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Research Article

### Ai aştıi illa iviakaicsi

# **Quality Changes in Sea Bass Patties Containing Lemon Essential Oil Loaded Oleogels**

Limon Esansiyel Yağı Yüklenmiş Oleojeller İçeren Levrek Köftelerinde Kalite Değişimleri

Emine Bakır<sup>1</sup>, Turgay Çetinkaya<sup>2,\*</sup>, Serhan Mantoğlu<sup>1</sup>

<sup>1</sup>Yalova University, Armutlu Vocational School, Food Processing Department, Yalova-TÜRKİYE <sup>2</sup>İstanbul University, Faculty of Aquatic Sciences, Department of Aquatic Biotechnology and Genomics, Department of Aquatic Biotechnology, İstanbul-TÜRKİYE

\*Corresponding author: turgaycetinkaya@istanbul.edu.tr

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Abstract: We aimed to deliver lemon essential oil utilizing oleogels and determined the effect on quality parameters of sea bass patties. Control patties were formulated using sunflower oil in minced sea bass. For the second and third groups, oleogels prepared with a candelilla wax-sunflower oil combination were added to minced meat at a ratio of 7.87% (w/w). For the third group, 0.0078% (w/w) lemon essential oil containing oleogel was added to the patty's formulation. Physicochemical, textural, and microbiological analyses were applied, and three groups were compared. Hardness increased in uncooked samples throughout the shelf life, with the control group being hardest, followed by oleogel and then lemon+oleogel groups. Higher adhesiveness was observed in oleogel groups. However, L\*, a\*, and b\* differences were more stable for oleogel groups through storage. Although the threshold limit was not exceeded, on 5<sup>th</sup> day, the lemon added oleogel group had lower total volatile basic nitrogen (TVBN) than other groups. pH values decreased for all groups throughout storage. While peroxide value (PV) and free fatty acid (FFA) values increased, no significant differences were observed between groups. Higher FFA levels in oleogel-containing patties suggest the interactions between various lipid degradation products. Oleogel addition did not show a significant effect on total mesophilic aerobic bacteria (TMAB) and total psychrophilic bacteria (TPB) results. Nevertheless, the results shed light on the necessity of novel oleogel substitute formulations to improve the quality of processed seafood products.

Özet: Çalışmamızda limon esansiyel yağı eklenen oleojellerin levrek köftelerinin pişirme kaybı, tekstür, renk ve fizikokimyasal parametreleri üzerindeki etkisi belirlenmiştir. Kontrol levrek köfteleri ayçiçek yağı kullanılarak formüle edilmiştir. İkinci grup için, kandelilla vaksı-ayçiçek yağı kombinasyonu ile hazırlanan oleojeller kıymaya %7,87 (a/a) oranında eklenmiştir. Üçüncü grupta ise oleojellere ek olarak formülasyona %0,0078 (a/a) oranında limon esansiyel yağı eklenmiştir. Sertlik, örneklerde pişmemiş raf ömrü boyunca artmıştır, sertlik kontrol>oleojel>limon+oleojel grupları olarak sıralanmıştır. Oleojel gruplarında daha yüksek yapışkanlık gözlenmiştir. Ancak, L\*, a\*, ve b\* farkları, depolama boyunca oleojel grupları için daha stabil olmuştur. Eşik değeri aşılmamasına rağmen, 5. günde limon eklenen oleojel grubu diğer gruplardan daha düşük toplam uçucu bazik azot (TVBN) değerine sahip olduğu belirlenmiştir. pH değerleri depolama boyunca tüm gruplarda azalmıştır. Peroksit ve serbest yağ asidi (FFA) değerleri artarken, gruplar arasında önemli bir fark gözlenmedi. Oleojel içeren köftelerdeki daha yüksek FFA seviyeleri, çeşitli lipit bozunma ürünleri arasındaki etkileşimleri düşündürmektedir. Oleojel ilavesi, toplam mezofilik aerobik bakteri (TMAB) ve toplam psikrofilik bakteri (TPB) sonuçları üzerinde önemli bir etki göstermemiştir. Bununla birlikte, sonuçlar işlenmiş deniz ürünlerinin kalitesini iyileştirmek için yeni oleojel ikame formülasyonlarının gerekliliğine ışık tutmaktadır.

### Keywords

- Minced meat
- Shelf life
- Oleogel
- Essential oil
- Sea bass patties

# Anahtar kelimeler

- Kıyma
- Raf ömrü
- Oleojel
- Esansiyel Yağ
- Levrek Köftesi

### 1. INTRODUCTION

One of the fundamental rules of maintaining health is balanced nutrition. Essential amino acids, fatty acids, vitamins, minerals, and water are indispensable components of a healthy diet. Aquatic foods provide these nutrients sufficiently, so including them in the diet is important. Fish patties have been consumed worldwide for many years and are prepared with different recipes. Rich in protein, they also contain omega-3 fatty acids and essential fatty acids.

Securing that the reformulated product sustains quality and sensory features over time is critical (Ferdaus et al., 2024). The limited shelf life of fish patties poses a significant challenge. Finding effective and non-toxic techniques to slow down spoilage and extend shelf life is challenging (Balıkçı et al., 2022). In oily fish products such as salmon and sea bass, oil release and oxidation may occur along with spoilage because of the high amount of omega-3 fatty acids presence, which are sensitive to oxidation. Oxidation can negatively affect the taste and nutritive value (Magsood & Benjakul, 2011; Parmar et al., 2022). Therefore, the use of plantbased extracts or essential oils as natural preservatives may be suitable to inhibit microbial growth or prevent oxidation in seafood (Balıkçı et al., 2022). Recently, there has been increased interest in antimicrobial agents derived from natural products. Aromatic plants stand out in this regard due to their high antimicrobial properties. The antimicrobial components of these plants are typically concentrated in volatile which are extracted through distillation and consist of fragrant, oil-soluble In addition to their antimicrobial properties, essential oils also possess therapeutic, antiseptic, and antibiotic characteristics (Keser & İzci, 2020). Wang, Mei, & Xie (2022) conducted a study in which they treated sea bass with different concentrations of lemon essential oil, demonstrating that this oil reduced oxidative stress levels and increased total antioxidant capacity. This suggests that lemon essential oil could be used in the fish balls.

