Food Science and Technology
ISSN 0101-2061 (Print)
ISSN 1678-457X (Online)

DOI: https://doi.org/10.1590/fst.41217

Effect of laurel (*Laurus nobilis*) and curcuma (*Curcuma longa*) on microbiological, chemical and sensory changes in vacuum packed sous-vide european sea bass (*Dicentrarchus labrax*) under chilled conditions

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Abstract

The study was conducted to evaluate the effect of ground laurel and curcuma on the quality changes (microbiological, chemical and sensory) of sous-vide processed gutted and vacuum packaged sea bass under refrigerated temperatures (+3°C). Sea bass samples were gutted and divided into 3 groups namely control (C), laurel added samples (LS) and curcuma added samples (CS) and 30 fish were used for each treatment group. Samples were stored under refrigerated (3 ± 1 °C) conditions for 60 days. Control group was vacuum packed and sous –vide processed and stored under chilled conditions while laurel and curcuma were added as a seasoning for the other groups. During the storage time the counts of total mesophilic aerobic bacteria (TMAB), total psychrophilic aerobic bacteria (TPAB) and Enterobacteriaceae have not reached the maximum allowable number of 7.00 log cfu/g. The highest TVB-N content (30.80 mg N/100g) was found for LS group at the end of the storage time. Regarding the changes in quality indicators the shelf life of the samples was found 45, 49 and 55 days for group C, LS and CS, respectively. The shelf life of the samples was extended 8.88 and 22.2% by addition of ground laurel and curcuma, respectively.

Keywords: sous-vide; seafood; quality; shelf-life; *Dicentrarchus labrax*.

Practical Application: laurel and curcuma was used to extend the shelf life of gutted sous-vied processed sea bass.

1 Introduction

In order to provide the main dietary source as proteins for human beings, seafood has a significant place and considered to be healthy food in the human diet. European sea bass (*Dicentrarchus labrax*) is gaining importance in Mediterranean countries since the acceptance of the sea bass by the consumers is high in terms of its textural properties, flavor, and sensory characteristics. The production of cultured sea bass increased by 100% between 2005 (37.290 tons) and 2014 (74.653 tons) in Turkey (Turkish Statistical Institute, 2016).

Sous vide is one of the effective processing methods for seafood in order to prolong the shelf life and maintain the safety of seafood. The definition of sous-vide is to cook the raw material under controlled temperatures (generally < 100°C) and time in heat durable vacuum pouches (Schellekens, 1996). Sous – vide products that are heated at mild temperatures for long period of time are subsequently cooled down (1-4°C) and kept under chilled conditions. In this context, the shelf life of sous vide products is between 6 to 42 days under refrigerated storage conditions (García-Linares et al., 2004).

Microbiological changes in perishable products such as seafood are relatively faster compared to semi or not- perishable products due to neutral pH, high water activity, and nutritional composition. The supported growth of both pathogenic and spoilage organisms could be observed due to the intrinsic parameters of seafood. In terms of seafood safety (i.e sous-vide seafood products) *Clostridium botulinum* Type B and E and *Listeria monocytogenes* are causing health risk after consumption of contaminated products or products that contain inadequately eliminated bacteria or spores. In accordance with the direction that is reported by (Advisory Committee on the Microbiological Safety of Food, 1992), a heat treatment at 70°C for 2 min or an equivalent heat process is required for the elimination of non-spore forming bacteria (i.e *L.monocytogenes*). In a study conducted by (Lindström et al., 2003) thermal inactivation of non-proteolytic *C. botolinum* type E spores in vacuum packed hot smoked fish products and model fish media were investigated and researchers were reported that the decimal reduction times for the heat-resistant spore fraction in rainbow trout medium were 255, 98, and 4.2 min at 75, 85, and 93°C and in whitefish medium were 55 and 7.1 min at 81 and 90°C, respectively. On the other hand, decimal reductions are also reported by Picouet et al. (2011) for total viable counts and Enterobacteriaceae. The stability of sous vide cooked salmon loins was investigated. In accordance with the results the decimal reductions for sous – vide cooked samples were found to be 4.5 log cfu/g for TVC and 3.0 log cfu/g for Enterobacteriaceae under at 40.7±0.1 °C for 19 min. of cooking conditions.

