The effect of black seed oil and olive oil on shelf life of dry-salted crucian carp (*Carassius carassius* Linneaus, 1758)

Çağlak E.*; Karslı B.

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Department of Processing Technology, Faculty of Fisheries, Recep Tayyip Erdogan University, 53100 Rize, Turkey
*Corresponding Author's Email: emre.caglak@erdogan.edu.tr

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Introduction

Seafood important in the health of human beings, is qualified as sensitive nutrients that have a limited shelf life (Gram and Huss, 2000). Due to their sensitivities, varieties of preservation methods have been used from past to present. One of these methods is salting that has been used traditionally. The salting process is divided into 3 groups including dry salting, brine salting and mixed salting (Erdem *et al*., 2005). With developing technology, the rate of salt is being decreased in salting process. In addition to this, different methods (chemical, herbal, etc.) are being used by the addition of several preservatives. One of these preservatives is olive oil which has the effect of an antioxidant on the product. Besides, it also contains a big amount of tocopherol which is well known and common antioxidants (Burt, 2004; Kykkidou *et al*., 2009). It is well known that black seed and its oil has an antimicrobial, antifungal and antihelmintic effects (Salem, 2005). Crucian carp is a freshwater fish. Global production of farmed crucian carp had reached 2 595 735 tonnes in 2013 (FAO, 2016). Turkey has been exporting it to the Middle Eastern countries in recent years. Currently, there has been no study on processing of crucian carp. Also, the effects of olive oil and black seed oil on shelf life and quality criteria of dry-salted seafoods have not been investigated.

The present study was conducted to show that crucian carp was salted with the dry salting method at the rate of 1:3 (salt:fish) and during this process, it was also aimed to determine the effects of olive oil and black seed oil (*Nigella sativa*) on the quality criterias of dry-salted crucian carp, to determine the product's shelf-life by biochemical, chemical, sensorial, microbiological analysis, to show the practicability of different processing methods.

Materials and methods

A total of 24 kg of fresh crucian carp (*Carassius carassius)* were purchased from a local fisherman. The fish were caught from Manyas Lake, Balıkesir, Turkey. The fish were transported to the Processing Technology Laboratory of Recep Tayyip Erdogan University Faculty of Fisheries in styrofoam boxes containing ice. The average length and weight of the sampled fish were 24.7±3.85 cm and 323±12.25 g, respectively. The fish were be-headed, and gutted and their fins were removed. Scales were cleaned out and washed to clean out the blood and mucus. Then, the fish were filleted and separated into 3 groups as first group (control group-only salted), second group (salted with black seed oil) and third group (salted with olive oil). The products of the second and third groups were kept in black seed oil and olive oil for one minute and then all the groups were taken into the process of dry salting method at the rate of 1:3 (salt:fish). In this process, the fish were put into glass jars as a layer of fish and a layer of salt, and they were stored at room temperature (20±2 ºC). The biochemical, chemical, microbiological and sensorial analyzes of the product groups were performed for 15 days in the first month and once a month in the following terms.

Biochemical analysis

The moisture, crude ash and crude protein contents of crucian carp were determined according to Norwitz (1970). The crude fat value was detected using the soxhlet methods (Bligh and Dyer, 1959).

Chemical analysis

The pH values of fish were measured according to Curran *et al*. (1980) by a pH meter (Hanna, HI 3220). Total volatile basic nitrogen (TVB-N) was determined according to the Lücke-Geidel method (Varlık *et al*., 1993). Thiobarbituric acid (TBA) was determined using the method by Tarladgis *et al*. (1960).

Microbiological analysis

Twenty-five grams of fish meat were mixed with 225 ml of 0.85% physiological saline solution (PSS). All serial dilutions were prepared in 0.85% PSS. Total viable counts of mesophilic and psychrophilic microorganisms were obtained using plate count agar incubated at 37 °C for 48 h and at 6.5 °C for 10 days, respectively. Violet red bile agar was used for total coliform: plates were incubated at 37 °C for 48 h (Halkman, 2005). Yeast-mold counts were taken after the incubation on potato dextrose agar at 30 °C for 3-5 days (Harrigan and McCance, 1976).