Solid fats are widely used in the food industry to enhance the functional properties of a diet. However, a study involving 29 different brands of products made with these solid fats found that, despite the presence of labels claiming "trans fatfree," trans fats were detected. Moreover, one product exceeded the regulatory limits for trans

fat content. Foods containing trans fats can contribute to various health problems, such as cardiovascular diseases and several types of cancer (Demir & Taşan, 2019). In this context, the oleogelation technique emerges as a new and popular method that allows for the use of fatty acids without causing any chemical changes in their structure. The immobilization of the liquid phase within the gel network occurs through the formation of cross-links or complex bonds by polymeric gelators due to chemical and physical interactions. Additionally, low molecular weight organic gelators produce large aggregates through physical interactions. Subsequently, the structure is strengthened by intermolecular interactions, including hydrogen bonds, van der Waals forces, and  $\pi$ - $\pi$ interactions, resulting in the formation of the final oleogel (Yenioğlu Demiralp et al., 2017). Oleogels have recently become common bioproducts due to their ability to replace other oils in food products. Studies on the use of oleogels in various meat and deli products have primarily focused on items made from red meat, such as sausages and salami (Çakır, 2021; Çalişkan, 2024; Toksöz, 2020; Tüysüz, 2024). To date, there has been no study on using oleogels as a sunflower oil replacer in fish patties for delivering lemon essential oil, making this research a first in this field.

Waxes have several advantages, such as being food-grade, low-cost, having good accessibility, and outstanding gelling ability. Generally, waxes consist of long-chained saturated fatty acids, C-20 to C-26, and fatty alcohols with long chain lengths (Hamidioglu et al., 2022). Oleogels are anhydrous colloidal systems formed by trapping a liquid oil using gelling agents such as gelators (Karimidastjerd et al., 2024). Oleogels produced with rice bran and candelilla waxes may maintain the stability of structure and nutritional value despite their low solid fat content. 1–3% w/w of candelilla wax is required to form an oleogel (Hamidioglu et al., 2022).

There are various studies on different versions of fish patties (Balıkçı et al., 2022; Bilgin & Metin, 2022; Can, 2012; Çapkın et al., 2020; Duman & Özpolat, 2011; Gökoğlu, 1994; Keser & İzci, 2020). For instance, fish patties from tench (*Tinca tinca*) with thyme (1%) and ginger (1%) were prepared, and it was found that the limit value (10<sup>7</sup> CFU/g) was exceeded on the 10<sup>th</sup> day of storage at 4°C. However, no literature has been found on the use of oleogel substitutes to

determine quality in the fish patties. For these reasons, the purpose of this research is to determine the effect of using an oleogel substitute formulation without and with lemon essential oil on sea bass fish patties' physical, microbiological, and chemical properties during 5 days of storage at 8°C.

## 2. MATERIAL and METHODS

### 2.1. Materials

Fresh filleted sea bass (Dicentrarchus labrax) were supplied from the fish market in Armutlu, Yalova region, and brought to the food processing laboratory on the same day. To accommodate also sensory panel requirements, the total sample consisted of fillets from nine fish, which were pooled to form a composite batch for patty preparation. Sunflower oil (Yudum), candelilla wax (Euphorbia cerifera) (Strahl & Pitsch wax), and lemon essential oil (Talya Herbal Products Trade and Industry Inc.) are used for oleogel production. Breadcrumbs and spices were used for the patties mixture (Baghdad Spice Food Industry and Industry Co. Ltd.), and garlic was purchased from local markets.

In the analyses conducted within the scope of this study; Ethanol, acetic acid, diethyl ether, chloroform, potassium iodide, starch, and sodium thiosulfate were provided from Merck. Sodium hydroxide, phenolphthalein, and other chemicals were purchased from Sigma Aldrich.

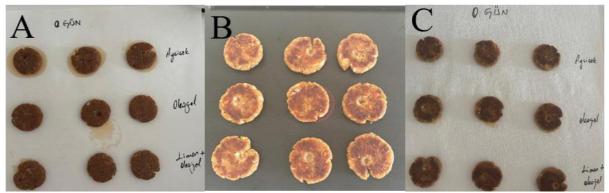
### 2.2. Preparation of Oleogels and Fish Patties

The production of oleogel was carried out by adopting the method of Öğütcü & Yilmaz (2014). To form the oleogel, candelilla wax (ratio of 2.5% w/w) and sunflower oil (ratio of 97.5% w/w) were heated separately in a water bath at 90°C. After the wax and oil were mixed into the oleogels, the essential oils were immediately

added at a rate of  $20~\mu L$  per 100~grams of sunflower oil. The mixture was then stirred for 30~seconds at 500~rpm using a magnetic stirrer to ensure homogeneity. The resulting mixture was transferred to containers and left at room temperature for 24~hours for oleogel formation.

The patty mixture was modified from the formulation presented by Keser & İzci (2020) and was determined through preliminary trials with sensory evaluations by the authors. As a result, three types of sea bass patties were prepared, including one control sample. Minced seabass fillets were added to the mixture in the amounts specified below:

The individual recipe for the control group was 100 grams of fish mince, 10 grams of breadcrumbs, 2 grams of salt, 0.5 grams of cumin, 0.5 grams of sweet paprika powder, 0.5 grams of allspice, 0.5 grams of ground black pepper, 3 grams of garlic, and 10 grams of sunflower oil (Control group). In oleogel group, the same formulation was used, except adding 10 grams of oleogel instead of sunflower oil (Oleogel group). For lemon+oleogel sample, 0.1% (w/w) lemon essential oil was added to 10 grams of oleogel. Therefore, in the end, oleogel ratio was fixed at 7.87% (w/w), and the lemon essential oil ratio was fixed at 0.0078% (w/w) in the lemon+oleogel group patties formulation. From the prepared mixtures, 20 grams of each mix were taken and shaped into patties using molds with a height of 12 mm and a diameter of 45 mm (Figure 1). The samples were covered with baking paper and stored in zip-lock bags at 8±1 °C for 5 days for further analysis. This abuse temperature was chosen to observe spoilage within a shorter period, as it accelerates microbial activity, thereby speeding up the spoilage process.



**Figure 1.** Photos of prepared control, oleogel, and lemon+oleogel groups from up to down. A: Uncooked, B: During grilling, C: After grilling.