Received 08 Dec., 2017
Accepted 29 May, 2018
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Temperature and duration of sous-vide cooking are the most significant factors for the sensory quality of seafood based sous-vide products (Creed, 1995). In a study reported by Gonzáles-Fandos et al. (2005) microbiological and sensory characteristics of sous-vide cooked salmon slices were determined. Three different combinations of time–temperature were taken into account (5 min/65 °C, 10 min/90 °C and 15 min/90 °C) in the study. Researchers have concluded that the treatment at 90 °C for 15 min was the most effective one to ensure the safety and to extend the shelf life of sous vide salmon. However, the sensory results were somewhat unstable regarding the temperature increase. In contrast with this, researchers have investigated the microbial safety and sensory quality of sous-vide processed rainbow trout (*Oncorhynchus mykiss)* at 90°C for 3.3 min, at 90°C for 1.04 min and at 70°C for 5.18 min. Researchers were reported that the sensory properties of rainbow trout samples were preserved for samples that processed at 90°C for 3.3 min and were occurred to be the most effective temperature and time combination among the treatments to ensure the safety and extend the shelf-life of sous-vide rainbow trout samples (Gonzáles-Fandos et al., 2004).

There are several studies for the assessments of microbiological, chemical and sensory quality of sous-vide processed meat (Aran, 2001; Nyati, 2000), poultry (Juneja et al., 2006; Wang et al., 2004) and seafood (Díaz et al., 2009; Sebastiá et al., 2010; Nieva-Echevarría et al., 2017, 2018; Llave et al., 2018; Çetinkaya et al., 2016; Ceylan & Şengör, 2017) as affected by temperature, processing time and sauce addition. However, there is no detailed study regarding determine the quality changes of sous vide processed sea bass which has a significant place in European countries and Turkey. In this context, the aim of this study is to determine the effect of laurel and curcuma on the sensory, chemical and microbiological quality of vacuum packed sous-vide processed sea bass under chilled conditions.

2 Materials and methods

A total of 100 fish were individually used at the average weight of 250 ± 15.25 g for conducting the experiment and were obtained from the commercial circuit (6 hours from harvesting) and transferred to the laboratory with drained polystyrene box in ice. The loin part of the fish without skin was used for all analysis. Specimens were packed individually and 33 packed of C group, 33 packed of laurel (LS) and curcuma samples (CS). Subsequently, fish were gutted and washed by following the Good Hygiene Practices (GHP) and Good Manufacturing Practices (GMP) at relatively low room temperature (approx. 15-17°C). Following this, ground laurel and curcuma was added on raw fish and specimens were packed in a polyethylene polyamide pouch with O_2 transmission rate of 80 cc/m^2/day and water vapor permeability of 8.5 gr/m^2/day. Specimens were individually packed by using a vacuum packaging machine (DZ-350-MS, Turkey). Heat process was performed for the sous-vide processing in a temperature controlled water bath (Memmert GmbH, Germany) at 90°C for 10 min (Gonzáles-Fandos et al., 2005). The ratio of fish/ingredients was 1:0.002 used in accordance with the preliminary study (results not shown).

The experiment was consisting of 3 batches namely sous vide cooked vacuum packed sea bass with no addition of ingredients as control (C), vacuum packed and laurel added samples (LS) and finally curcuma added specimens (CS). In each batch of the study approx. 30 samples were used and 2 samples were analyzed in each sampling day (i.e one for sensory analysis and one for chemical and microbiological analysis. The intervals of the sampling days varying from 4 to 6 days. When necessary the third sample was used for sensory, chemical or microbial analysis). Samples were stored under refrigerated (3 ± 1 °C) conditions for 60 days. All analysis mentioned below were carried out in duplicates.

Ten grams of samples from each batch were aseptically weighted with 90 ml of 0.1% buffered peptone water (Merck Darmstadt Germany) and homogenized in a stomacher for 2 min. Decimal dilutions were prepared from the first diluent. Pour plate method was used for the enumeration of total mesophilic aerobic (TMAB), total psychrophilic aerobic bacteria (TPAB) and members of Enterobacteriaceae family. For the enumeration of TMAB and TPAB plate count agar (Merck Darmstadt Germany) was used and incubated at 30°C and 6°C for 3 and 10 days, respectively (International Organization for Standardization, 2001). Violate Red Bile Glucose Agar (Merck Darmstadt Germany) was the media for counting Enterobacteriaceae family members and incubated at 37°C for 2 days. Purple/red colonies with purple halo were counted as the members of Enterobacteriaceae family members.

The pH of fish muscle was measured in a fish/water (1:10, w/v) homogenate, using a pH-meter (HANNA HI-221). Total volatile basic nitrogen (TVB-N) was determined by steam distillation according to the method described by (Antonacopoulos & Vyncke, 1989) Trimethil amine – nigrogen (TMA-N) content of the samples was determined in accordance the method of (Association of Official Analytical Chemists, 1998).

