Food Science and Technology
ISSN 0101-2061 (Print)
ISSN 1678-457X (Online)
DOI: https://doi.org/10.1590/fst.01318

Influence of different essential oils on marinated anchovy (*Engraulis encrasicolus* L. 1758) during refrigerated storage

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Abstract

This study aims to determine the effect of different essential oils on the physical, chemical and microbiological properties, and the shelf life of the cold marinated anchovies (*Engraulis encrasicolus* L. 1758) stored at 4 ± 2 °C. The anchovies were marinated for twenty-four hours at 4 ± 2 °C in brine, consisting of salt (10%), alcohol vinegar (4%) and citric acid (0.2%), and then stored for six months at 4 ± 2 °C in sunflower seed oil. The study consists of the following groups: only sunflower seed oil (group A), 0.1% rosemary oil (group B), 0.1% coriander oil (group C), 0.1% laurel oil (group D) and 0.1% garlic oil (group E). When the results of the TVB-N and microbiological analysis were considered, it were established that all the groups maintained their freshness during the six month period, and that group E containing garlic oil, had the minimum TVB-N ratio and the lowest total mesophilic bacteria count by the end of the storage period.

Keywords: storage life; anchovy; marinade; TVB-N; microbiology.

Practical Application: Cold marinate is an aromatic and delicious product. Different food additives extend the shelf life of foods. The influence of natural additives on the shelf life of sea foods has been examined recently. Adding different essential oils on cold anchovy marinade were prolong the shelf life. Essential oils are safely for marinated anchovy and fit for human consumption for a period of 6 months.

1 Introduction

Of the nearly six hundred and eight thousand tons of seafood that was caught in Turkey in 2013, one hundred and eighty thousand tons were anchovies (Tuik, 2015). The anchovies caught in the East and West Black Sea regions are consumed fresh, while the surplus is either frozen or processed in fish flour and oil factories. The processing and marination of anchovies that were frozen during the fishing season is beneficial, both in terms of extending employment throughout the year and of raising fish prices by increasing the product range available on the market. Cold anchovy marinate is an aromatic product, which is obtained as a result of thoroughly bleeding out and filtering the thawed fish in water, then placing them in acid + salt brine. Turkey ranks somewhat below the world average in terms of seafood consumption. The per capita fish consumption might be increased by encouraging fish consumption outside the fishing season. The marination process is faster and easier than other processing methods such as salting, conservation and drying. Sea foods is treated in different acid and salt solutions, and end product being sealed and stored in airtight packaging or in different oils.

The effects of spices are depending on the microorganisms type and the chemical composition of spices, essential oil concentration and materials and method used in addition to doing extraction process (Farag et al., 1989; Ehrich et al., 1995; Dülger et al., 1999). The phenols within the essential oils change the permeability of the cell by altering the altering the membrane structure of the bacterium cell, thereby increasing its permeability and ultimately disrupting the osmotic pressure balance and the structure of the proteins that hold together the bacterium's cell wall. Once the cell permeability has been disrupted, water loss due to the intensity of the H+ and K+ cations within the bacterium's cytoplasm results in the death of the cell (Heipieper et al., 1991; Stammati et al., 1999). Coriander and garlic are listed among the herbs and spices that could be used for preserving foodstuff in terms of their antimicrobial effects (Beuchat & Golden, 1989).

Thanks to globalization and busy living conditions, food-processing technology has seen a rapid development in recent years, and people have been increasingly on the lookout for products lining the market shelves that offer different tastes and have a different appearance. Products enriched with different flavors and spices have found a place on market shelves. In previous study (Turan et al., 2017) we determined the lipid oxidation and sensory attributes of anchovy marinated which had been used the same marinate solution and essential oils. However, in this study we have tried to change the overall product by adding different essential oils to the traditional cold marinate. And we aimed to study (i) the storage life of marinated anchovies; and (ii) the effect of essential oils on the physico-chemical and microbiological qualities of the marinated anchovy products.

