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Assessing Quality Changes in Carp Fish Meatballs Enriched with Different Mustard Oil Levels

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Abstract: This study aimed to assess the microbiological and sensory changes in carp (*Cyprinus carpio*) meatballs enriched with black mustard seed oil at different concentrations (3% and 6%). Carp fish meat was minced and divided into three distinct meatball groups, which were subsequently stored in sealed refrigerator bags at 4±1ºC. Sensory (appearance, taste, smell, texture and overall acceptability) and microbiological (total aerobic mesophilic bacteria, psychrotrophic bacteria, yeast and mold) analyzes were performed on the 1st, 7th and 14th days of storage. Significant differences were observed between the control group and the essential oil added groups in terms of microbiological properties during storage ($p<0.05$). The results of sensory analyses showed that there were statistically significant differences between the groups, and the control group was the most preferred group across all parameters. According to the results of research, it has been determined that the addition of mustard seed essential oil reduces the number of bacteria and delays spoilage.

Keywords: *Cyprinus carpio*, Fish ball, Microbiological analysis, Mustard oil, Shelf life, Sensory changes analysis

INTRODUCTION

Carps constitute a large part of inland water fishing in Turkish. Carp (*Cyprinus Carpio*) be-longs to the Cyprinidae family and is a cold-resistant fish species with high economic value that is widely found in almost all waters in Turkey. In addition, it can easily adapt to the environment and its feed intake and evaluation is high. It can develop rapidly under suitable conditions (Balık et al., 2006; Çağıltay, 2011; Süle, 2011).

It is known that fish and other aquatic products, which are not popular with consumers in Southwest Asia and Far East countries, European countries and the United States, are trans-formed into new products with different processing techniques. Valuable fish species are of-ten sold as fresh, frozen fillets or as smoked, salted, marinated or canned products. However, there are many types that are traditionally processed into different shapes made from minced meat. Fish mince allows the use of fish species with low economic value, especially those that cannot find a sufficient market demand due to their bones. It converts the remaining edible parts after the fillet process into processed products such as fish mince (Yanar and Fenercioğlu, 1999; Süle, 2011).

The mustard plant, renowned for its rich content, has been employed in alternative medicine since the discovery of its anti-inflammatory properties centuries ago. Widely recognized as a spice and sauce in kitchens, mustard provides many health benefits owing to its abundant nutritional content. As a herbaceous plant with yellow flowers, it belongs to the cruciferous family (*Brassicaceae*). The use of mustard in foods not only enhances flavor but also extends the product's shelf life due to its antimicrobial activity. Among the approximately ten different species, three are commonly used as spices: black (*Brassica nigra*), white or yellow (*Brassica alba*) and brown (*Brassica juncea*). Although yellow and brown mustard types are popular choices in many countries, black mustard, distinguished by its unique, intense, sharp, and bitter taste, boasts a higher oil content. Black mustard seed contains allyl isothiocyanate, which imparts a characteristic aroma, and various beneficial components such as amino acids, oligosaccharides, minerals, and fatty acids. In addition, mustard seeds exhibit antioxidant effects due to bioactive compounds like phenolic acid and phytin. Black mustard has a strong antimicrobial effect against bacteria, yeast and mold species, especially many food pathogens. This effect is attributed to the presence of allyl isothiocyanate, parahydroxybenzyl isothiocyanate, and glucosinolates. In traditional medicine, black mustard seeds are used as a laxative, diuretic, stimulant, emetic, appetite stimulant, anti-asthma, anti-cough, anti-congestion, anti-neuralgia and more (Mejia-Garibay et al., 2015; Gıdık, 2016; Tırancıoğlu, 2017; Şahiner, 2019; Đorđević et al., 2020; Lakwani, 2021; Lakwani et al., 2022).

Mustard oil is a vegetable oil produced by squeezing the seeds of the mustard plant using various methods, and has been a preferred choice for its health benefits since ancient times up to the present day. It is rich in monounsaturated and polyunsaturated fatty acids (MUFA and PUFA), as well as omega-3 and omega-6 fatty acids (Anonymous, 2023; Đorđević et al., 2023). Black mustard oil possesses strong

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antimicrobial properties due to its allyl isothiocyanate (Çoşkun, 2006).
This study aimed to determine the microbiological and sensory changes in carp (*Cyprinus carpio*) meatballs enriched with black mustard seed oil at two different concentrations (3% and 6%).