# 2.3. Physical Analysis of Uncooked and Grilled Fish Patties

For cooking loss, prepared patties were weighed in three repetitions before grilling. Then, the front and back sides of the patties were cooked for approximately 3 minutes in the Tefal electric grill device in the fish cooking mode. Finally, the grilled patties were weighed, and the weight loss percentage after cooking was calculated according to the method of Samuel Wu et al. (2021). Hardness, adhesiveness, and chewiness values were determined using the CT3 device (Brookfield) and TA8 ball probe in accordance with the operating guidelines provided by Brookfield Engineering (Ametek Brookfield, 2023) and Serdaroğlu, & Karaman, M. (2025) with 3 replications each for uncooked and grilled patties on days 0, 1, 2, 3, 4, and 5. The parameters are as follows: Test speed 3 mm/s; pre-test speed 2 mm/s, Target depth 4 mm. Color parameters from three different parts of patties, L\* lightness (black to white 0-100), a\* (redness + and green-), b\* (yellowness + and blueness -), were obtained using Fru® Precise Color Reader with 8 mm caliber, which adheres to standards of International Commission on Illumination (CIE) for colorimetry commonly used in food quality control.

# **2.4.** Microbiological Analysis of Uncooked Fish Patties

During the storage period, TMAB and TPB counts were performed on prepared uncooked patties. 10 g of each sample was taken and homogenized with 90 mL peptone water (0.1%) in a stomacher. Serial dilutions were prepared for each sample, and 1 ml of the diluted sample was transferred to petri dishes and the pour plate method was applied as indicated in the FDA standard (Maturin & Peeler, 1988). The samples were incubated at 35 °C for 48 hours for TMAB. To determine the TPB change, the inoculated petri dishes were incubated in an incubator set at 7 °C for 10 days.

# 2.5. Chemical Analysis of Uncooked Fish Patties

HANNA HI 2211 pH meter was used for pH analysis following the procedure outlined in ISO 2918:1975 (ISO, 1975). Five grams of each sample were taken, and 50 mL of distilled water was added with three repetitions. The mixture was homogenized using a homogenizer at 75,000 rpm until the meat dissolved.

For PV and FFA determination, the method of Quansar et al. (2025) was modified.10 grams of

the weighed sea bass patties were treated with 50 mL of chloroform, homogenized for 1 minute using an Ultra Turrax, and then filtered. Following this, 10 mL of distilled water was added, and the phases were separated in a separating funnel, allowing the chloroform phase to be collected to obtain lipid extracts from the patties. These lipid extracts were used for peroxide and FFA analyses.

For FFA analysis, 5 mL of the lipid extract was taken and mixed with 10 mL of an ethanol/diethyl ether (1:1) solution, to which the phenolphthalein indicator was added. This mixture was then titrated with 0.01 N NaOH. The FFA content was expressed as grams of oleic acid per 100 g of sea bass patties sample and calculated using the following formula:

FFA%

blank sample

$$= \frac{Titration(a - b) \times n \times 282 \times 100 \times dilution factor}{1000 \times W}$$
a=ml of NaOH solution used in titration

n= normality of NaOH b=ml of NaOH solution used in titrated with

282=oleic acid molecular weight W=weight of sample

For peroxide analysis, 10 mL of the lipid extract was taken, and 15 mL of acetic acid was added. After adding 1 mL of potassium iodide, the mixture was kept in the dark with a closed lid for 5 minutes. At the end of this period, 1 mL of starch was added to the samples, which were then titrated with 0.01 N Na<sub>2</sub>S<sub>2</sub>O<sub>4</sub>. The amount of titrant used was recorded and calculated using the following formula:

PV (meq oxygen/kg of fish patties)

$$= \frac{V \times n \times 1000 \times dilution \ factor}{W}$$
V= ml of Na<sub>2</sub>S<sub>2</sub>O<sub>4</sub> used in titration
n= normality
W=weight of sample

TVBN content determined using the method of Güroy et al. (2019). The homogenized 10g sample was transferred to a Kjeldahl tube, and 1 g MgO and 100 mL distilled water were added with two repetitions. In addition, 100 mL of distilled water, 10 ml 3% boric acid, and 7 drops of Tachiro indicator are added erlenmeyer flask. The prepared samples are used in the Kjeldahl

Distillation device (Behr S3), and the distillation process is continued until distillate is collected in the Erlenmeyer flask. The obtained distillate is titrated with 0.1N HCl acid, and the TVBN ratio is calculated as;

$$TVBN = \frac{Comsumption \times 100 \times 1.4}{Sample\ Weight}$$

### 2.6. Sensory Evaluation

Hedonic test was applied to the 10 panelists according to the prepared forms. Cooked three types of patties were coded and scored between 1-5 by panelists. Sensory analysis was performed only on the initial day.

### 2.7. Statistical Analysis

The data obtained from the analyses were statistically analyzed using the IBM SPSS 29.0.0.0 (241) software with the Tukey test ( $P \le 0.05$ ). In the Tables, mean values in the rows with a different superscript letter are presented as

significantly different between groups ( $p \le 0.05$ ).

### 3. RESULTS AND DISCUSSION

# 3.1. Cooking Loss

Grilled patties lead to both moisture loss and fat loss due to the heat during grilling, causing fat to ooze from the patties. According to Figure 2, results fluctuated, and more cooking loss was obtained in oleogel-added samples on all days except day 4. Similar loss range percentages were found in previous studies (Serdaroğlu & Karaman, 2025). Keenan et al. (2015) stated that increasing fish oil substitutes resulted in increased cooking loss and reduced hardness in beef burgers. In another study, it was reported that using olive oil oleogel instead of animal fat in producing chicken burger patties increased cooking loss (Tüysüz, 2024). Increasing cooking loss after oleogel addition in our study may be attributed to factors such as the type of oleogel, its concentration, and the seafood matrix.

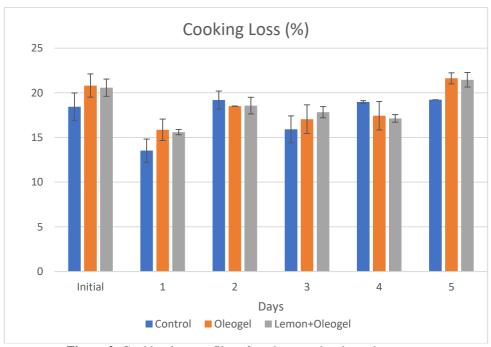


Figure 2. Cooking loss profiles of sea bass patties through storage.