Received 19 Jan., 2018
Accepted 09 Sept., 2018
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2 Materials and methods

2.1 Raw materials

Frozen anchovies (*Engraulis encrasicolus* L. 1758) were provided by the Başoğlu Fish Company (Sinop, Turkey). In total, 36 kg of frozen anchovies were used in the study. The anchovies were transferred to the laboratory in a plastic fish box and thawed in cold water. Before marinating, the fish were beheaded and gutted, the bones removed manually to obtain fillets. The anchovies were washed with cold water and soaked in 5% salt-water solution until the blood had drained away. Alcohol vinegar was used in accordance with the TS1880 EN 13188 standard (Turkish Standard, 2003). The brine used which is composed from pure rock salt (this was suitable for Turkish Food Codex) and after brine process, sunflower seed oil (Bizim Oil, Ülker, İstanbul, Turkey) was added on marinade. The essential oils added were rosemary oil (*Rosamary officinalis*), coriander oil (*Coriander sativum*), laurel oil (*Lauri expressum*) and garlic oil (*Allium sativum*) (0.1%).

2.2 Marination process

The marination process was performed in 4% (v/v) alcohol vinegar+10%salt+0.02% citric acid solution at 4 °C for 24h. The solution to fish ratio was 2:1. After marination process, fish fillets were drained from solution. All of them were placed in the plastic box and sunflower oil was added. These products were divided into five groups. Groups were: including only sunflower seed oil (group A), 0.1% rosemary oil (group B), 0.1% coriander oil (group C), 0.1% laurel oil (group D) and 0.1% garlic oil (group E), respectively. And they stored at 4 ± 2 °C for 6 months in a refrigerator.

Physicochemical and microbiological analyzes were carried out during the first day of storage and during 6 months. The study was consisting of three replicate.

2.3 Physicochemical analysis

Manthey et al. (1988) method was used for measuring the pH value, by using digital pH meter (WTW Multi 340i). Total volatile basic nitrogen (TVB-N, mg/100 g) values were determined according to the method described by Ludorff & Meyer (1973). Water activity were measured with Novasina LabSwift-Aw meter.

2.4 Microbiological analysis

For all microbiological analyzes, 10 g of sample was aseptically weighed and transferred into 90 mL of sterile buffered peptone water (MERCK 1.07228.0500) and homogenized by Stomacher (MAYO HG400) for 3 min. And then, the decimal dilutions were prepared with 0.85% NaCl. Total Aerobic Mesophilic Bacteria (TMAB) and Psychotropic bacteria (TPAB) were determined by using surface cultivation method and Plate Count Agar (PCA, Merck 105463.0500) was used as a medium. Plates were incubated at 10 °C for 7 days for TPAB and for 2 days at 37 °C for TMAB. Total yeast and mould count was enumerated on Potato Dextrose Agar (PDA, Merck 1.10130) at 22-25 °C for 4-5days. Coliform bacteria were incubated for 18-24 hour at 35-37 °C in Violet Red Bile Agar (VRBA, Merck 1.01406) (Halkman, 2005). Total Lactic acid bacteria (LAB) were incubated on the De Man, Rogosa and Sharpe Agar (MRS, Merck code: 1.10660.0500) at 37 °C for 3 days (Kostaki et al., 2009; Stamatis & Arkoudelos, 2007). Microbiological datas were transformed into logarithms of the number of colony forming units (CFU/g).

2.5 Statistical analysis

Statistical evaluations of physicochemical and microbiological analyses were made using MINITAB 16 by one-way analysis of variance (ANOVA) using a significance level of $p < 0.05$ (%95) by Tukey's test.

3 Results and discussion

The initial pH (Table 1), A_w (Table 2) and TVB-N (Table 3) values of the fish fillets were identified as 6.42, 0.961 and 15.59 mg/100 g, respectively. The pH, Aw and TVB-N values of the marinades kept in the acid + salt brine were reduced to 3.98, 0.915 and 3.25 mg/100 g, respectively ($p < 0.05$). The pH values of all the groups did not exceed 4.38 during the six months of storage. Topuz et al. (2014) and Šimat et al. (2011) reported that the initial pH values of the anchovies were 6.49 and 6.16, which were reduced respectively to 4.18 and 5.77 after marinating. The pH value required for marinating is between 4 and 4.5. In this pH value, tissue cathepsins become more active. Consequently,

Table 1. Changes in pH value of marinated anchovy fillet samples sauced with sun flower seed oil and different essential oils during refrigerated storage at 4 ± 2 °C.