MATERIALS AND METHODS

Material

The carp fish used in the research were supplied from a local seller in Ardahan Province, and the black mustard seed oil was procured from the market.

Method

Carp fish, with an average weight of approximately 5 kg, were obtained in adherence to cold chain regulations and brought to the Atatürk University Fisheries Faculty Processing Laboratory. To prepare the fish balls, the carp fish were ground into minced meat and 10% breadcrumbs, 5% onion, 2% salt, 0.5% black pepper, 0.5% red pepper, 0.5% cumin were add-ed and thoroughly kneaded. Then, the fish balls were divided into 3 groups: control (A), 3% black mustard seed oil addition (B) and 6% black mustard seed oil addition (C). The meatballs were packaged in sealed refrigerator bags and stored at 4±1°C, and sensory evaluations (appearance, taste, smell, texture and overall acceptability) and microbiological analyses (total aerobic mesophilic bacteria, psychrotrophic bacteria, yeast and mold) were conducted on the 1st, 7th and 14th days of storage.

Microbiological Analysis

A meatball sample (10 g) was taken, 90 ml of sterile saline was added and homogenized in the Stomacher device (Lab Stomacher Blender 400-BA 7021 Seward Medical, England). Total aerobic mesophilic and psychrotrophic bacteria were enumerated using Plate Count Agar and incubated for 2 days at 30°C and 10 days at 4°C, respectively. Potato Dextrose Agar was used for yeast-mold counting and incubated at 25ºC for 5 days. The results of the microbiological analysis are given as log cfu/g (Baumgart et al., 1986; Anonymous, 1992; Halkman, 2005).

Sensory Analysis

Meatball samples were evaluated in terms of appearance, taste, smell, texture and overall acceptability parameters by a panelist group of 8 people. Fried meatball samples were scored on a scale of 1 to 5 (Altuğ Onoğur and Elmacı, 2011).

Statistical Analysis

The study was designed following a completely randomized trial plan and conducted with two replications. Data evaluation was performed using the SPSS 17.0 program, and the results were compared using Duncan's multiple comparison tests.

RESULTS and DISCUSSION

Microbiological Analysis

These naturally occurring antimicrobial and antioxidant agents are highly complex mixtures of often hundreds of individual aromatic essential oil compounds extracted from different plant materials such as leaves, barks, stems, roots, flowers and fruits (Koç, 2023). The results of the microbiological analysis of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) are given in Table 1. Total aerobic mesophilic bacteria are important parameters that provide information on food hygiene throughout storage and pro-duction of food (Dinçoğlu and Çalışkan, 2022). The acceptable limit for total aerobic mesophilic bacteria in fresh fish and seafood is set at 7 log cfu/g (ICMSF, 1986). Notably, these limits were exceeded in the control group on the 14th day of storage. However, it was observed that the total bacterial count in the mustard oil-added groups remained below the acceptable limit throughout the storage period. This may be due to the protective role of mustard oil against spoilage bacteria (Çoşkun, 2006). Statistically significant differences were found between the groups during storage ($p<0.05$). While the highest total aerobic mesophilic bacteria count was in the control groups, the lowest bacterial load was in the group C. Application of 6% mustard essential oil was more effective against TAMB. It has been observed that the antibacterial effect increases as the essential oil concentration increases. Essential oils are produced from parts of plants as defense mechanisms against microorganisms. Lemongrass leaf essential oil has been reported to have an important role in extending the shelf life of fish cakes by reducing the microbial load (Kamona and Alzobaay, 2021). In our research findings, it was determined that mustard oil has an inhibitory effect on bacteria depending on concentration. Similar to the present findings, Keser and İzci (2020) noted that the control samples exceeded the limit value (7 log cfu/g), while the essential oil-added meatball samples did not on the 7th day of storage. Hać-Szymańczuk et al. (2021) found that the number of mesophilic aerobic bacteria increased as the storage time increased in all groups in the chicken meatballs prepared with different rosemary formulations. Dinçoğlu and Çalışkan (2022) reported a low-er number of TMAB in meatball groups containing lavender essential oil compared to the control groups ($p<0.05$). At the end of storage, it was determined that the mackerel meatball groups with herb extract (thyme, rosemary and basil) showed a significant difference ($p<0.05$) in terms of TAMB numbers compared to the control group (Balıkçı, 2015).