The sensory analysis of the samples was performed by 10 trained (British Standards Institution, 2014) panelists from Fishing and Processing Technology Department in Suleyman Demirel University (International Organization for Standardization, 1994). Torry scheme was used for sensory evaluation of the samples. Regarding torry scheme samples were analyzed in terms of their flavor, odor and texture. Ten point scale was used to quantify the sensory results (10-8, excellent; 7-5, good and ≤4 not acceptable). The attributes of each parameter were determined in accordance with particular changes such as flavor (i.e soil, seaweed, bitter, sour, putrid), odor (i.e fresh, seaweed, neutral, metallic, rancid) and texture (hardness, juiciness, gumminess) (International Organization for Standardization, 1985).

For the determination of significant effects among the groups and storage time Tukey's HSD test was used in SPSS® 20.0 package program. The significant level was set at $p<0.05$. Principal Component Analysis (PCA) was performed for each of the mean scores of determinative attributes namely time, TMAB, TPAB, Enterobacteriaceae count, TVB-N, TMA-N, pH and sensory results of control LS and CS samples. The PCA is used to describe the observed data by inter-correlated quantitative dependent variables. Moreover, the similarities and differences among the mean results of the variables that repeated measures from the study were described by PCA. The quality of PCA method was determined by cross-validation method (Abdi & Williams 2010; Zhou et al., 2017). Multivariate analysis was performed by using Minitab 16 statistical package program (Minitab Ltd. USA).

3 Results and discussion

The counts of microorganisms in sous-vide processed vacuum packaged sea bass samples were presented in Table 1. The initial TMAB counts of C, LS and CS were found to be 1.00, 1.00 and 3.25 ± 0.19 log cfu/g, respectively. Regarding time increase, the counts of TMAB fluctuated. After 35 days of storage, the counts of TMAB were found to be 5.57 ± 0.04, 2.22 ± 0.03 and 3.97 ± 0.006 log cfu/g, respectively. The differences between the groups were statistically significant ($p < 0.05$). The population of TMAB was slightly increased for LS and CS samples after day 35 and at the end of the storage time the highest TMAB counts compared to those of C and CS samples were found in LS samples with 6.30 ± 0.32 log cfu/g ($p < 0.05$). The initial TMAB counts were lower than the study conducted by Gonzáles-Fandos et al. (2005) for raw salmon processed by sous-vide method. In the study, the counts of mesophiles were found to be 4.77±0.42 log cfu/g for raw salmon. In another study the initial counts of mesophilic bacteria were reported to be 1.36 log cfu/g for sous-vide processed mackerel fillets. The counts of mesophilic bacteria were increased to 5.35 log cfu/g after 9 week of chilled (2°C) storage for sous vide mackerel fillets (Dogruyol & Mol, 2017). Çağlak et al. (2017) were investigated the temperature effect of sous-vide technique on the quality changes of zander (*Sander lucioperca*) fillets. Researchers reported the initial total mesophilic aerobic counts (TMAB) of zander fillets 3.80 log cfu/g and the counts of TMAB were found to be 6.49 log cfu/g on day 42 for zander fillets sous-vide processed at 60°C and 6.34 and 3.43 log cfu/g fort he fillets which were abused to 70 and 80°C during sous-vide. It is obvious that sous-vide processing in which the temperature had a significant effect in decreasing the counts of the microbial population. However, the results of this study showed that the growth of TMAB was not inhibited or eliminated by the addition of ground laurel and curcuma in sous-vide sea bass. This could be due to the natural microflora and microbial load in seasoning material (Ereifej et al., 2015).

The changes in TPAB count of sous-vide processed vacuum packed sea bass samples under chilled conditions were shown in Table 1. The initial counts of TPAB of C, LS and CS samples were found to be 1.03 ± 0.05, 1.16 ± 0.22 and 1.07 ± 0.10 log cfu/g, respectively. Psychrophilic bacteria were multiplied during the storage and found to be 3.38 ± 0.30 and 5.47 ± 0.04 log cfu/g on day 27 and 35 for C samples, respectively. At the end of the storage time, the counts of TPAB were found to be 4.00 ± 2.32 log cfu/g. The highest TPAB count was found in LS samples 6.12 ± 0.42 log cfu/g while lower counts were found in CS samples 5.75 ± 0.30 log cfu/g at the end of the storage time. During 59 days of storage, none of the samples were exceeded the permitted limit of 7.00 log cfu/g which is the acceptable microorganism limit for freshwater and marine species (International Commission on Microbiological Specifications for Foods, 1978). Higher results were reported for sous-vide processed mackerel fillets on day zero and at the end of 9 weeks chilled storage such as 0.67 and 7.43 log cfu/g, respectively (Dogruyol & Mol, 2017). In contrast, similar results were obtained in sous-vide processed zander fillets for the counts of total psychrophilic aerobic bacteria (TPAB) which are abused to 80°C (Çağlak et al., 2017).