		A	*B*	*C*	*D*	*E*
Raw anchovy		6.42 ± 0.00^{Aa}	6.42 ± 0.00^{Aa}	6.42 ± 0.00^{Aa}	6.42 ± 0.00^{Aa}	6.42 ± 0.00^{Aa}
Marine anchovy		3.98 ± 0.08^{Abc}	3.98 ± 0.08^{Ade}	3.98 ± 0.08^{Ad}	3.98 ± 0.08^{Ad}	3.98 ± 0.08^{Ac}
Storage time (months)	*1*	3.83 ± 0.01^{Cc}	4.15 ± 0.01^{Bcd}	4.21 ± 0.01^{Bc}	4.22 ± 0.03^{Bbc}	4.39 ± 0.01^{Ab}
	2	4.00 ± 0.05^{Bbc}	4.19 ± 0.01^{ABbc}	4.38 ± 0.04^{Ab}	4.16 ± 0.06^{ABcd}	4.11 ± 0.09^{Bc}
	3	3.88 ± 0.03^{Cc}	4.05 ± 0.03^{ABcde}	4.17 ± 0.02^{Ac}	4.09 ± 0.05^{ABcd}	3.97 ± 0.03^{BCc}
	4	4.15 ± 0.06^{Bb}	4.33 ± 0.01^{Ab}	4.37 ± 0.03^{Ab}	4.41 ± 0.04^{Ab}	4.38 ± 0.02^{Ab}
	5	4.01 ± 0.03^{BCbc}	3.97 ± 0.04^{Ce}	4.19 ± 0.02^{Ac}	4.16 ± 0.01^{Acd}	4.11 ± 0.01^{ABc}
	6	4.09 ± 0.04^{ABb}	4.04 ± 0.04^{Bcde}	4.22 ± 0.01^{Ac}	4.06 ± 0.04^{Bcd}	4.08 ± 0.03^{ABc}

A = Control group; B = Marinated anchovy with rosemary oil; C = Marinated anchovy with coriander oil; D = Marinated anchovy with laurel oil; E = Marinated anchovy with garlic oil; ↓ (a, b….) Means with different lowercase letters in the same column are significantly different ($p < 0.05$); → (A, B...) Means with different capital letters in the same row are significantly different ($p < 0.05$).

Table 2. Changes in A_w value of marinated anchovy fillet samples sauced with sun flower oil and different essential oils during refrigerated storage at +4 ± 2 °C.

		A	*B*	*C*	*D*	*E*
Raw anchovy		0.961 ± 0.001^{Aa}	0.961 ± 0.001^{Aa}	0.961 ± 0.001^{Aa}	0.961 ± 0.001^{Aa}	0.961 ± 0.001^{Aa}
Marine anchovy		0.915 ± 0.001^{Ae}	0.915 ± 0.001^{Ae}	0.915 ± 0.001^{Aef}	0.915 ± 0.001^{Acd}	0.915 ± 0.001^{Ad}
Storage time (months)	*1*	0.916 ± 0.001^{Ae}	0.896 ± 0.002^{Cef}	0.917 ± 0.002^{Aef}	0.909 ± 0.003^{ABd}	0.905 ± 0.001^{Be}
	2	0.924 ± 0.002^{Ad}	0.924 ± 0.005^{Acde}	0.925 ± 0.002^{Ade}	0.920 ± 0.001^{Ac}	0.867 ± 0.002^{Bef}
	3	0.932 ± 0.001^{Ac}	0.928 ± 0.001^{ABbcd}	0.932 ± 0.001^{ABbc}	0.928 ± 0.001^{Bb}	0.932 ± 0.001^{ABb}
	4	0.932 ± 0.001^{ABc}	0.931 ± 0.001^{ABbc}	0.927 ± 0.001^{Bcd}	0.933 ± 0.001^{Ab}	0.935 ±0.001^{Ab}
	5	0.938 ± 0.001^{Ab}	0.936 ± 0.001^{Ab}	0.935 ± 0.002^{Ab}	0.929 ± 0.001^{Bb}	0.933 ± 0.001^{ABb}
	6	0.923 ± 0.001^{ABd}	0.921 ±0.001^{ABCde}	0.918 ± 0.001^{Cef}	0.919 ± 0.001^{BCc}	0.924 ± 0.001^{Ac}

