effects becoming increasingly pronounced as the concentration of the essential oil was elevated. The observed elevation in sensory scores can be ascribed to the antioxidant properties and the citrus flavor profile characteristic of lemongrass essential oil. Research indicates that bulk oregano essential oil, particularly when utilized in nanoemulsified form, has significant potential as a natural preservative for extending the shelf life of mayonnaise (Rashidi & Amin Shamekhi, 2023). Another study indicated that the incorporation of ginger into mayonnaise substantially enhanced its sensory attributes, with the degree of enhancement being contingent upon the concentration of ginger employed (Kishk & Elsheshetawy,

2013). The outcomes of the sensory evaluations indicated that an increase in the concentration of lemongrass essential oil was positively correlated with the aroma, flavor, texture, color, and overall acceptance of the mayonnaise among the participants.

The findings from the sensory evaluation of taste across various mayonnaise samples are presented in Figure 12. The findings indicate that throughout the storage period, lemongrass essential oil exerted a noteworthy influence on the sensory profile, specifically the flavor, of the various mayonnaise samples. On the initial day of the study, no statistically significant differences were detected in the sensory evaluation of taste across the various treatments. However, beginning on day 30, a statistically significant difference was identified. A notable downward trend in the sensory evaluation of taste was observed across all studied samples. The incorporation of increased quantities of lemongrass essential oil into various mayonnaise formulations substantially enhanced the sensory acceptability concerning taste, as evidenced by the evaluation results. This level of acceptability may be attributed to the presence of lemon flavor compounds in lemongrass essential oil (Kishk & Elsheshetawy, 2013).



**Fig. 12.** Comparison of the average taste scores over two months of storage at a significance level of 0.05.

The findings from the sensory evaluation of aroma and olfactory characteristics across various mayonnaise samples are presented in Figure 13. The findings indicate that, throughout the storage period, the incorporation of lemongrass essential oil significantly influenced the aroma and scent profiles of the various mayonnaise samples. On the initial day of the study, no statistically significant differences were observed in the sensory evaluation of aroma and odor across the various treatment groups. Nonetheless, commencing from day 30, a statistically significant difference was evident. A noteworthy decline in the sensory evaluation of aroma and olfactory characteristics was observed across all samples examined. The increment of lemongrass essential oil concentration in the various mayonnaise samples demonstrated a significant enhancement in the sensory acceptability concerning aroma and olfactory characteristics. The acceptability of this phenomenon can be attributed to the presence of the lemon aroma and the olfactory characteristics associated with lemongrass essential oil. By the conclusion of the second month of storage, treatments T3 and T2 were assessed by the evaluators to be at an equivalent level of quality. (Rashidi & Amin Shamekhi, 2023)

The findings from the sensory evaluation of color across various mayonnaise samples are presented in Figure 14. The findings indicate that throughout the storage period, the application of lemongrass essential oil exerted a significant influence on the color characteristics of the various mayonnaise samples analyzed. On the initial day of the study, the sensory evaluation of color did not reveal any statistically significant differences among the various treatments. Starting from day 30, a statistically

significant difference was observed. A notable downward trend in the sensory evaluation of color was identified across all samples analyzed. The incremental incorporation of lemongrass essential oil into various mayonnaise samples led to a noteworthy enhancement in the sensory acceptability of the color attribute. The findings suggest that lemongrass essential oil effectively mitigates perceptible color alterations by suppressing the oxidation of lipids. (Farzad Kazemipour, 2024)

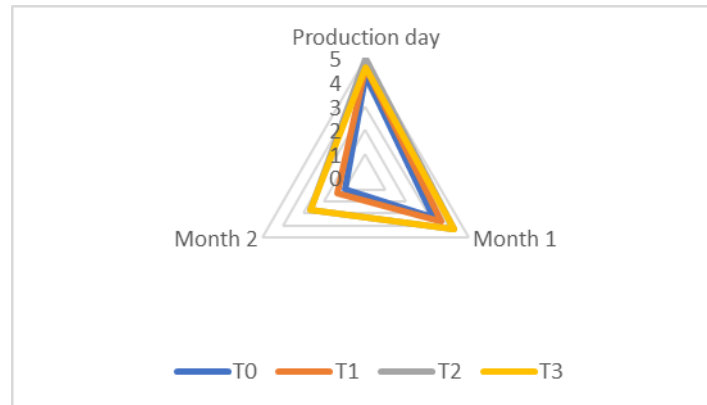
The findings from the sensory evaluation of texture across various mayonnaise samples are presented in Figure 15. The findings indicate that during the storage period, lemongrass essential oil exerted a considerable influence on the texture of the various mayonnaise samples. On the inaugural day of the study, the analysis of sensory evaluations pertaining to texture revealed no statistically significant differences among the various treatments. Starting from day 30, a statistically significant difference was observed. A notable decreasing trend in the sensory evaluation of texture was identified across all samples analyzed in the study. The incorporation of higher concentrations of lemongrass essential oil in various mayonnaise samples significantly enhanced the acceptability of the sensory evaluation concerning texture.