Psychrotrophic bacteria play a significant role and cause changes in texture, smell and taste through the production of different metabolic compounds (aldehydes, volatile sulphides, bio-genic amines and ketones) (Ozogul and Uçar, 2013). During storage, the control group (A) exhibited the highest psychrotrophic bacteria count while the group C had the lowest. The difference in mustard essential oil concentration significantly influenced the growth of psychrotrophic bacteria in the samples ($p < 0.05$). Although the number of psychrotrophic bacteria count increased in all groups during the storage period, the highest increase was

detected in the control group. The number of psychrotrophic bacteria showed significant differences ac-cording to storage days and groups ($p<0.05$). Özalp Özen and Soyer (2017) emphasized that the addition of plant extract reduced the psychrophilic bacteria count in mackerel minced meat stored by adding different herbal extracts (green tea, grape seed and pomegranate peel). Erecevit Sönmez et al., (2020), reported that the number of psychrotrophic bacteria in carp fish prepared with different amounts of ginger (0.1%, 0.25%, 0.5%) oil was lower in the 0.5% added group than in the other groups. Şevik et al. (2022) emphasized that the addition of herbal extract reduced the total number of aerobic psychrophilic bacteria. These findings align with the results of our study.

The increase in the number of yeast and mold in the control group samples during the storage period was higher than in the mustard seed oil added groups. Yeast and mold numbers increased in all groups during the storage period. It was determined that there were significant differences between the groups during storage ($p<0.05$). Can (2012) found that adding different amounts of eugenol (0.5% and 1%) to meatballs prepared with mirror carp reduced the number of yeast and mold compared to other groups. In another study, Can and Emir Çoban (2012) noted that applying 1% thymol reduced the number of yeast and mold compared to other groups. Erol and İlhak (2015) reported that there was a slight increase in the yeast and mold numbers of meatballs made from thymol mirror carp, although it varied according to the groups.

Table 1. Microbiological analysis results of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) (log cfu/g).

		Samples		
	Days	A	B	C
Total aerobic mesophilic bacteria	1	3.83±0.14^{a}	2.28±0.08^{a}	2.25±0.14^{a}
	7	5.48±0.12^{b}	4.26±0.33^{b}	4.09±0.16^{b}
	14	7.07±0.98^{c}	6.43±0.14^{c}	5.94±0,08^{c}
Psychrotrophic bacteria	1	3.92±0.05^{a}	2.53±0.07^{a}	2.59±0.01^{a}
	7	5.26±0.14^{b}	4.75±0.07^{b}	4.35±0.12^{b}
	14	7.33±0.31^{c}	6.68±0.14^{c}	6.22±0.31^{c}
Yeast and mold	1	2.09±0.13^{a}	2.00±0.00^{a}	2.00±0.00^{a}
	7	2.83±0.06^{b}	2.26±0.17^{a}	2.06±0.08^{a}
	14	3.78±0.33^{c}	2.88±0.16^{b}	2.78±0.04^{b}

All values are the mean ± standard deviation. Different letters (a, d) in the same column indicate significant differences ($p<0.05$). A: control; B: 3% black mustard seed oil; C: 6% black mustard seed oil

Sensory Analysis

Sensory properties are important parameters in the acceptance of the foods by consumers (Shafaghi Rad and Nouri, 2023). Essential oils have a unique strong odor, taste and color. Oxygenated derivatives formed by oxidation of terpenes give the essential oil its unique smell, taste and therapeutic properties (Çelen, 2006). The results of sensory analysis of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) are illustrated in Figure 1. The control group (A) samples were the most preferred by the panelists across all criteria (appearance, taste, smell, texture and overall acceptability). It is suggested that the extracts used give an unusual taste and smell to the fish, leading to a lower overall acceptability. Statistically significant differences were observed between the groups in terms of all parameters ($p<0.05$). Sensory parameters showed a significant decrease in all sample groups parallel to the storage duration. Sinalbin (in white mustard) and sinigrin (in black mustard) produce the pungent mustard oil (Yaralı, 2023). It was determined that essential oil con-centration was given low scores by the panelists in terms of sensory parameters. Similarly, Mattje et al. (2019) reported that fish burgers containing ginger oil received low scores from panelists due to their intense ginger flavor in terms of taste and overall evaluation. In line with our study, Zhang et al. (2021) noted decreases in color, odor, texture and overall acceptability parameters in grass carp supplemented with thyme essential oil during storage. Emir Çoban and Tuna Keleştemur (2017) reported that catfish burgers prepared using essential oil received the highest score in sensory criteria when 0.4% Zataria multiflora Boiss essential oil was added. Corbo et al. (2009) emphasized that no significant difference was detected between sensory control and treated cod fish burgers at the beginning of storage. In another study, the use of rosemary extract has been reported to improve the sensory quality of mackerel burgers (Uçak et al., 2011).