Sensory analysis

Sensory evaluations were conducted by six experienced panelists, according to the method described by Altuğ and Elmacı (2005). The samples were assessed on odor, taste, appearance, and texture characteristics using a 10-point descriptive scale. A score of 8–10 indicated “very good” quality, a score of 6.0–7.9 was considered as “good” quality, a score of 5.9–4.1 met “the limit of acceptability”, while a score of 1–4 was regarded as “spoiled, unacceptable”.

Statistical analysis

The data obtained were analyzed by analysis of variance (ANOVA), and when significant differences were found, comparisons among means were carried out by using a Tukey test (p<0.05) under the program JMP 5.0.1 (SAS Institute Inc., Cary, NC, USA; Sümbüloğlu and Sümbüloğlu, 2000).

Results and discussion

The moisture content of fresh samples was determined as 78.02%. With the effect of the salting process, all of the groups had a loss of water on the 15th day of the storage (Table 1). Significant differences were found between moisture amount of fresh samples and the salted groups (p<0.05). Similarly, a recent study showed that the amount of moisture of pearl mullet decreased from 73.45% to 40-49.92% by the effect of salting (Kılınççeker and Küçüköner, 2003).

The content of the crude ash in the fresh fish was determined as 1.54% (Table 1). After the salting processes, the content of the crude ash increased significantly, depending on the salting process (p<0.05). The changes among the groups were found no significant except on the 30th and 150th days of storage (p>0.05).

Table 1: Changes of biochemical contents (%) of dry-salted crucian carp with different additives.

Days	Groups	Moisture	Crude Ash	Crude Protein	Crude Fat	Salt
Fresh	C	$78.02\pm0.94_{A}$	$1.54\pm0.10_{A}$	$17.16\pm1.92_{ABC}$	$2.64\pm0.01_{AE}$	$0.20\pm0.01_{A}$
	C	$58.10\pm0.12^{a}_{B}$	$19.95\pm0.58^{a}_{B}$	$15.21\pm0.27^{a}_{A}$	$3.89\pm0.11^{a}_{B}$	$11.40\pm0.06^{c}_{B}$
15	BSO	$55.72\pm0.59^{b}_{B}$	$18.73\pm0.34^{a}_{C}$	$18.75\pm0.40^{a}_{A}$	$4.08\pm0.02^{a}_{B}$	$11.60\pm0.03^{b}_{B}$
	OO	$55.65\pm0.32^{b}_{BC}$	$19.30\pm0.33^{a}_{B}$	$17.47\pm2.27^{a}_{A}$	$5.78\pm0.01^{b}_{B}$	$12.40\pm0.01^{a}_{B}$
	C	$58.04\pm0.16^{a}_{B}$	$21.06\pm0.16^{a}_{B}$	$16.88\pm0.42^{a}_{A}$	$3.38\pm0.05^{a}_{C}$	$13.80\pm0.01^{a}_{C}$
30	BSO	$56.40\pm0.29^{b}_{B}$	$19.99\pm0.09^{b}_{BC}$	$15.49\pm0.18^{a}_{C}$	$5.51\pm0.02^{b}_{C}$	$13.10\pm0.00^{b}_{C}$
	OO	$55.36\pm0.39^{b}_{C}$	$19.91\pm0.34^{b}_{B}$	$16.42\pm0.84^{a}_{A}$	$5.90\pm0.08^{c}_{B}$	$10.70\pm0.01^{c}_{C}$
	C	$56.58\pm0.22^{a}_{B}$	$20.56\pm0.74^{a}_{B}$	$16.85\pm0.25^{b}_{A}$	$3.72\pm0.13^{a}_{B}$	$13.20\pm0.03^{c}_{D}$
60	BSO	$56.75\pm0.93^{a}_{B}$	$20.31\pm0.20^{a}_{BC}$	$18.44\pm0.14^{a}_{AB}$	$4.01\pm0.06^{a}_{B}$	$13.80\pm0.01^{b}_{D}$
	OO	$57.61\pm0.01^{a}_{BC}$	$20.71\pm0.10^{a}_{B}$	$17.40\pm0.27^{b}_{A}$	$4.58\pm0.07^{b}_{C}$	$13.90\pm0.00^{a}_{D}$
	C	$57.08\pm0.13^{a}_{B}$	$19.26\pm1.52^{a}_{B}$	$17.40\pm0.39^{a}_{A}$	$2.20\pm0.03^{a}_{D}$	$13.00\pm0.00^{b}_{E}$
90	BSO	$56.53\pm0.75^{a}_{B}$	$21.98\pm1.19^{a}_{B}$	$16.24\pm0.53^{a}_{ABC}$	$5.82\pm0.23^{b}_{C}$	$13.00\pm0.14^{b}_{C}$
	OO	$56.30\pm1.18^{a}_{BC}$	$21.80\pm1.79^{a}_{B}$	$16.09\pm0.32^{a}_{A}$	$4.50\pm0.02^{c}_{C}$	$13.90\pm0.03^{a}_{D}$
	C	$57.70\pm0.72^{a}_{B}$	$20.38\pm0.56^{a}_{B}$	$17.29\pm1.40^{a}_{A}$	$2.84\pm0.05^{a}_{AE}$	$14.70\pm0.01^{a}_{F}$
120	BSO	$57.70\pm1.95^{a}_{B}$	$20.27\pm0.79^{a}_{BC}$	$15.79\pm0.04^{a}_{BC}$	$4.95\pm0.01^{b}_{D}$	$11.80\pm0.04^{b}_{B}$
	OO	$57.94\pm0.65^{a}_{B}$	$20.23\pm0.75^{a}_{B}$	$15.51\pm0.63^{a}_{A}$	$4.68\pm0.01^{c}_{C}$	$12.70\pm0.00^{ab}_{E}$
	C	$58.35\pm0.93^{a}_{B}$	$19.91\pm0.16^{a}_{B}$	$17.34\pm1.46^{a}_{A}$	$2.42\pm0.02^{a}_{DE}$	$14.20\pm0.00^{b}_{G}$
150	BSO	$54.42\pm0.01^{b}_{B}$	$18.76\pm0.07^{b}_{C}$	$17.17\pm0.01^{a}_{ABC}$	$5.83\pm0.05^{b}_{C}$	$13.40\pm0.07^{c}_{E}$
	OO	$58.02\pm0.16^{a}_{B}$	$20.85\pm0.11^{c}_{B}$	$16.53\pm0.30^{a}_{A}$	$4.63\pm0.06^{c}_{C}$	$14.50\pm0.03^{a}_{F}$
	C	$57.85\pm0.01^{a}_{B}$	$21.19\pm0.09^{a}_{B}$	$16.40\pm1.12^{a}_{A}$	$2.24\pm0.03^{a}_{D}$	$14.10\pm0.03^{b}_{G}$
180	BSO	$56.57\pm0.11^{b}_{B}$	$21.08\pm0.11^{a}_{B}$	$17.50\pm0.19^{a}_{ABC}$	$4.48\pm0.03^{b}_{F}$	$14.70\pm0.00^{a}_{F}$
	OO	$56.24\pm0.51^{b}_{BC}$	$21.05\pm0.13^{a}_{B}$	$16.40\pm 0.15^{a}_{A}$	$4.68\pm0.07^{c}_{C}$	$14.70\pm0.03^{a}_{G}$