A = Control group; B = Marinated anchovy with rosemary oil; C = Marinated anchovy with coriander oil; D = Marinated anchovy with laurel oil; E = Marinated anchovy with garlic oil; ↓ (a, b….) Means with different lowercase letters in the same column are significantly different ($p < 0.05$); → (A, B...) Means with different capital letters in the same row are significantly different ($p < 0.05$).

Table 3. Changes in TVB-N value of marinated anchovy fillet samples sauced with sunflower oil and different essential oils during refrigerated storage at 4 ± 2 °C.

		A	*B*	*C*	*D*	*E*
Raw anchovy		15.59 ± 0.27^{Ab}	15.59 ± 0.27^{Ab}	15.59 ± 0.27^{Ab}	15.59 ± 0.27^{Ab}	15.59 ± 0.27^{Aa}
Marine anchovy		3.25 ± 0.21^{Ae}	3.25 ± 0.21^{Ae}	3.25 ± 0.21^{Ad}	3.25 ± 0.21^{Ae}	3.25 ± 0.21^{Ac}
Storage time (months)	*1*	14.41 ± 0.61^{Abc}	13.20 ± 0.20^{ABc}	14.62 ± 0.37^{Ab}	13.67 ± 0.36^{ABbc}	12.14 ± 0.7^{Bcd}
	2	12.58 ± 0.64^{Ac}	11.96 ± 0.80^{Acd}	13.88 ± 0.68^{Abc}	12.58 ± 0.18^{Ac}	11.77 ± 0.36^{Acd}
	3	8.84 ± 0.28^{Cd}	10.07 ± 0.29B^{Cd}	12.14 ± 0.26^{Ac}	10.48 ± 0.20^{Bd}	9.89 ± 0.44B^{Cd}
	4	14.45 ± 0.63^{Abc}	11.73 ± 0.32^{Bcd}	13.97 ± 0.57^{ABbc}	15.61 ± 0.65^{Ab}	11.81 ± 0.42^{Bcd}
	5	24.88 ± 054^{Aa}	19.15 ± 0.36^{Ba}	19.43 ± 0.29^{Ba}	18.24 ± 0.71^{Ba}	12.44 ± 0.72^{Cbc}
	6	24.10 ± 0.19^{Aa}	20.11 ± 0.66^{Ba}	20.13 ± 0.44^{Ba}	18.07 ± 0.45^{Ca}	14.67 ± 0.43^{Dab}

A = Control group; B = Marinated anchovy with rosemary oil; C = Marinated anchovy with coriander oil; D = Marinated anchovy with laurel oil; E = Marinated anchovy with garlic oil; ↓ (a, b….) Means with different lowercase letters in the same column are significantly different ($p < 0.05$); → (A, B...) Means with different capital letters in the same row are significantly different ($p < 0.05$).

muscle proteins are broken down into peptides and amino acids, which results in the development of the taste and flavour that are peculiar to the marinade (Gökoğlu, 2002). As a result of the measurements made at the end of the 1st, 2nd, 3rd and 4th months, it was established that the differences between the pH values of the marinades with the added rosemary, coriander and laurel essential oils were not significant ($p > 0.05$). As is the case with our study, Gökoğlu et al. (2009) reported that there was not any significant change in the pH values of anchovy marinades involving concentrated pomegranate juice throughout their storage in the refrigerator.

Salt is used primarily for reducing the A_w value in addition to adding flavour. Salting is used because most bacteria, fungi and other potentially pathogenic organisms cannot survive in high salt conditions due to the osmotic pressure that salt creates. Any living cell in an environment with high concentrations of salt will become dehydrated through osmosis, and die or become inactivated. This extraction of water from the fish meat caused a decrease in water activity (Sampels, 2015). Capaccioni et al. (2011) reported that the A_w values of anchovy (*Engraulis anchoita*) fillets and marinated anchovies were 0.99 and 0.94, respectively. Similar results were found in this research. The A_w values of all the groups were under the A_w value of the fresh fish fillets throughout the storage period (Table 2). The same results were found by Šimat et al. (2011) in the case of marinated anchovies.