Regarding the TPAB results of this study hurdle effect of heat application, laurel and curcuma add and chilled storage are resulted to delay the growth of psychrophilic bacteria count. Additionally, a decrease in the counts and delay in growth of psychrophilic bacteria could be due to cold shock during

Table 1. The effects of laurel and curcuma addition on microbiological quality of sous-vide processed gutted vacuum packed sea bass at 3°C (Mean ± S.D. in log cfu/g).

Storage time (days)	TMAB C	TMAB LS	TMAB CS	TPAB C	TPAB LS	TPAB CS	Enterobacteriaceae C	Enterobacteriaceae LS	Enterobacteriaceae CS
0	1.03 ± 0.05^{aA}	1.00 ± 0.00^{aA}	3.25 ± 0.19^{abB}	1.03 ± 0.05^{a}	1.16 ± 0.22^{a}	1.07 ± 0.10^{a}	2.00 ± 0.00^{abB}	1.05 ± 0.08^{aA}	1.05 ± 0.08^{aA}
7	4.38 ± 0.32^{eC}	1.00 ± 0.00^{aA}	3.97 ± 0.009^{bcB}	1.08 ± 0.12	1.03 ± 0.05	1.19 ± 0.26	2.98 ± 0.32^{abB}	1.18 ± 0.25^{A}	2.59 ± 0.19^{abB}
13	1.38 ± 0.12^{abAB}	1.03 ± 0.05^{aA}	2.96 ± 0.04^{aB}	1.19 ± 0.26	1.16 ± 0.22	1.16 ± 0.22	1.23 ± 0.33^{a}	1.07 ± 0.10	1.52 ± 0.73^{ab}
17	2.68 ± 0.02^{cdA}	2.92 ± 0.01^{cB}	3.34 ± 0.36^{abBC}	1.16 ± 0.22	1.03 ± 0.05	1.10 ± 0.14	1.03 ± 0.05	1.23 ± 0.33	1.05 ± 0.08^{a}
21	1.79 ± 0.02^{abB}	1.19 ± 0.26^{aA}	3.44 ± 0.02^{abC}	1.19 ± 0.26	1.08 ± 0.12	1.03 ± 0.05	1.16 ± 0.22	1.26 ± 0.37	1.03 ± 0.05^{a}
27	1.73 ± 0.05^{abA}	4.06 ± 0.01^{dB}	4.26 ± 0.05^{cC}	3.38 ± 0.30^{abC}	1.11 ± 0.16^{A}	2.51 ± 0.00^{B}	1.03 ± 0.05^{A}	1.07 ± 0.10^{A}	3.34 ± 0.06^{bB}
31	2.09 ± 0.19^{bcA}	2.78 ± 0.11^{bcB}	2.94 ± 0.05^{aBC}	3.19 ± 0.15^{abB}	1.17 ± 0.24^{A}	1.19 ± 0.28^{A}	1.05 ± 0.08	1.12 ± 0.18	1.07 ± 0.10^{a}
35	5.57 ± 0.04^{fC}	2.22 ± 0.03^{bcA}	3.97 ± 0.006^{bcB}	5.47 ± 0.04^{bB}	1.15 ± 0.21^{A}	1.22 ± 0.19^{A}	1.03 ± 0.05	1.05 ± 0.08	1.05 ± 0.08^{a}
41	3.32 ± 0.10^{dA}	4.48 ± 0.12^{dB}	3.28 ± 0.15^{abA}	1.71 ± 1.01^{ab}	1.19 ± 0.28	1.19 ± 0.26	1.03 ± 0.05	1.16 ± 0.22	1.07 ± 0.10^{a}
45	1.15 ± 0.21^{aA}	2.05 ± 0.38^{bB}	2.96 ± 0.50^{aB}	1.15 ± 0.21^{a}	1.17 ± 0.24	1.25 ± 0.35	1.08 ± 0.12	1.07 ± 0.10	1.05 ± 0.08^{a}
49	1.15 ± 0.21^{aA}	5.43 ± 0.18^{eC}	2.94 ± 0.00^{aB}	1.15 ± 0.21^{a}	1.53 ± 0.08	1.69 ± 0.12	1.08 ± 0.12	1.05 ± 0.08	1.16 ± 0.22^{a}
55	1.69 ± 0.00^{abA}	4.00 ± 0.21^{dC}	3.82 ± 0.06^{bcB}	2.74 ± 1.25^{ab}	2.57 ± 1.11	2.15 ± 1.47	1.95 ± 1.65	2.00 ± 1.61^{ab}	2.07 ± 1.55^{ab}
59	2.85 ± 0.53^{cdA}	6.30 ± 0.32^{fB}	6.01 ± 0.02^{dB}	4.00 ± 2.32^{ab}	6.12 ± 0.42^{b}	5.75 ± 0.30^{b}	3.90 ± 2.66^{bA}	3.92 ± 2.64^{bA}	5.89 ± 0.22^{cB}

storage. Results of this study were in agreement with those of reported by (Jeyasekaran et al., 2011) that investigated the microbiological quality of cuttlefish (*Sephia pharaonis*) fillets stored under chilled conditions.