# **3.2.** Texture Parameters of Uncooked and Grilled Patties

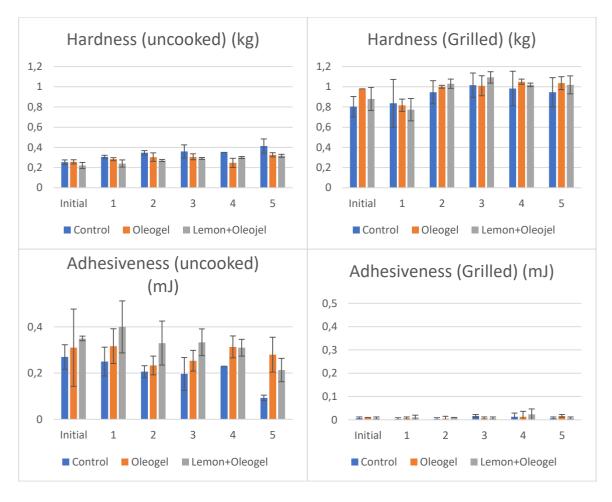
Textural parameters of fish are associated with the structural integrity of muscle tissue, with post-mortem tissue disintegration gradually occurring leading to meat tenderization through autolysis. The effect of added oleogels on the textural properties of sea bass patties including hardness, adhesion, and springiness values during

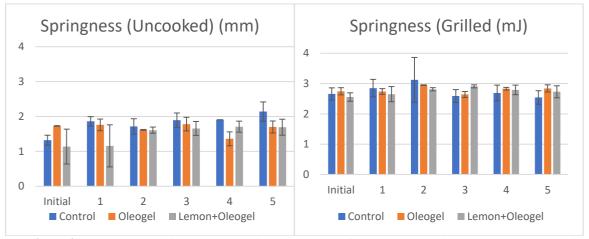
their shelf life was assessed. The hardness values showed an increase in each group of uncooked products throughout their shelf life under the experimental conditions (Figure 3). Oleogel addition decreased the hardness of uncooked sea bass patties. Throughout 1-5 days, the hardest group was the control, followed by the oleogel group and then the lemon-added group. Similar studies are reporting a reduction in the hardness

of uncooked patties with the use of oleogel (Gao et al., 2021; Gómez-Estaca et al., 2020; Martins et al., 2019; Moghtadaei et al., 2018; Oh et al., 2019). In a study evaluating the production of beef burgers with varying levels of oleogel made with beeswax and sesame oil, decreased hardness was attributed to the smaller size of fat globules, which resulted from the replacement of animal fat (Moghtadaei et al., 2018). In another study using oleogel as a replacer for beef fat in beef heart patties, hardness was significantly reduced compared to patties with beef fat (Gao et al., 2021). Similarly, oleogels prepared hydroxypropyl methylcellulose as a fat replacer in meat patties significantly decreased the hardness values compared to beef fat and canola oil. These studies highlight that maintaining and enhancing the softness of meat patties requires retaining moisture and fat within the patties (Oh et al., 2019). Considering that oleogels retain the

oil structure more effectively compared to controls, the softness of patties containing oleogel can be explained by this property. Moreover, at the end of the storage higher hardness of control samples, could be attributed to water and oil loss.

Looking into hardness in grilled products, it was increased until 3<sup>rd</sup> day for control but decreased in the following storage days. The hardness values of oleogel-containing samples were higher compared to the control group on all days except 1<sup>st</sup> day. Davidovich-Pinhas, Barbut, & Marangoni (2015) indicated that the temperature increase during cooking disrupts the hydrogen bonds in oleogel structures and destabilizes the polymer network, the hardness values of oleogel-containing products after cooking were affected by oil leakage from the oleogel structure and aggregation of the polymer network.





**Figure 3:** Hardness, adhesiveness, and springiness values of uncooked and grilled sea bass patties during storage.

Adhesiveness values, approximately between 0.1-0.5, showed a decreasing trend during storage in each group of uncooked patties, and control samples had lower adhesiveness compared to the oleogel groups in all days. Higher adhesiveness could be linked to oleogel-enhanced ability to retain water or oil. In grilled products, adhesion was close to zero and even zero itself in some repetitions (0 to 0.05). The cooking process eliminated the effects of the tissues responsible for adhesion, resulting in adhesion values being recorded close to zero or zero in patties.

There were fluctuations in the springiness of samples through storage. The springiness increased in the control group until the 1<sup>st</sup> day and between days 2-5 throughout the storage of uncooked products, while it was more stable in other groups. Bou et al. (2017) reported that storage of fish patties caused changes in springiness, although it showed an uncertain trend, and the lack of studies examining the instrumental texture of fish balls during storage, which was also a problem encountered in this study, caused difficulties in comparing the results.

### 3.3. Color Values

L\*, a\*, and b\* parameters were included in Table 1. An increase in the L\*-lightness values of

all groups was observed during storage. The increase can be attributed to the oxidation of lipids and proteins, which leads to a lighter appearance. There were no significant differences between groups (p>0.05). Similarly, in a study by Kara (2017), sea bass patties' L\* values were found to be 40.61 on the initial day. However, this value was increased to 58-68 at the end of storage, which is higher than our results. Reductions in a\* and b\* values were observed during storage. Interestingly, a\* reduction was less (13.1%) for lemon essential oil added oleogel group (Control;18.26% Oleogel;21.33%). Furthermore, a\*-redness was higher for oleogel groups at the end. The reduction in redness could be due to the degradation of pigments such as myoglobin and hemoglobin. The lesser reduction in the lemon essential oil added oleogel group could be due to the antioxidant properties of lemon essential oil, which helps to preserve the red pigments. Similarly, b\*-yellowness reduction was much more for the control group (22.8%) oleogel (20.32%)compared to lemon+oleogel group (13.00%), which could be due to protection against oxidative degradation. Overall, the presence of lemon essential oil and oleogel could have a protective effect against color degradation.