One of the main chemical analyses used in determining the quality deterioration of fish meat is TVB-N. The TVB-N value of the fish fillets before the marination was approximately five times the value obtained after marination (Table 3). Similarly, we found, in another study (Kılınç & Cakli, 2004), that the TVB-N value of sardines was reduced after the marination process following the thawing of the frozen fish. The reason for this decline was the dissolution of the TVB-N components by the brine that contains acid and salt (Kılınç & Cakli, 2004).

The acid added to the brine neutralized the nitrogenous components found in the fish meat and, as a result, the pH values tended more towards the acidic, while the TVB-N development in all the groups slowed down throughout the storage period. In particular, by the end of the storage period, the TVB-N value of group E - containing garlic essential oil - was found to be the lowest (14.67 mgN/100 g) among the groups ($p < 0.05$). The TVB-N value of the control group was 24.88 mg/100 g in the fifth month, and this did not change statistically in the sixth month ($p > 0.05$). No significant changes were found by the end of the storage period in the TVB-N values of the groups treated with rosemary and coriander essential oils ($p > 0.05$). A level of 30 mg/100 g was considered the maximum limit above which fishery products are considered unfit for human consumption (Sikorski et al., 1989). The TVB-N values were noted throughout the storage period, and the essential oils were found to have an

effect on the TVB-N. A similar increase in TVB-N values has been reported in the case of marinated sardines (Özden & Baygar, 2003; Kılınç & Cakli, 2005), brined anchovies (Karaçam et al., 2002); marinated horse mackerel (Erdem & Bilgin, 2005) and marinated vacuum-packed Pacific saury (Sallam et al., 2007).

The microbiological analysis data are given in Tables 4-6. The total aerobic mesophilic bacteria (TMAB), psychotropic bacteria (TPAB) and total yeast-mould counts of the raw material were found to be 2.84, 1.88 and 3.62LogCFU/g, respectively. After marinating, all these microorganisms were inhibited. The same results were determined by the other studies involving marinated sardines (Kılınç & Cakli, 2004); marinated anchovies (Günşen et al., 2011); trout fillets in grape vinegar and apple cider vinegar (Kılınç & Yavuz, 2011); marinated crayfish with a rosemary and thyme essential oils additive (Duman et al., 2012) and marinated Pacific saury (Sallam et al., 2007). Calo et al.

Table 4. Total mesophilic bacteria counts of marinated anchovy fillet samples sauced with sunflower seed oil and different essential oils during refrigerated storage at 4 ± 2 °C.

		A	*B*	*C*	*D*	*E*
Raw anchovy		2.84 ± 0.02^{Abcd}	2.84 ± 0.02^{Ab}	2.84 ± 0.02^{Acd}	2.84 ± 0.02^{Ab}	2.84 ± 0.02^{Aab}
After 5% salt marination		2.56 ± 0.04^{Ad}	2.56 ± 0.04^{Ab}	2.56 ± 0.04^{Ad}	2.56 ± 0.04^{Ab}	2.56 ± 0.04^{Ab}
Marine anchovy		1.38 ± 0.16^{Ae}	1.38 ± 0.16^{Ac}	1.38 ± 0.16^{Ae}	1.38 ± 0.16^{Ad}	1.38 ± 0.16^{Ac}
Storage time (months)	*1*	2.61 ± 0.03^{Ad}	1.34 ± 0.33^{Bc}	0.96 ± 0.00^{Bf}	0.96 ± 0.00^{Be}	1.18 ± 0.12^{Bc}
	2	2.68 ± 0.04^{Acd}	0.96 ± 0.00^{Cc}	0.96 ± 0.00^{Cf}	2.10 ± 0.07^{Bc}	0.96 ± 0.00^{Cc}
	3	2.96 ± 0.01^{Abc}	2.71 ± 0.04^{ABb}	2.52 ± 0.04^{Bd}	2.87 ± 0.00^{Ab}	1.30 ± 0.14^{Cc}
	4	3.13 ± 0.02^{Ab}	2.56 ± 0.02^{Cb}	2.89 ± 0.01^{Bc}	2.87 ± 0.04^{Bb}	2.59 ± 0.02^{Cb}
	5	4.51 ± 0.01^{Aa}	3.12 ± 0.04^{Db}	4.25 ± 0.02^{Ba}	3.38 ± 0.05^{Ca}	3.22 ± 0.03^{CDa}
	6	4.65 ± 0.02^{Aa}	4.11 ± 0.05^{Ba}	3.78 ±0.01^{Cb}	3.46 ± 0.01^{Da}	3.26 ± 0.04^{Ea}