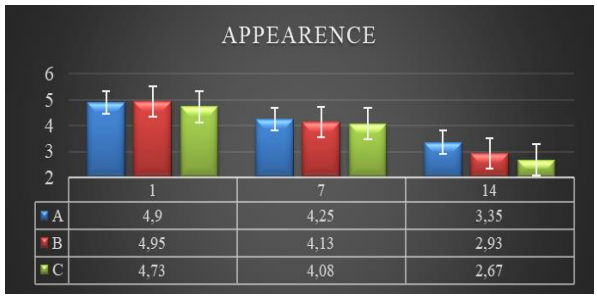

Figure 1. Changes in appearence parameters of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) (log cfu/g). A: control; B: 3% black mustard seed oil; C: 6% black mustard seed oil

Figure 2. Changes in taste parameters of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) (log cfu/g). A: control; B: 3% black mustard seed oil; C: 6% black mustard seed oil

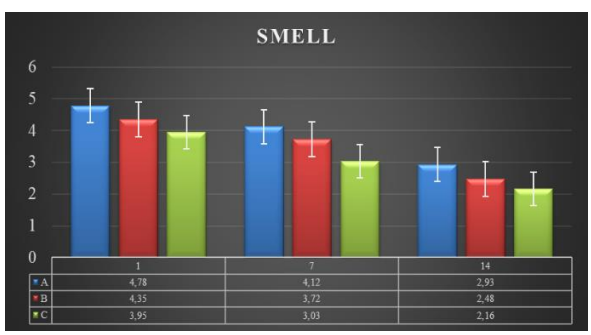

Figure 3. Changes in smell parameters of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) (log cfu/g). A: control; B: 3% black mustard seed oil; C: 6% black mustard seed oil

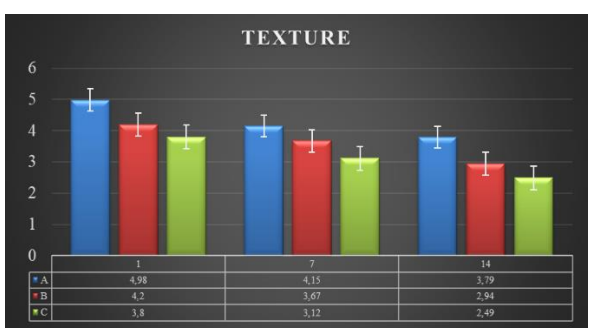

Figure 4. Changes in texture parameters of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) (log cfu/g). A: control; B: 3% black mustard seed oil; C: 6% black mustard seed oil

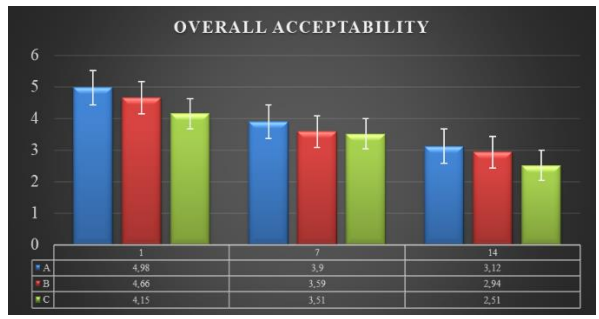

Figure 5. Changes in overall acceptability parameters of carp (*C. carpio*) meatballs enriched with black mustard seed oil at different rates (3% and 6%) (log cfu/g). A: control; B: 3% black mustard seed oil; C: 6% black mustard seed oil

CONCLUSION

In this study, it has been determined that black mustard seed oil has positive effects on the microbiological properties of carp fish meatballs and increases their shelf life. As a result of sensory analysis, it was determined that the control group samples were liked the most in terms of all parameters, and it is predicted that this is due to the essential oil used giving the fish an unusual taste and smell. It has been observed that such important essential oils, which provide many health benefits, can be easily used in fish and fish products. It is also important to prepare such meatballs and offer them as an alternative product to individuals who do not consume fish.

COMPLIANCE WITH ETHICAL STANDARDS

Conflict of interest

The authors declared no conflict of interest. All the authors read and approved the final manuscript.

ETHICAL APPROVAL

Not required.

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CONSENT FOR PUBLICATION

Applicable.

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