**Table 1.** L\*, a\*, b\*, results of uncooked sea bass patties during the storage period.

Parameter	Day	Control	Oleogel	Lemon+Oleogel
	Initial	36.36±11.53 <sup>a</sup>	38.24±12.17 <sup>a</sup>	$37.4\pm12.10^{a}$
	1	$36.10\pm11.48^{a}$	$38.91\pm12.37^{a}$	$39.16\pm12.42^{a}$
L*	2	$33.24\pm14.83^a$	$38.81\pm12.31^{a}$	$39.76\pm12.70^{a}$
$\mathbf{L}^{*}$	3	$37.82\pm11.99^a$	$39.52\pm12.55^{a}$	$39.22\pm12.44^{a}$
	4	$37.34\pm11.84^{a}$	$38.79 \pm 12.30^a$	$40.41\pm12.79^{a}$
	5	$37.48\pm11.89^{a}$	$41.59\pm13.23^{a}$	$40.80\pm12.92^{a}$
	Initial	10.13±3.34 <sup>a</sup>	10.64±3.55 <sup>a</sup>	10.38±3.36 <sup>a</sup>
	1	$8.61\pm2.79^{a}$	$9.68\pm3.19^{a}$	$9.24\pm2.81^{a}$
a*	2	$8.65\pm2.86^{a}$	$8.92\pm2.89^{a}$	$8.86\pm3.11^{a}$
a.	3	$8.39\pm2.77^{a}$	$8.86\pm2.92^{a}$	$9.09\pm2.95^{a}$
	4	$7.96\pm2.75^{a}$	$9.81\pm3.14^{a}$	$8.55\pm3.02^{a}$
	5	$8.28\pm2.73^{a}$	$8.37\pm2.70^{a}$	$9.02\pm2.96^{a}$
	Initial	31.40±10.82 <sup>a</sup>	$30.06\pm10.30^{a}$	29.29±9.46 <sup>a</sup>
	1	$25.28\pm8.22^{a}$	$27.40\pm9.12^{a}$	$26.39\pm8.43^{a}$
b*	2	25.23±8.31 <sup>a</sup>	$25.03\pm8.13^{a}$	$25.22\pm8.36^{a}$
D	3	$24.13\pm7.97^{a}$	$24.83\pm8.12^{a}$	$25.93\pm8.35^{a}$
	4	$23.89\pm8.10^{a}$	$27.25\pm8.73^{a}$	$24.62\pm8.34^{a}$
	5	$24.24\pm7.99^{a}$	$24.49 \pm 7.87^{a}$	$25.48\pm8.35^{a}$

The difference between the averages shown with different letters in the same row is significant (P < 0.05).

# 3.4. Microbiological Results of Uncooked Patties

It is expected that initial microbial counts of minced fish meat are higher than fish fillets. The initial TMAB and TPB results of groups were in the range of 3.88-4.31 log CFU/g and 3.28-4.47 log CFU/g, respectively (Table 2). Similarly, Çapkın (2008) found TMAB count of the initial found fish patties from tench (Tinca tinca) as 4.46±0.02 log CFU/g and the TPB count as 3.73±0.02 log CFU/g. Fish balls containing lemon essential oil showed lower TMAB count on the initial day than other groups, suggesting an immediate antimicrobial effect of lemon. Lemon essential oil is rich in citral, limonene, and linalool, compounds known for their broadspectrum antimicrobial activity against mesophilic bacteria. Although TMAB growth was faster for oleogel samples, lemon addition inhibited growth on all days except day 1. However, TPB counts were higher on initial, day 1 and day 5 for lemon-containing groups. This could be due to the gradual release of essential oil active compounds from the oleogel matrix, which may enhance the efficacy of lemon essential oil over time.

Citric acid from lemon might inhibit other competing bacteria, allowing psychrophilic bacteria to dominate and grow more rapidly. The maximum permissible limit of 7 log CFU/g (Swanson et al., 2011) was already exceeded on day 3 for the oleogel group (TMAB:7.34; TPB:7.31), which was significantly different. Control samples had the lowest TMAB counts (8.61 log CFU/g) at the end compared to the oleogel groups. Can (2012) stated that mirror carp fish balls TMAB values on day 0 were 4.48±0.1 log CFU/g, and increased up to 8.82±0.1 log CFU/g on day 9, which is similar to our results. In a study by İzci & Umut (2023), fish balls stored at 4 °C had TPB 4.67±0.01-9.04±0.01 log CFU/g during storage. In another study by Erol & İlhak (2015), TPB counts reached 7.55±0.18 log CFU/g for the mirror carp patties stored at 4 °C on the 6<sup>th</sup> day. In our study, closer ranges were obtained earlier on the 4<sup>th</sup> day, probably because of the storage temperature difference.

**Table 2.** Total mesophilic aerobic bacteria and total psychrophilic bacteria counts of uncooked sea bass patties.

TMAB	Control	Oleogel	Lemon+Oleogel
Initial	$4.31\pm0.13^{a}$	$3.94\pm0.30^{a}$	$3.88 \pm 0.07^{a}$
1	$5.00\pm0.41^{a}$	$5.23\pm0.08^{a}$	$5.55\pm0.22^{a}$
2	$6.64\pm0.29^{a}$	$6.69\pm0.18^{a}$	$6.43\pm0.24^{a}$
3	$6.70\pm0.28^{a}$	$7.34\pm0.12^{b}$	$6.87 \pm 0.07^{\mathrm{a}}$
4	$7.43\pm0.23^{a}$	$7.85\pm0.24^{a}$	$7.83\pm0.20^{a}$
5	$8.61\pm0.14^{a}$	$8.88 \pm 0.15^{a}$	$8.82 \pm 0.13^{a}$
TPB	Control	Oleogel	Lemon+Oleogel
Initial	$3.28\pm0.37^{a}$	$3.96\pm0.14^{b}$	$4.47 \pm 0.07^{\rm b}$
1	$5.18\pm0.15^{a}$	$5.37\pm0.12^{a}$	$5.60\pm0.13^{a}$
2	$6.39\pm0.24^{a}$	$6.58\pm0.36^{a}$	$6.25\pm0.15^{a}$
3	$6.90{\pm}0.04^{\mathrm{a}}$	$7.31 \pm 0.09^{b}$	$6.89 \pm 0.07^{\mathrm{a}}$
4	$7.41\pm0.19^{a}$	$7.37\pm0.09^{a}$	$7.33\pm0.28^{a}$
5	$8.79\pm0.08^{a}$	$8.66\pm0.06^{a}$	$8.97 \pm 0.08^{\mathrm{b}}$

The difference between the averages shown with different letters in the same row is significant (P < 0.05).