A = Control group; B = Marinated anchovy with rosemary oil; C = Marinated anchovy with coriander oil; D = Marinated anchovy with laurel oil; E = Marinated anchovy with garlic oil; ↓ (a, b….) Means with different lowercase letters in the same column are significantly different ($p < 0.05$); → (A, B...) Means with different capital letters in the same row are significantly different ($p < 0.05$).

Table 5. Total physcrotrophic bacteria counts of marinated anchovy fillet samples sauced with sunflower seed oil and different essential oils during refrigerated storage at 4 ± 2 °C.

		A	*B*	*C*	*D*	*E*
Raw anchovy		1.88 ± 0.08^{Abc}	1.88 ± 0.08^{Aa}	1.88 ± 0.08^{Aab}	1.88 ± 0.08^{Aa}b	1.88 ± 0.08^{Aab}
After 5% salt marination		0.96 ± 0.00^{Ad}	0.96 ± 0.00^{Ab}	0.96 ± 0.00^{Ad}	0.96 ± 0.00^{Ac}	0.96 ± 0.00^{Ac}
Marine anchovy		0.96 ± 0.00^{Ad}	0.96 ± 0.00^{Ab}	0.96 ± 0.00^{Ad}	0.96 ± 0.00^{Ac}	0.96 ± 0.00^{Ac}
Storage time (months)	*1*	2.11 ± 0.08^{Aab}	1.46 ± 0.25^{Aab}	1.96 ± 0.00^{Aa}	1.21 ± 0.22^{Abc}	1.21 ± 0.22^{Ac}
	2	0.96 ± 0.00^{Ad}	0.96 ± 0.00^{Ab}	0.96 ± 0.00^{Ad}	0.96 ± 0.00^{Ac}	0.96 ± 0.00^{Ac}
	3	1.56 ± 0.00^{Ac}	0.96 ± 0.00^{Cb}	1.47 ± 0.03^{ABbc}	1.27 ± 0.10^{Bbc}	1.35 ± 0.04^{ABbc}
	4	1.11 ± 0.08^{Ad}	1.03 ± 0.07^{Ab}	0.96 ± 0.00^{Ad}	1.03 ± 0.06^{Ac}	0.96 ± 0.00^{Ac}
	5	1.96 ± 0.00^{Aab}	1.21 ± 0.22^{ABab}	0.96 ± 0.00^{Bd}	1.90 ± 0.29^{ABab}	1.46 ± 0.25^{ABabc}
	6	2.28 ± 0.16^{Aa}	1.46 ± 0.25^{ABab}	1.21 ± 0.22^{Bcd}	2.18 ± 0.07^{Aa}	2.03 ± 0.07^{ABa}

A = Control group; B = Marinated anchovy with rosemary oil; C = Marinated anchovy with coriander oil; D = Marinated anchovy with laurel oil; E = Marinated anchovy with garlic oil; ↓ (a, b….) Means with different lowercase letters in the same column are significantly different ($p < 0.05$); → (A, B...) Means with different capital letters in the same row are significantly different ($p < 0.05$).

Table 6. Total yeast and mould counts of marinated anchovy fillet samples sauced with sun flower seed oil and different essential oils during refrigerated storage at +4 ± 2 °C.