The presence of Enterobacteriaceae family in freshwater and marine fish species closely related to the quality and safety issues (Ghanem et al., 2014; Goulas et al., 2005). The initial count of Enterobacteriaceae family was found to be 2.00 ± 0.00, 1.05 ± 0.08 and 1.05 ± 0.08 log cfu/g for C, LS and CS samples. The only significant increase in Enterobacteriaceae count was found at the end of storage time ($p<0.05$). The results agreed with those from Gonzáles-Fandos et al. (2004, 2005) that investigated the microbiological safety and sensory quality of rainbow trout (*Oncorhynchus mykiss*) processed by sous-vide method and microbiological safety and sensory characteristics of salmon slices processed by sous-vide method, respectively.

The value of TVB-N is one of the most acceptable parameters for determination the quality of seafood. However, it mostly reflects the later stage of the spoilage in fishery products (Riquixo, 1998). The initial amount of TVB-N depends on species and varies between 10 to 15 mg N/100 g sample except for pelagic fish species (Etienne, 2005). The changes in TVB-N values of sous-vide vacuum packed sea bass samples are shown in Figure 1a. The initial TVB-N value of C, LS and CS samples were found to be 14.00 ± 3.95, 9.80 ± 1.97 and 15.40 ± 1.97 mg N/100 g sample. During 35 days of storage TVB-N values were not exceeded 20 mg N/100 sample. The highest amount of TVB-N values were found in LS samples at the end of the storage time (30.80 ± 7.91 mg N/100 g). The mean values of group LS were found statistically significant ($p<0.05$) compared to those of other groups. The tested groups (C, LS and CS) were exhibited good quality in terms of TVB-N values during 59 days of storage and none of the samples were exceed the acceptability limit of 35-40 mg N/100 g sample as reported by EU guidelines (European Union, 1995). Similar results for the increase in TVB-N values regarding storage time have been reported for lemon juice added sous-vide bonito (*Sarda* sarda) (Cocansu et al., 2011). Çetinkaya et al. (2017) were investigated the effect of rosemary on the quality changes of sous-vide processed rainbow trout. While initial TVB-N values of rosemary added samples were reported 13.36 mg N / 100 g, after 45 days of storage the TVB-N values of rosemary added samples were found to be 21.94 mg N / 100 g. In another study, initial TVB-N values of makerel fillets were found to be 17.71 and at the end of 9 weeks chilled storage TVB-N values were incresed to 36.53 mg N/100 g (Dogruyol & Mol, 2017).

The changes in TMA-N content of sous-vide processed vacuum packaged sea bass samples were shown in Figure 1b. The initial TMA-N content of C, LS and CS samples were found to be 0.57 ± 0.32, 3.16 ± 0.03 and 4.43 ± 0.09 mg N/100g sample, respectively. The content of TMA-N in all samples was below the acceptable limits of 5-10 mg N/100 g (Sikorski et al., 1990) and 8 mg N/100 g sample (Varlık et al., 1993) at the end of the storage time. The differences between the groups were found statistically significant ($p<0.05$). The results of TMA-N content of the samples were in agreement to those of reported by (Baygar et al., 2012) for sea bass marinades under chilled conditions. However, at the end of the storage time higher amount of TMA-N was observed for the LS (4.51 ± 0.09) and CS (4.57 ± 0.01) added vacuum packaged sea bass samples than those of reported by Baygar et al. (2012) (32.2 mg N /kg for scale fillets and 29.7 mg N/kg for descaled samples). TMA-N product of reduced TMA-N oxide by the spoilage bacteria during the storage of the marine fish. The higher amount of TMA-N in this study could be due to the microbial growth and higher pH compared to marinated/organic acid added sous-vide samples (Cocansu et al., 2011).