TPB counts were higher in lemon-containing groups on the initial day, day 1, and day 5. This suggests a selective antimicrobial effect, where lemon essential oil may suppress TMAB more effectively, thereby reducing microbial competition and allowing TPB to proliferate. Citric acid, a major component of lemon, is known to lower pH, which can inhibit many bacteria but may favor the growth of acid-tolerant psychrophiles.

# 3.5. Chemical Analysis of Uncooked Patties 3.5.1 pH

Uncooked fillets' pH was determined before patties preparation, which was found 6.52±0.09 (Table 3). After the preparation of patties, this value was decreased to 6.14. As discussed previously, the addition of lemon essential oil decreases the pH because of citric acid, and in the range pH 5 to 6, myofibrillar proteins are less

soluble and tend to aggregate. A lower pH for the lemon+oleogel-containing sample could explain this trend. Until the 4<sup>th</sup> day, lemon+oleogel-containing sample showed higher pH stability compared to other samples. Oleogel-added samples had the lowest pH on days 4, 5, and 6. All pH profiles demonstrated clear acidification as a function of time. All samples demonstrated a pH decrease at the end of the storage period.

Hemoglobin has been reported to be a major catalyst in fish. Reduction in pH may be related to lipid oxidation due to enhanced autoxidation of hemoglobin. The ability to form heat-induced gels is reduced at low pH because, when myofibrillar proteins approach their isoelectric point, they become less soluble and aggregate. Therefore, pH reduction can affect texture, which was confirmed with hardness results in previous sections.

Table 3: pH values of uncooked sea bass patties.

Days	Control	Oleogel	Lemon+Oleogel
Initial	$6.25\pm0.07^{a}$	$6.24\pm0.02^{\mathrm{b}}$	$6.14\pm0.01^{b}$
1	$6.16\pm0.13^{a}$	$6.08\pm0.03^{\mathrm{b}}$	$6.11\pm0.02^{ab}$
2	$6.32\pm0.10^{a}$	$6.21\pm0.03^{b}$	$6.21\pm0.11^{ab}$
3	$6.30\pm0.06^{a}$	$6.20\pm0.01^{b}$	$6.16\pm0.03^{b}$
4	$6.23\pm0.04^{a}$	$5.9\pm0.02^{\rm b}$	$6.22\pm0.04^{a}$
5	$6.08\pm0.08^{a}$	$5.37 \pm 0.04^{b}$	$5.66 \pm 0.03^{b}$

The difference between the averages shown with different letters in the same row is significant (P < 0.05).

## 3.5.2. Peroxide value

Lipid oxidation is one of the most significant factor affecting the quality deterioration of stored aquatic products. Parameters that influence the extent of oxidation in lipids include the sensitivity to lipid oxidation, high of unsaturated acids concentration fatty (particularly polyunsaturated fatty acids), exposure of the product to high temperatures, the presence of pro-oxidants and antioxidants, and processing conditions such as grinding (Delgado-Pando et al., 2011). Increased exposure to oxygen due to grinding the sea bass may affect the fatty acid profile of the fish meat, and the use of a fatty fish species is expected to negatively contribute to lipid deterioration in the patties. In general,

fish oil is composed of approximately 20% saturated fatty acids and 80% unsaturated fatty acids, indicating a much higher proportion of unsaturated fatty acids compared to other animal fats. Considering all these factors, a significant portion of the spoilage in fish occurs in fish oil, and this spoilage manifests as rancidity in the final product, leading to sensory rejection. Since the initial products of unsaturated fatty acid oxidation are peroxides, the determination of PV can reveal even subtle rancidities that are not perceptible sensorially in the early days of storage. The PV, in brief, indicates the amount of oxygen bound to the molecular structure of fats and provides information about the oxidative stability of the analyzed product (Çakır, 2021). PV is measured to evaluate the formation of primary oxidation products and defines the degree of lipid oxidation. In the analysis of the fresh fish fillet sample, the PV was recorded as 13.88 meq O<sub>2</sub>/kg. This value increased after patties preparation, but the lemon sample had the lowest value compared to others (Table 4). The PV was found to range between 15.22 and 20.66 meq O<sub>2</sub>/kg on the initial day. The difference between the fish fillet and the sea bass patties is attributed to the oils and oleogels added to the patties. PV should generally be below 25 meg O<sub>2</sub>/kg in fatty food products (Elgin, 2019). In all products, the PV remained below this threshold limit throughout the storage period. The PV of the control group was found to be lower than that of the other two groups prepared with oleogel at the end. Significant difference was only observed on the initial day. These results indicate that the addition of oleogel did not contribute to protecting the lipids in the sea bass patties from primary oxidation. Indeed, some studies align with these findings. Delgado-Pando et al. (2011) found that despite low lipid oxidation levels, replacing animal fats with healthier combinations in Frankfurter-type formulations resulted in increased lipid oxidation. It should also be noted that oleogel systems, due to their large surface area, were more exposed to pro-oxidants in the environment, leading to increased oxidation levels. Exposure to high temperatures is also a prominent factor among those determining the formation of lipid (Delgado-Pando oxidation et al., 2011: Panagiotopoulou et al., 2016).

Table 4: Peroxide values in uncooked sea bass patties.

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Days	Control	Oleogel	Lemon+Oleojel
Initial	18.55±2.24 <sup>a</sup>	20.66±2.36 <sup>b</sup>	15.22±3.67 <sup>a</sup>
1	$17.75\pm5.75^{a}$	$16.00\pm1.00^{a}$	$18.75\pm1.75^{a}$
2	$8.66\pm0.94^{a}$	$7.66\pm0.0^{\mathrm{a}}$	$7.22\pm0.58^{a}$
3	$13.00\pm3.14^{a}$	$17.44 \pm 1.40^{a}$	$17.00\pm2.60^{a}$
4	$15.55\pm2.12^{a}$	$16.77\pm0.87^{a}$	19.44±2.81 <sup>a</sup>
5	$16.22\pm2.30^{a}$	$16.55\pm0.96^{a}$	$16.88 \pm 0.68^{a}$

The difference between the averages shown with different letters in the same row is significant (P < 0.05).