The different subscript capital letters (A, B, C, D, E, F, G) in the same column represent statistical differences detected within the same group in different storage period ($p<0.05$). The different small subscript letters (a, b, c) in the same row represent statistical differences detected among groups in the same storage day (p<0.05). Values are mean + standard deviation. **C:** Control group, **BSO:** Black seed oil group, **OO:** Olive oil group.

Bilgin *et al*. (2007) stated that the amount of crude ash of *Salmo trutta macrostigma* increased from 1.33% in fresh to 11.20% in the dry salted samples and 5.90% in brine samples.

During the storage period, there were not any significant changes between protein contents of the fresh and the other groups ($p>0.05$). The crude protein content of the salted fillets during the storage was found between 15.21-18.75%. Kılınççeker and Küçüköner (2003) determined that the amount of crude protein (19.20%) of fresh pearl mullet increased up to 19.23% after dry salting (E method), but no significant changes were detected.

After the dry salting process, crude fat value increased to 3.89 of C, 4.08 of BSO and 5.78 of OO on the 15th day (Table 1). During the storage period, irregular increases and decreases were determined in all groups. The changes in crude fat were found significant among the salted groups with fresh products in all the storage days (except for on the 120th and 150th day of the control group) ($p<0.05$). The crude fat value of control group was found less than the value of fresh products at the end of the storage period and it showed a parallelism with the other studies (Bilgin *et al*., 2007; Ahmed *et al*., 2010). But, the fat values of the other groups increased due to additives added.