Mean values ± standard deviations of the pH values of the samples during storage were presented in Figure 1c. The initial pH values of C samples (6.95 ± 0.03) were significantly ($p<0.05$)

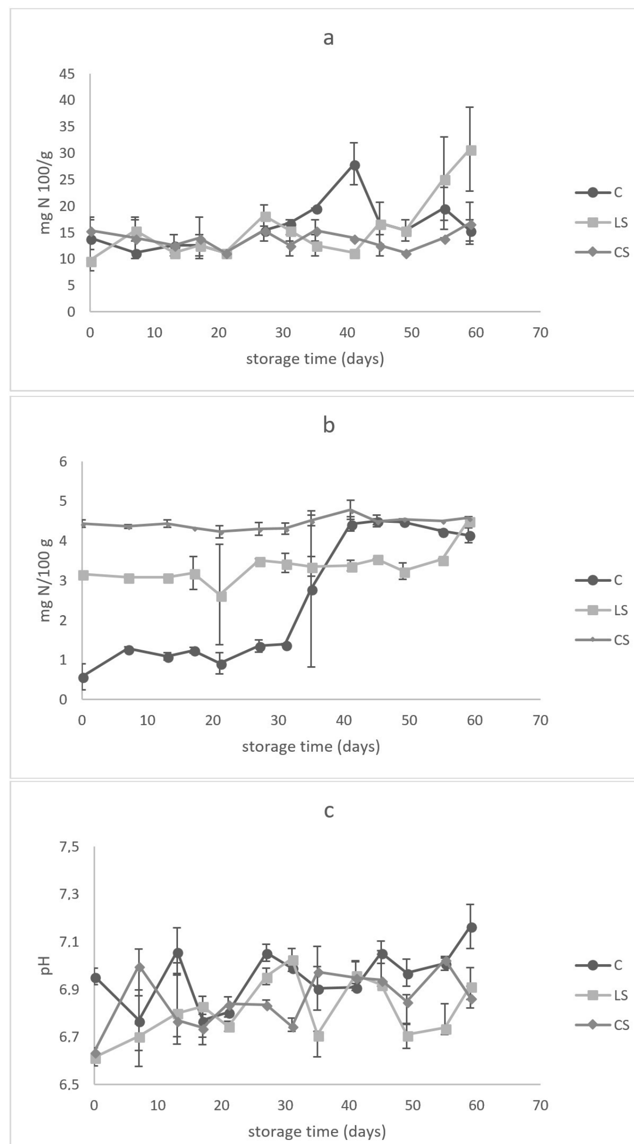

Figure 1. The effects of laurel and curcuma addition on chemical quality of sous-i processed vacuum packed sea bass at 3°C. (a) Total Volatile Basic Nitrogen (TVB-N) (mg N/100 g), (b) Trimethylamine Nitrogen (TMA-N) (mg N/100 g) and (c) pH.

Table 2. The effects of laurel and curcuma addition on sensory quality of sous-vide processed vacuum packed sea bass under chilled conditions at 3°C (Mean ± S.D).