The findings of the sensory evaluation concerning overall acceptance of various mayonnaise samples are depicted in Figure 16. The findings indicate that the inclusion of lemongrass essential oil during the storage period significantly influenced the overall acceptance of the various mayonnaise samples. On the initial day of the study, there were no statistically significant differences detected in the overall acceptance sensory evaluation across the various treatments.

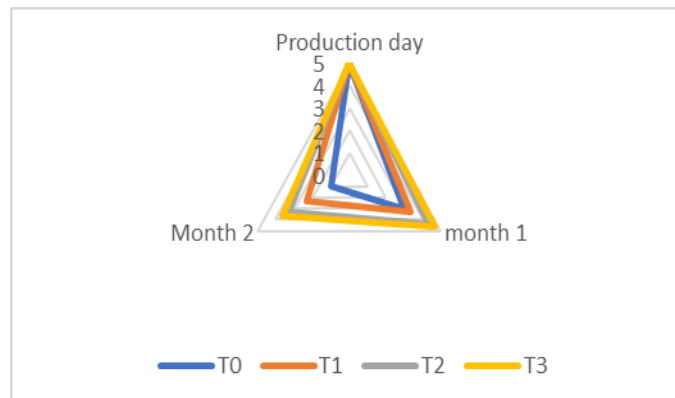


Nevertheless, beginning on day 30, a statistically significant difference was observed. A notable decline in the overall acceptance ratings during sensory evaluation was observed across all samples examined in this study in Figure 17. On Day 30, Treatment 2 (T2) demonstrated a higher acceptance score

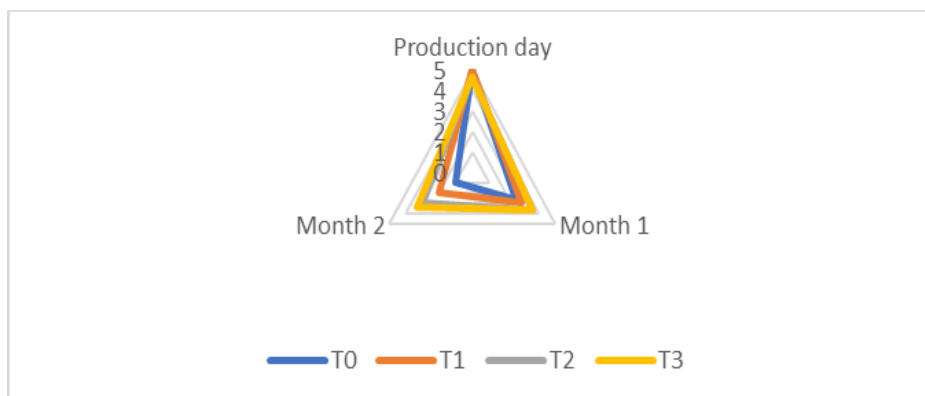
compared to the treatment containing a greater concentration of essential oil (T3) as assessed by the evaluators. An increase in the concentration of lemongrass essential oil across various mayonnaise samples markedly preserved the overall acceptability of the texture evaluation. (Farzad Kazemipour, 2024)