The salt content of fresh crucian carp was found as 0.20 % (Table 1). At the end of the storage period, the salt amount was calculated as 14.10% (C), 14.70% (BSO) and 14.70% (OO). In addition, the control group was found to be different from the other groups ($p<0.05$). During the storage period, differences were observed among the groups ($p<0.05$). Data of the present study was found similar with results of studies applied the different salting methods in literature (Kılınççeker and Küçüköner, 2003; Turan *et al*., 2007).

The pH value of raw crucian carp was determined as 7.26. With the results of the applied processes, a decrease was seen in all groups (Fig. 1a). At the end of the storage period, the statistical differences among groups were found no significant ($p>0.05$). Roth *et al*. (2005) stated that the average pH value of Atlantic salmon smolt was 7.27. Şentürk (1994) also stated that the pH value of the fresh shrimp sample was 7.2. It could be observed that there is a different pH value in different species of fisheries. Many literary studies state that, after the death of fish, changes in pH value occur due to the influence of some factors such as the glycogen level of the muscles and fishing methods (Varlik *et al*., 1993; Tzikas *et al*., 2007).

The amount of TVB-N increased during the storage period. TVB-N amount of the control group only exceeded the consumable value limit on the 150th day. At the end of the storage period, minimum increase occurred in black seed oiled products while maximum increase occurred in the control group (Fig. 1b). The changes among the storage days were found statistically significant ($p<0.05$). Similarly, Süle (2011) determined that the TVB-N value (14.95 mg/100g of raw) of surimi *Carassius gibelio* was increased during the storage period and it was determined as 27.97 m/100g on the 90th

day of storage.

The TBA value increased in the all groups during the storage and exceeded the consumable limit value on the 150th day in the control group, and on the 180th day in the black seed oiled and the olive oiled groups (Fig. 1c). Differences among TBA values of the groups at the end of the storage period (p<0.05) were found significant. İnanlı and Patır (2004) detected the TBA value of sorbated and salted trout fillets increased during the storage period, however none of the groups exceeded values of consumable limit during the 84 days of storage. When comparing the results of this research with the data of the 90th day of the present study, it was observed that similar results were obtained.