		A	*B*	*C*	*D*	*E*
Raw anchovy		3.62 ± 0.06^{Aa}	3.62 ± 0.06^{Aa}	3.62 ± 0.06^{Aa}	3.62 ± 0.06^{Aa}	3.62 ± 0.06^{Aa}
After 5% salt marination		3.44 ± 0.02^{Aa}	3.44 ± 0.02^{Aa}	3.44 ± 0.02^{Aa}	3.44A ± 0.02^{Aa}	3.44 ± 0.02^{Aa}
Marine anchovy		0.75 ± 0.11^{Ae}	0.75 ± 0.11^{Ac}	0.75 ± 0.11^{Af}	1.48 ± 0.07^{Ad}	0.75 ± 0.11^{Ad}
Storage time (months)	*1*	1.48 ± 0.07^{Ac}	1.20 ± 0.12^{Abc}	1.23 ± 0.09^{Ade}	1.28 ± 0.12^{Ac}	1.42 ± 0.05^{Ac}
	2	1.11 ± 0.08^{Bde}	1.47 ± 0.03^{Ab}	1.52 ± 0.05^{Acd}	1.38 ± 0.06^{Ac}	1.53 ± 0.03^{Ac}
	3	1.38 ± 0.06^{Bcd}	1.61 ± 0.02^{ABb}	1.52 ± 0.05^{ABcd}	1.38 ± 0.06^{Bc}	1.63 ± 0.04^{Ac}
	4	1.48 ± 0.07^{Ac}	1.44 ± 0.15^{Ab}	1.11 ± 0.08^{Aef}	1.18 ± 0.07^{Ac}	1.41 ± 0.08^{Ac}
	5	1.93 ± 0.04^{Ab}	1.60 ± 0.06^{Bb}	1.89 ± 0.07^{Abc}	2.05 ± 0.03^{Ab}	2.07 ± 0.08^{Ab}
	6	2.06 ± 0.03^{Ab}	1.65 ± 0.05^{Bb}	1.97 ± 0.06^{Ab}	2.12 ± 0.04^{Ab}	2.09 ± 0.07^{Ab}

A = Control group; B = Marinated anchovy with rosemary oil; C = Marinated anchovy with coriander oil; D = Marinated anchovy with laurel oil; E = Marinated anchovy with garlic oil; ↓ (a, b….) Means with different lowercase letters in the same column are significantly different ($p < 0.05$); → (A, B...) Means with different capital letters in the same row are significantly different ($p < 0.05$).

(2015) noted that aromatic plants and their components had been examined as potential inhibitors of bacterial growth, and most of their properties had been linked to essential oils and other secondary plant metabolites.

The TMAB count of all the groups did not exceed the limit value (10^6) during the storage period. At the end of the storage period, the minimum TMAB load was identified in the group that contained garlic essential oil (3.26LogCFU/g) ($p < 0.05$). The highest and lowest TPAB values during the storage period were found to be 2.28LogCFU/g in the control group, and 1.21LogCFU/g in the group which contains coriander essential oil. The total yeast and mould values of all the groups did not exceed 10^3 throughout the storage period. At the end of the storage period, the total yeast and mould values of all groups were similar ($p > 0.05$) with the exception of group B (rosemary essential oil added group) which was lowest in terms of total yeast and mould values. In the marinade, lower pH results in retarding bacterial growth (Sampels, 2015). Coriander essential oils and extracts possess promising antibacterial, antifungal and antioxidative properties as various chemical components are contained in different parts of the plant. This therefore plays an important role in maintaining the shelf life of foods by preventing their spoilage (Mandal & Mandal, 2015).

No coliform and lactic acid bacteria were found and identified throughout the storage period, due to the marination process. Similarly, it was reported that no *E. coli* and Lactobacillus spp. were identified in the marinated anchovies (*E. encrasicholus*) that were packaged using different methods (Günşen et al., 2011), and no Coliform bacteria was identified in cold marinated anchovies (*E. anchoita*) (Fuselli et al., 1994).

4 Conclusion

Based on the results of this study, it can be said that, during six months of storage in the refrigerator, the number of all microorganisms tested were below the limit values and that the essential oils have a positive effect on microorganism inhibition. Moreover, the group with the garlic essential oil is a lot more satisfactory than the other groups in terms of the results obtained.

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