Storage time (days)	C-flavour	C-odour	C-texture	C-Total Sensory changes	LS-flavour	LS-odour	LS-texture	LS-Total Sensory changes	CS-flavour	CS-odour	CS-texture	CS-Total Sensory changes
1	6.50 ± 1.35^{bcd}	6.60 ± 1.50^{ef}	6.40 ± 1.50^{de}	19.50 ± 0.10	7.20 ± 1.98^{b}	7.30 ± 1.15^{de}	6.90 ± 1.72^{efg}	21.40 ± 0.20	6.70 ± 1.49^{b}	6.60 ± 0.96^{def}	7.30 ± 1.49^{e}	20.60 ± 0.37
7	7.30 ± 1.05^{cd}	8.00 ± 2.49^{fg}	6.00 ± 1.70^{cde}	21.30 ± 1.10	7.10 ± 1.19^{b}	8.70 ± 0.67^{e}	6.30 ± 1.49^{ef}	22.10 ± 1.22	7.30 ± 1.05^{b}	7.70 ± 1.49^{f}	5.40 ± 1.42^{cde}	20.40 ± 1.22
13	7.60 ± 1.50^{d}	8.90 ± 1.28^{g}	7.40 ± 1.57^{e}	23.90 ± 0.81	6.90 ± 1.96^{b}	7.60 ± 1.50^{de}	8.50 ± 1.71^{g}	23.00 ± 0.80	7.50 ± 1.50^{b}	7.90 ± 1.28^{f}	7.20 ± 2.04^{e}	22.60 ± 0.35
17	6.20 ± 0.78^{bc}	6.60 ± 1.07^{ef}	5.80 ± 1.03^{cde}	18.60 ± 0.40	7.30 ± 1.05^{b}	6.70 ± 0.94^{d}	6.70 ± 0.82^{efg}	20.70 ± 0.34	7.70 ± 0.67^{b}	7.90 ± 0.56^{f}	6.90 ± 1.19^{e}	22.50 ± 0.52
21	6.60 ± 1.17^{bcd}	6.50 ± 1.26^{def}	5.60 ± 1.57^{bcde}	18.70 ± 0.55	7.30 ± 0.67^{b}	7.40 ± 1.34^{de}	7.00 ± 1.05^{fg}	21.70 ± 0.20	7.40 ± 1.07^{b}	7.50 ± 0.84^{ef}	6.80 ± 1.31^{e}	21.70 ± 0.37
27	5.80 ± 0.42^{b}	5.70 ± 0.82^{cde}	5.50 ± 0.52^{bcde}	17.00 ± 0.15	6.00 ± 0.47^{b}	5.70 ± 1.05^{cd}	5.80 ± 0.78^{de}	17.50 ± 0.15	$6{,}80 \pm 0.91^{b}$	6.70 ± 0.82^{def}	6.10 ± 0.31^{ed}	19.60 ± 0.37
31	1.00 ± 0.0^{a}	5.70 ± 1.49^{cde}	5.80 ± 1.75^{cde}	12.50 ± 0.07	1.00 ± 0.0^{a}	5.80 ± 2.09^{cd}	4.80 ± 1.68^{cde}	11.60 ± 0.70	1.00 ± 0.0^{a}	7.20 ± 2.29^{ef}	5.50 ± 1.58^{cde}	13.70 ± 1.20
35	1.00 ± 0.0	4.10 ± 2.28^{abc}	4.90 ± 1.37^{bcd}	10.00 ± 0.56	1.00 ± 0.0	2.90 ± 1.66^{ab}	3.30 ± 1.56^{abc}	7.20 ± 0.28	1.00 ± 0.0	4.40 ± 1.83^{bc}	4.70 ± 1.70^{bcd}	10.10 ± 0.21
41	1.00 ± 0.0	3.90 ± 1.19^{abc}	3.60 ± 1.07^{ab}	8.50 ± 0.21	1.00 ± 0.0	4.20 ± 1.68^{bc}	3.70 ± 1.25^{bcd}	8.90 ± 0.35	1.00 ± 0.0	4.30 ± 1.05^{bc}	3.80 ± 0.91^{bc}	9.10 ± 0.35
45	1.00 ± 0.0	4.30 ± 1.82^{bcd}	4.70 ± 2.26^{bcd}	10.00 ± 0.28	1.00 ± 0.0	5.90 ± 1.52^{cd}	6.00 ± 1.82^{ef}	12.90 ± 0.07	1.00 ± 0.0	5.70 ± 1.63^{cde}	6.00 ± 2.10^{ed}	12.70 ± 0.21
49	1.00 ± 0.0	4.50 ± 1.08^{bcde}	4.20 ± 1.03^{bc}	9.70 ± 0.21	1.00 ± 0.0	5.80 ± 0.91^{cd}	4.80 ± 0.78^{cde}	11.60 ± 0.70	1.00 ± 0.0	4.80 ± 0.91^{bcd}	4.70 ± 1.05^{bcd}	10.50 ± 0.07
55	1.00 ± 0.0	2.40 ± 0.84^{bc}	1.80 ± 0.78^{a}	5.20 ± 0.42	1.00 ± 0.0	1.70 ± 0.67^{a}	2.50 ± 2.32^{ab}	5.20 ± 0.56	1.00 ± 0.0	2.90 ± 0.99^{ab}	3.10 ± 0.99^{ab}	7.00 ± 0.14
59	1.00 ± 0.0	2.0 ± 0.81^{a}	1.90 ± 0.56^{a}	4.90 ± 0.07	1.00 ± 0.0	1.00 ± 0.00^{a}	1.20 ± 0.42^{a}	3.20 ± 0.14	1.00 ± 0.0	1.50 ± 0.52^{a}	1.70 ± 0.67^{a}	4.20 ± 0.14

higher than those of laurel and curcuma added samples. During 59 days of storage, the pH values were changed from 6.61 to 6.91 for LS and 6.64 to 6.86 for CS and significant differences ($p<0.05$) were only found between storage time for each of the sample. Lower results were reported for the sea bass stored on ice (Bojanic et al., 2009). The variation of the pH values could be the result of pre-slaughter and processing methods which could cause pH variations during storage (Bagni et al., 2007).

Sensory scores of the tested samples were presented in Table 2. The initial flavor, odor and texture results of the samples are indicating that the samples were of good quality. The differences between treatments were not significant ($p>0.05$) at the beginning of the storage trial for C, LS and CS samples. The flavor changes of the samples were dramatically decreased ($p<0.05$) after 27 days of the storage. However, laurel (6.00 ± 0.47) and curcuma (6.80 ± 0.91) added samples had higher flavor scores than those of control (5.80 ± 0.42) samples. One of the most determinative parameters for the quality changes of seafood is odor. During 59 days of storage odor scores of the samples were slightly changed. On day 35 the sensory scores of the samples were reached to unacceptable limits. Lowest scores were found in LS samples (2.90 ± 1.66) on day 35. While C and CS samples had 4.10 ± 2.28 and 4.40 ± 1.83, respectively. Regarding texture changes lowest scores were obtained after day 55 for all of the samples (Table 2). The contribution of variables was calculated by PCA to evaluate the effect of seasoning on the quality of vacuum packed sea bass during 59 days of storage and the results were shown in Figure 2. The 1st principal component (PC1) explained 51% of total variations. Positive correlations were found between chemical (pH, TVB-N, TMA) and microbiological (TMA and TPA) changes in accordance with storage time. Regarding the sensory changes, negative strong correlations were found between chemical and microbiological changes. In contrast to this, the changes in the counts of Enterobacteriaceae family were positively correlated with the changes of sensory results (Figure 2).