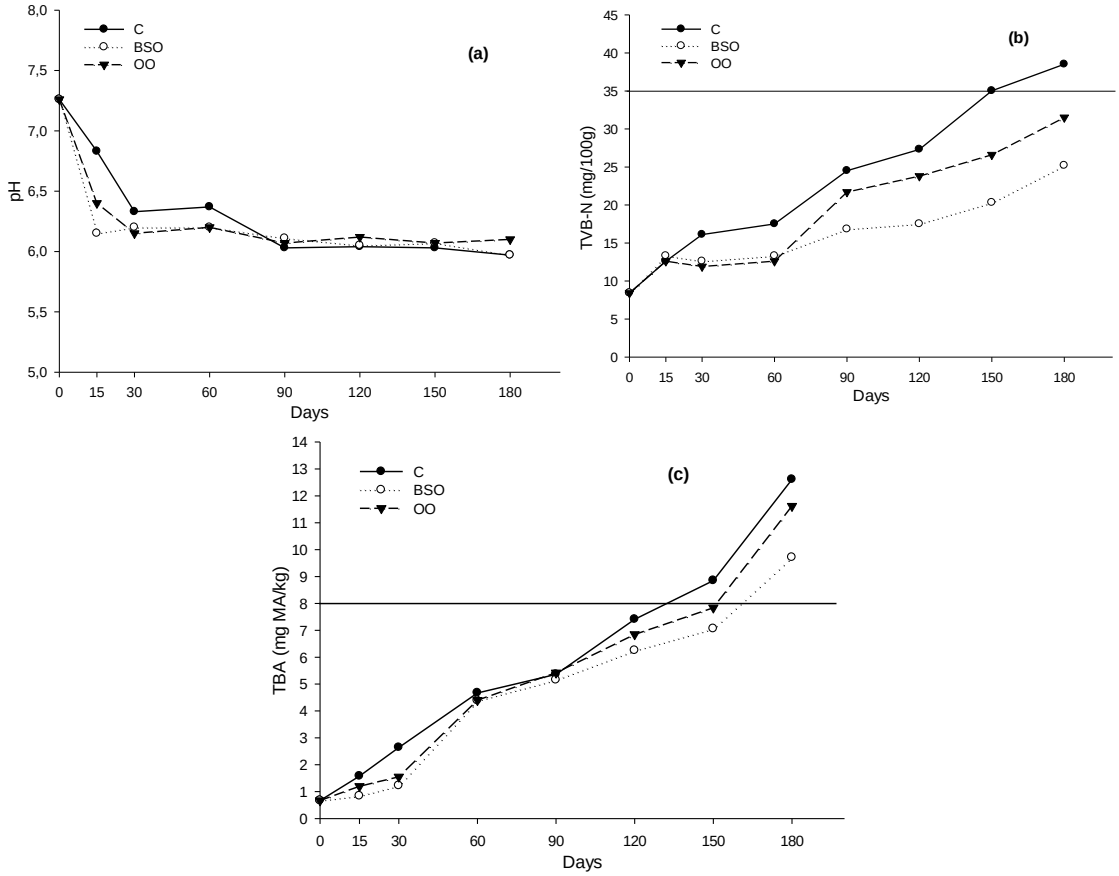

Figure 1: The results of chemical analysis of dry-salted crucian carp with different additives.

The total mesophilic aerobic bacteria (TMAB) count was found as 1.76 log CFU/g in the raw and it decreased in all groups due to the effect of salting. The TMAB count was found below 1.47 log CFU/g in all groups during the storage time except for the 150th day (2.48) and 180th day (2.90) of olive oiled products. Besides, yeast-mold and total coliform counts of fresh and salted fillets found below the level of 1.47 log CFU/g for all of the storage days. It was determined that

all groups remained within the microbiological quality values during the storage period. Similarly, Ahmed *et al*. (2010) observed a decrease in TVC of *Hydrocynus forskalii* salted during the storage period.

The sensory evaluations were considered by panelists during the storage period and there was a decrease in their likes based on the storage process. It was determined that the black seed oiled group was the most liked product while the control group was least liked product. The odour of the black seed oil gave a different aroma to the product and it gained likes of panelists. The control group was found under the limit value (4.0) in terms of sensory liking on the 150th day of storage. It was also determined that olive oiled fillets (3.88) and black seed oiled fillets (3.98) went down below the limit value at the end of a 180-day storage period. The statistical differences among the groups during the storage period was found significant (p<0.05). Kenar *et al*. (2010) stated that the sardines including extracts of rosemary and sage according to the sensory evaluations had a 7 day longer shelf-life than the control group at 3±1 °C. It was thought that the differences in sensory evaluations occurred from the antioxidant and antimicrobial effects of the additives.

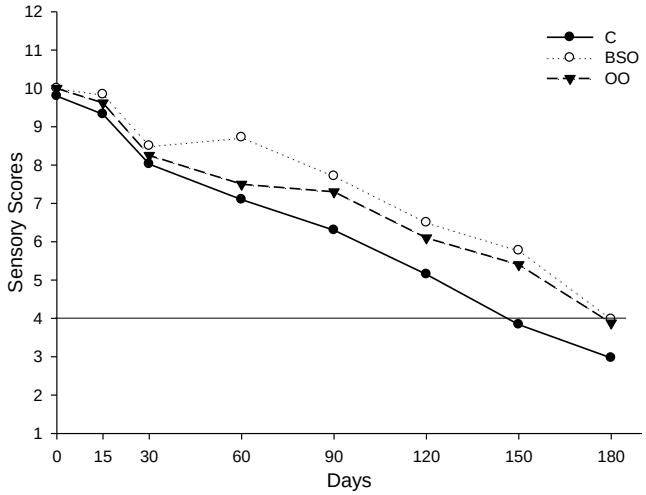

Figure 2: The results of sensory analysis of dry-salted crucian carp with different additives.

In conclusion, microbial growths were limited with the antiseptic and bactericide effects of the salt, and microbial spoilage was not observed in dry-salted crucian carp during the storage period. The usage of the black seed oil and olive oil in the salting process had a more positive effect than the control group in terms of its sensorial and chemical specialities. The black seed oil gave a different aroma and odour to the salted fillets and gained the most likes in terms of sensorial evaluation. Also, it was observed that black seed oil had a positive contribution to the shelf-life of the product. According to the results of sensorial, chemical and microbiological analysis, it was determined that the control group can be preserved for 120 days and

the olive oiled and black seed oiled products could be preserved for 150 days.

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