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Evaluation of the Microbiological, Chemical and Sensory Quality of Carp Processed by the Sous Vide Method

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***Abstract*—**This study evaluated the microbiological quality and the sensory characteristics of carp fillets processed by the sousvide method when stored at 2 and 10 °C. Four different combinations of sauced–storage were studied then stored at 2 or 10 °C was evaluate periodically sensory, microbiological and chemical quality. Batches stored at 2 °C had lower growth rates of mesophiles and psychrotrophs. Moreover, these counts decreased by increasing the heating temperature and time. Staphylococcus aureus, Bacillus cereus, Clostridium perfringens and Listeria monocytogenes were not found in any of the samples. The heat treatment of 90 °C for 15 min and sauced was the most effective to ensure the safety and extend the shelf-life of sousvide carp preserving its sensory characteristics. This study establishes the microbiological quality of sous vide carp and emphasizes the relevance of the raw materials, heat treatment and storage temperature to ensure the safety of the product.

***Keywords*—**Sous- vide methods, carp, sauce, microbiological, chemical and sensory quality

I. Introduction

FRESH fish and marine products are extremely perishable as compared to other fresh meat commodities. The hygienic quality of fish and marine products declines rapidly due to microbial cross-contamination from various sources, ultimately leading to spoilage. Carp, as defined a freshwater fish species, has been one of the most widely cultured species all over the world due to its fast growth rate, easy cultivation and high feed efficiency ratio. Carp farms have proliferated in the last decades and a subsequent oversaturation of the trout market has occurred in some areas. Another issue to be taken into account is that fish has a very short shelf-life due to its high water content and its neutral pH [1]. All these reasons support the need to further extend this product to markets far away from the production sites, increase its shelf-life and even diversify the offer in order to meet an increasing consumer demand for convenient and safe fresh food of high organoleptic quality, free of additives and preservatives and with the appearance and taste of freshly homeprepared food as a consequence of the reduction in the time devoted to cooking at home.Catering services, food processing plants and retail sectors are employing novel methods to deliver home-made style meat-based meals of high quality and with a long shelf-life [2]-[4]. Present trends involve cooking the meat inside the final packaging in its own juice or accompanied by a sauce in order to make the cooking, preservation treatment and, frequently, final presentation a one-step process. Sous vide technology is defined ‘‘food cooked under controlled conditions of temperature and time inside heat-stable vacuum pouches”. The sous vide technology could be a reasonable choice, as it allows to obtain products with an extended shelf-life and a quality similar to that of fresh food (Schellekens and Martens, 1992). Sous vide or vacuum cooked food is defined as ‘‘raw materials or raw materials with intermediate foods, that are cooked under controlled conditions of temperature and time inside heat-stable vacuum pouches’’ [5]. Sous vide method involves cooking/ pasteurisation temperatures of 65–95 ºC applied over long periods (upto 16 h), followed by rapid cooling to attain a temperature of 3 ºC in the centre of the product [2]. Dishes are stored at temperatures below 3.3 ºC to prevent the growth of Clostridium botulinum, Bacillus cereus and other pathogenic microbes resistant to the pasteurisation [6]. However, refrigerated sous vide meat can suffer spoilage by the action of lactic acid bacteria [7], [8] which produce sour off-flavours and off-odours, milky exudates, a slimy texture and CO2, which may cause swelling of the pack and/or greening [9]. Moulds and yeasts can also grow in refrigerated sous vide meats [10]-[12]. In addition, meat prepared by this method may undergo proteolysis, lipolysis and enzymatic and chemical oxidation during refrigerated storage, leading to changes in texture, colour, odour and flavour, sometimes accompanied by a loss of firmness, darkening, rancidity, sourness and other off-odours and off-flavours.

The UK Advisory Committee on the Microbiological Safety of Food recommends for cooked-chilled products with an extended shelf-life of more than 10 days a heat treatment at 90ºC for 10 min or equivalent lethality and strict chill conditions to control Clostridium botulinum. In order to eliminate non-sporeforming pathogens such as Listeria monocytogenes, a heat treatment at 70ºC for 2 min or an equivalent heat process is required [13]. An adequate heat treatment must achieve at least a six-log reduction cycle in the psychrotrophic strains of Clostridium botulinum and Listeria monocytogenes. The treatments necessary to reach a significant reduction in the number of Cl. botulinum spores cause unacceptable thermal damages to some products, and for that reason less severe heat treatments have been proposed. However, additional hurdles should be incorporated [14].The aim of this work was to evaluate the shelf-life, microbiological, chemical quality and sensorial

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characteristics of marinate carp processed by the sous vide method under different storage conditions at 2ºC and 10ºC.