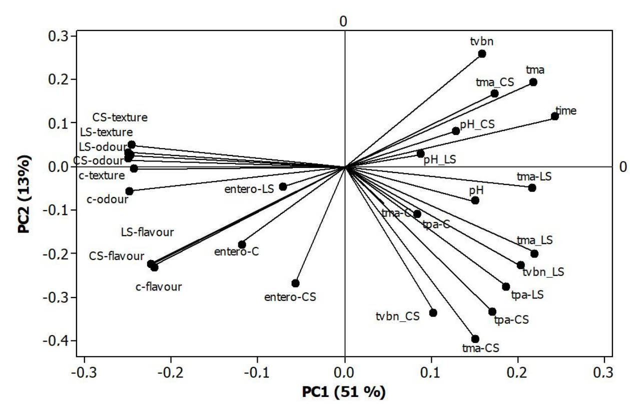

Figure 2. Loading plots of sous-vide processed vacuum packed sea bass at 3°C.

Second principal component (PC2) explained 13% of the variations. Obviously, PC2 contributes to differentiating the contributions of the variables and groups. The location of pH, TMA, odor and texture load of LS and CS samples were on the up side of the plots while TMA, Enterobacteriaceae, TPA, TVB-N, pH, flavor, odor and texture loads of C, CS and LS samples were located on the downside of the loading plots.

From the score plot, it is obvious that Enterobacteriaceae count of LS samples are not significantly contributed to explain the quality loss of vacuum packed sous vide sea bass samples (Figure 3). However, the effects of Enterobacteriaceae counts, TMA, odor TPAB, flavor and texture for the control samples and TMA, TVB, TPAB, pH for LS samples and finally TMA for

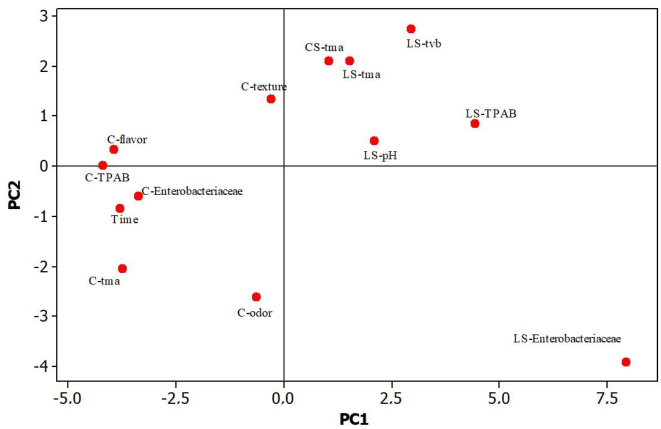

Figure 3. Score plots of sous-vide processed vacuum packed sea bass at 3°C.

CS samples on the determination of the shelf life of vacuum packed sous vide processed sea bass samples were explained.

By taking into account the changes in quality parameters, the addition of ground laurel and curcuma in vacuum packed sous-vide processed sea bass did significantly ($p<0.05$) extend the shelf life of processed samples (approx. 8 and 22%). Moreover, results from this study showed that seasoning has positive effects on sensory results and maintain the quality during the storage.

4 Conclusions

The quality of the processed (i.e sous-vide) products, particularly in seafood, strictly depends on initial quality indices (microbiological, chemical and sensory) of the raw material. Maintenance of quality and safety of a final product should provide satisfactory results in terms of consumption. Sous-vide processing is one of the most popular and emerging technology to maintain and protect seafood from spoilage and being a source of food-borne pathogens as there are several steps in the processing such as temperature application and modified atmosphere (i.e vacuum packaging). Results of this study showed that the shelf life of seasoned gutted and vacuum packed sea bass samples was extended approximately 4 and 10 days for LS and CS added samples, respectively. Higher concentration of ground laurel and curcuma could prolong the shelf life of the product, however appreciation of the sous-vide processed sea bass samples by the consumers would be lower based on sensory results.

Acknowledgements

Authors thank The Scientific and Technological Research Council (TÜBİTAK-2209/A – University Students Domestic Research Projects) for funding the project.

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