As indicated in a study on the partial replacement of beef tallow with oleogels prepared with sesame oil and beeswax in beef burgers, the higher PV's of the oleogels compared to the control were highlighted because of heat exposure during oleogel production (Moghtadaei et al., 2018). The catalytic effect of degradation products formed in lipids is another issue to consider. Lipid degradation progresses exponentially, and the products formed as soon as degradation begins or those already present in the environment can act as catalysts for further deterioration. The excessive release of FFA can undergo oxidation, leading to rancidity in the product and an increase in PV due to the formation of oxidation products (Toksöz, 2020). Accordingly, the higher PV observed in oleogelcontaining patties could also be attributed to the catalytic effect of lipid degradation products.

The peroxide analyses conducted revealed fluctuations on different days across all groups of patties. These results can be attributed to the behavior of oil degradation, as peroxides measured in the peroxide analysis are primary oxidation products. The observed decrease in PV following an initial increase in the sea bass patties is because peroxides, which show an increasing trend in the early stages of lipid oxidation. decrease during tend to propagation stage because their formation rates are lower than their degradation rates. In other words, hydrogen peroxides that form in the initial stages of oxidation convert into secondary oxidation products (such as malondialdehydes) in the subsequent stages. Similar results were observed in a study by Elgin (2019), where patties prepared with added cranberry extract and then frozen for shelf-life analysis after cooking showed an increase in PV during the first three months, followed by a decrease. Likewise, in a study investigating the oleogelation of fish oil with various natural waxes, PV increased throughout the storage period at 35°C for 21 days in the oleogel samples, while the control group, consisting only of fish oil, initially showed an increase in PV followed by a decrease. The authors attributed this to the progression of degradation levels, whereby primary degradation products degrade into secondary oxidation products, leading to lower PV (Hwang et al., 2018). In a study on the replacement of beef tallow with oleogel in beef patties, the control group samples, which did not have beef tallow replaced, showed a decrease in PV during storage, followed by an increase. In groups with 75% and 100% replacement, no significant effect of storage duration on the PV of oleogels was observed. However, in the 50% replacement samples, PV increased on the 9th day and subsequently decreased in the following storage days. This phenomenon was attributed to the formation of peroxides from initially oxidized FFA, which then converted into secondary oxidation products (Çalişkan, 2024).

The PV of patties made from oleogels with added lemon essential oil showed that the lemon essential oil did not exhibit the expected effect of delaying the oxidation in the patties. This can be attributed to the fact that lemon essential oil was used only by mixing it with sunflower oil without any additional encapsulation process to retain the oil in the system. Similarly, in a study investigating the effects of encapsulated citric acid on the shelf life of sea bass patties (Bou et al., 2017), it was reported that hydroperoxides, a primary oxidation product, increased during the first three weeks of storage, with no significant difference observed during the 4-8 weeks of storage. Additionally, the study found that patties with encapsulated citric acid showed lower levels of hydroperoxides compared to the control group, but there was no notable difference in PV, indicating that the addition of pure anhydrous citric acid did not have a positive effect. Furthermore, the essential lemon oil used in this study is also sensitive to oxidation, and considering the catalytic effects of lipid degradation, the observed results are understandable.

### 3.5.3. Free Fatty Acid

FFAs are formed through the hydrolysis of in triglycerides by lipases bonds (Hamidioglu et al., 2022). FFA value serves as a legal limit and is used to determine the usable life of frying oils. The FFA value of the fish fillet was recorded as 0.0658% (Table 5). For sea bass patties, it was found to range between 0.0909% and 0.1065% which was higher than fillet. In European countries, legal limits for the FFA content of frying oils must be within the range of 0.9%-2.5% (Çakır, 2021). The results presented in this study were lower than the legal limit throughout the storage period. However, the FFA values increased within each group throughout storage. In a study by Hamidioglu et al. (2022), oleogels prepared using rice bran wax and candelilla wax as oleogelators with hemp oil were used as fat replacements in beef patties. It was demonstrated that the FFA content increased in all products.

Similar to the PV results, the control group exhibited lower FFA values compared to those with oleogel. Significant differences were observed on days 4 and 5. A study reporting similar results indicated that the FFA value could increase depending on the amount of oleogelator used and the preparation time in sesame oil-based oleogels. The researchers noted that an increase in the amount of wax in the oleogel and an extension of the cooling time during the preparation of the oleogel significantly elevated the FFA value (Moghtadaei et al., 2018). Accordingly, it can be concluded that the preparation process also influences the FFA content in the final product.

**Table 5:** Free fatty acid values in uncooked sea bass patties.

Days	Control	Oleogel	Lemon+Oleojel
Initial	$0.0909\pm0.014^{a}$	$0.1034\pm0.009^{a}$	$0.1065\pm0.005^{a}$
1	$0.1762\pm0.050^{a}$	$0.1762\pm0.050^{a}$	$0.1410\pm0.0^{a}$
2	$0.1410\pm0.047^{a}$	$0.1880\pm0.047^{a}$	$0.1410\pm0.0^{a}$
3	$0.1285\pm0.011^{a}$	$0.1567\pm0.012^{a}$	$0.1504\pm0.008^{a}$
4	$0.1316\pm0.009^{a}$	$0.1911 \pm 0.005^{b}$	$0.1661\pm0.005^{\mathrm{b}}$
5	$0.1441\pm0.011^{a}$	$0.1942 \pm 0.005^{b}$	$0.1942 \pm 0.005^{b}$

Although it was not significant, lemon+oleogel group showed slightly lower FFA values than the oleogel-only group on days 1, 2, 3, and 4, suggesting a partial protective effect from lemon essential oil, probably due to citral and limonene compounds. These compounds may help scavenge free radicals and thereby slow lipid degradation.