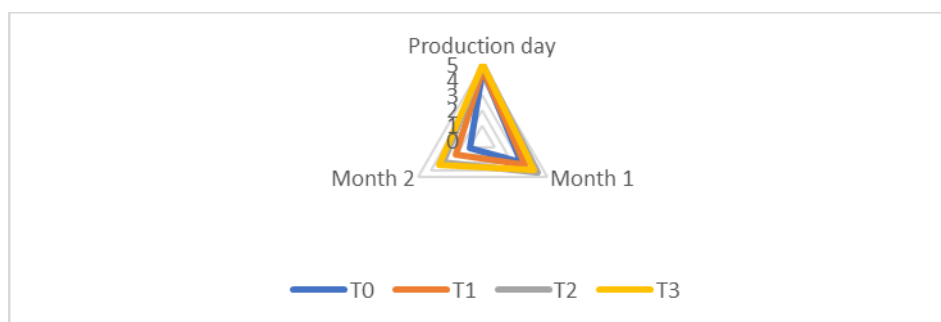
**Fig. 13.** Comparison of the average perfume scores over two months of storage at a significance level of 0.05.



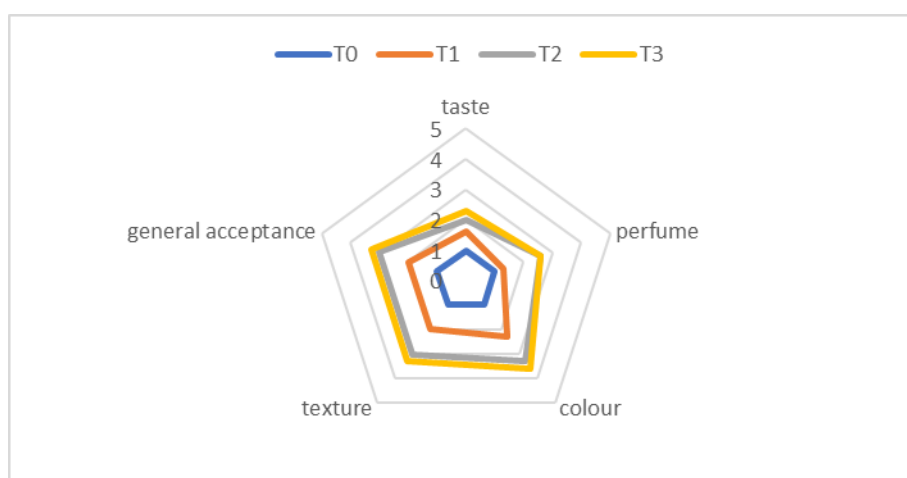
**Fig. 14.** Comparison of the average color scores during two months of storage at a significance level of 0.05.



**Fig. 15.** Comparison of the average texture scores during two months of storage at a significance level of 0.05.



**Fig. 16.** Comparison of the mean general acceptance scores over two months of storage at a significance level of 0.05.



**Fig. 17.** Results of the assessment of sensory characteristics after two months of storage at a significance level of 0.05.

## Conclusion

Given the potential risks associated with chemical preservatives for consumers, it is imperative to substitute these compounds with natural preservatives in the formulation of mayonnaise. Lemongrass essential oil represents one of the natural compounds utilized in the present study. The findings of this study indicate that lemongrass essential oil exhibits commendable antioxidant and antimicrobial properties, attributable to its phenolic complex. The findings from the antioxidant assays conducted on samples infused with lemongrass essential oil (designated as T1, T2, and T3) demonstrated a positive efficacy in inhibiting lipid oxidation within mayonnaise formulations. The T3 treatment significantly attenuated the

upward trend of peroxide values, anisidine values, thiobarbituric acid levels, and the TOTOX index throughout the storage period. The pH levels and acidity measurements of samples containing mayonnaise (T1, T2, T3) remained within the standard range following the two-month storage period. Lemongrass essential oil demonstrated the capability to preserve the rheological properties at an optimal level. Consequently, the T3 treatment effectively mitigated the decline in stability and viscosity, while also inhibiting the tendency for two-phase formation during the storage period. Furthermore, it successfully hindered microbial growth following the storage duration. The findings from the sensory evaluations indicated a positive correlation between the concentration of lemongrass essential

oil and the acceptance of its sensory attributes by the evaluators. Specifically, the T3 treatment achieved the highest scores in the assessment of taste, aroma, color, texture, and overall acceptance, reflecting a favorable reception of these sensory properties at elevated oil concentrations. It is generally advised to incorporate lemongrass essential oil into food products that are prone to oxidation. Considering the comprehensive analysis of the results and the sensory evaluation findings, the T3 treatment has been identified as the superior treatment within the context of this study.

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