### 3.5.4. TVB-N Results

TVB-N is an index/indicator known as the product of bacterial spoilage and endogenous enzymes and is used to determine the chemical quality of aquatic products. TVB-N for fresh sea bass fillet was found to be  $11.38\pm1.88$  mg N/100 g on the initial day (Table 6), being consistent with the results of the study from Chen et al. (2022), who found 11.54 mg N/100 g. When

the TVB-N analysis results were performed throughout storage, the control group values varied between  $7.46\pm0.86$ ,  $20.22\pm0.18$ ; the oleogel group values varied between 9.14±0.64 and 22.80±0.78; and the lemon group values varied between 6.76±0.10 and 19.52±0.88. No statistical difference was found between the groups in the analysis periods on initial, 1, 2, and days (p>0.05). There were statistically significant differences on the 4<sup>th</sup> and 5<sup>th</sup> days (p<0.05). After that, the lowest TVB-N result was obtained in the lemon group on the 5<sup>th</sup> analysis day and on the 4th day in the control group. At the end of the storage, TVB-N was still lower than the deteriorated fish threshold limits (Yue et al., 2023).

**Table 6:** Determined total volatile basic nitrogen values in uncooked sea bass patties.

Days	Control	Oleogel	Lemon+Oleogel
Initial	$7.46\pm0.86^{a}$	$9.14\pm0.64^{a}$	$6.76\pm0.10^{a}$
1	$9.13\pm1.52^{a}$	$13.40\pm0.06^{a}$	$12.59\pm1.29^{a}$
2	$10.31 \pm 1.05^{a}$	$12.52\pm0.69^{a}$	$12.32\pm0.01^{a}$
3	$13.94\pm3.90^{a}$	$13.55\pm1.82^{a}$	$10.90\pm1.33^{a}$
4	$14.86 \pm 0.35^{a}$	$18.07 \pm 1.70a^{b}$	$19.37 \pm 0.46^{b}$
5	$15.50\pm1.39^{a}$	$22.41\pm5.36^{b}$	$14.95\pm0.97^{a}$

### 3.5.5 Sensory Results

Sensory analyses were conducted on the initial day to investigate consumer taste in addition to chemical analyses. The reason patties have been diversified over the years is not because of perfect patty formulation has not yet been reached, but because different flavors emerge when different ingredients are added, producing products that appeal to different sensory profiles. Consumer expectations or preferences are highly diverse and variable. In this context, while patties are made with beef meat in some regions, in others they are prepared with lentils or only bulgur wheat (fellah meatballs, uncooked meatballs). This reflects both consumer preferences and financial wellbeing. In addition to these, as mentioned, studies are not conducted for perfect patty formulations that we have not yet reached, and it is science to diversify products and work on trying the untried. In this context, this study was carried out using sea bass, a relatively valuable fish, and oleogel, which we thought would make a difference, was also included in the patties. Sensory analysis results indicated that fish patties with oleogel were more prominent in consumer preferences than the control (Figure 4).

When the sensory analysis results of fish patties were examined, the fish patty with oleogel was superior to the control in terms of appearance, texture, smell, taste, and acceptability. The control group received the lowest score in all evaluation parameters. The patty containing oleogel stood out clearly

according to the taste and overall impression evaluations, followed by the patty containing oleogel with lemon essential oil and the control group. It is also understood from the responses given to the last question of the sensory test, which is the question of "would you buy it?", that the control patty prepared with sunflower oil is not preferred. According to these results, it has been shown that adding oleogel to fish patties has

a positive effect on sensory preferences. In contrast to this study, studies have been conducted on chicken patties prepared using different proportions of oleogel, showing that the addition of oleogel or the increase in its proportion in the patties is inversely proportional to sensory acceptability (Tüysüz, 2024). The reason for this contrast may be related to the type of meat used or the oleogel amount.

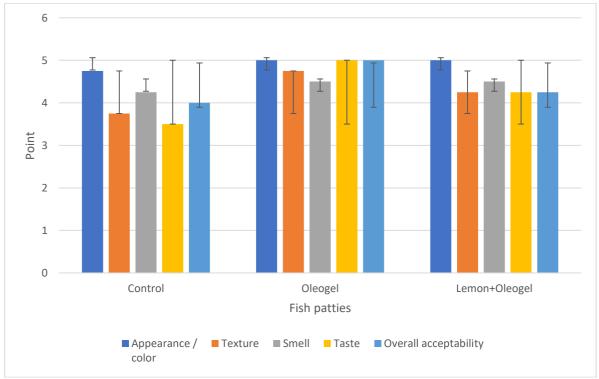


Figure 4. Sensory scores of cooked sea bass groups on the initial day.

# 4. CONCLUSION

Physical analysis of both uncooked and grilled fish patties for a period of 5 days is recorded with a very little amount of lemon added oleogels. Oleogels created a network that can trap water, reducing water loss during storage, but increased cooking loss.

The gradual pH decline in sea bass patties—especially in oleogel-added samples—reflects acidification during storage, likely driven by citric acid and lipid oxidation, which in turn affected protein solubility and enhanced the texture softness. The higher PV and FFA values in patties containing oleogel compared to the control may be due to the continuous exposure to heat and oxygen during the preparation period of oleogels. The presence of a tiny amount of lemon essential oil in oleogel showed a positive effect on color stability. However, they did not show

any significant effect on TMAB and TPB. Despite an overall increase in TVB-N values during storage, samples lemon group showed the most effective inhibition by day 5. The contribution of oleogel to the preferability of fish patties showed a positive impact on taste, smell, appearance, and overall acceptability scores. Results indicate that using essential oils in oleogels could align with trends favoring fewer synthetic additives. Furthermore, enhanced softness of texture contributes to improved mouthfeel in fish patties, showing oleogel's value in patties for commercial purposes. In the future, it will be necessary to sustain the texture and oxidative stability of seafood products with oleogels. For this purpose, controlled release of lemon oil via nanogels could improve gel strength and oxidative protection, creating a microbarrier against spoilage.

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### **CONFLICT OF INTEREST**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## **AUTHOR CONTRIBUTIONS**

Fiction: TÇ, EB; Literature: TÇ, EB; Methodology: TÇ, EB, SM; Performing the experiment: TÇ, EB, SM; Data analysis: TÇ, EB; Manuscript writing: TÇ, EB, All authors approved the final draft.

## ETHICAL STATEMENTS

None

## DATA AVAILABILITY STATEMENT

Data supporting the findings of the present study are available from the corresponding author upon reasonable request.

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