II. Material and Methods

A.Sample Preparation

The carp (Cyprinus carpio L. 1758), between 14 and 16 kg in weight, were captured from local market. Having been transferred to the laboratory, the fish were beheaded, gutted and washed. Then, they were filleted. The filet were divided similar thickness, each of which weighed 70-80 grams. Prepared fillets were separated into two groups. The first group was assigned as the control group. Three percent dry salting process was applied to control group samples. Second group was added in the sauce (30% tomato paste, 20% lemon juice, 30% oil, 10% garlic, 4% water, 3% salt, 1% red pepper, 1% cumin and 1% thyme). This type of sauce was chosen preliminary studies, the most appreciated formula. The weight of the sauce used was 20% of the fish weight. The storage temperature/sauce combinations were tested as a four different groups. Control and 2 ºC for storage (a), control and 10 ºC (b), sauced and 2 ºC (c) and sauced and 10 ºC (d). Each portion was packaged into a polyethylenepolyamide pouch with an O2 permeability of 25–30 cm3/m2/24 h and a water steam permeability of 5 g m2/24 h at 25ºC. The pouches were heat sealed using a vacuum sealing machine. The heating process was carried out in a steam oven (Arçelik, MF 2009,Turkey). All samples were cooked in an oven at 90 ºC for 15 minute. The heating profiles of vacuum-packed samples were obtained with a thermocouple (HI 9057 KJT thermocouple, Hanna instruments, Portugal) located in the geometric center of the sample. After heating, the samples were immediately chilled until reaching an internal temperature of 4 ºC. After chilling, samples were stored at 2 and 10 ºC for 0, 7, 14, 28, 42 and 56 days. Three experiments were carried out. The following determinations were made in each experiment microbiological, chemical and sensory analysis.

B. Microbiological analyses

Twenty-five grams of carp fillet were aseptically weighed and homogenized in a Stomacher for 2 min with 225 ml of sterile peptone water (0.1% peptone). Further decimal dilutions were made with the same diluent. The total number of mesophilic micro-organisms was determined on Plate Count Agar (PCA, Oxoid CM 325) following the pour plate method, and incubated at 30ºC for 72 h [15]. Psychrotrophs were determined on Plate Count Agar with an incubation temperature of 7ºC for 10 days, following the pour plate method [15]. Anaerobes were determined on PCA incubated under anaerobic conditions at 30ºC for 72 h [15]. Lactic acid bacteria were determined in MRS (Oxoid) incubated at 30ºC for 72 h [16]. Enterobacteriaceae were determined on plates of Violet Red Bile Glucose Agar (Difco, Detroit, MI). The plates were overlaid before the incubation at 37ºC for 18–24 h [15]. Staphylococcus aureus was enumerated by plating on Baird-Parker agar (Oxoid) following the surface plate method. The incubation temperature used was 37ºC (18–24 h). Suspected colonies were subjected to a DNAse test (Difco, Detroit, MI) [15]. Bacillus cereus was enumerated on Bacillus selective agar (Oxoid) incubated at 30ºC for 48 h [15]. Clostridium perfringens was enumerated on SPS agar (Oxoid) incubated at 37ºC for 48 h under anaerobic condition [15]. For Clostridium spp., 25 g of sample were homogenized and incubated at 25ºC for 24 h in Cooked Meat Medium (Oxoid), after incubation 0.1 ml of suspension was inoculated in Reinforced Clostridial Medium agar (Oxoid), and incubated at 25ºC for 10 days under anaerobiosis. Calculations of the number of strictly anaerobic bacteria were based on the proportion of isolated colonies no growing in air [17]. The presence of Listeria spp. was investigated as follows: a 25 g sample was homogenized with 225 ml of Listeria Enrichment Broth (LEB, Merck, Darmstadt) in a Stomacher. The enrichment broth was incubated at 30ºC for 48 h. LEB cultures were streaked on Palcam agar and then the plates were incubated at 37ºC for 48 h and analysed for the presence of Listeria characteristic colonies [17] , [18]. All the analyses were performed in duplicate.

C.Chemical Analyses

The method reported by Varlık [19], was employed in determination of TVB-N amount of the samples. Thiobarbituric acid value (TBA, mg malonaldehyde/kg) was determined using a spectrophotometric method [20].

D. Sensory Analyses

For the sensory analysis, samples were heated in a covered plastic container using a microwave (Balay S.A., South Korea) at full power (850 W) for 2.5 min until reaching an internal temperature of 72 ºC, as measured by a thermometer. The warmed samples were then presented to the eight panelists in small aluminium trays. The panelists were selected and trained according to ISO standards [21]. The quality of each sample was evaluated using texture, taste, color, smell, appearance and total assessment characteristic. Each characteristic was scored using a point scale ranging from 1 to 5, corresponding respectively to “very bad”, “bad”, “normal”, “good” and “very good” [22].

E. Statistical analyses

Analysis of the data was conducted using Statistical Analysis System (SAS) package program. Values between groups and within group-between days were compared. Data were subjected to variance analysis in accordance with 3 x 11 x 3 x 1 factorial design and in terms of fix effects and inter-variable interactions so that “repetition number x sampling time x test groups x number of samples examined at one instance from each test group”. According to General Linear Models (GLM) procedure, Fisher’s smallest squares average (LSD) test was used. Standard deviation figures of all averages were calculated [23]. Significance of variance value was determined as 0.05.

III. Results

A. Microbiological Quality

Microbiological results are shown in Figs.1. The raw carp fillets had initial mesophiles and anaerobes counts of 3.88 and 2.1 log cfu/g, respectively (Figs.1. a and b). The mean

mesophile counts of the carp filets that received a heat treatment control and sauced were stored at 10 ºC (b and d groups) were above 9 log cfu/g and 7 log cfu/g after 56 days, respectively. The mesophiles and anaerobes counts in the carp filets that received a and c groups were 4.55 and 3.00 log cfu/g 56 days of storage. The heat treatment had a significant effect between a, c and b, d groups on the mesophiles and anaerobes counts ($p<0.05$). The final mesophiles and anaerobes counts were significantly lower ($p<0.05$) in the samples stored at 2 ºC than in those stored at 10 ºC. In fact, the surviving cells after the sauced and heat treatment were hardly capable of growing at 2 ºC until day 56. Mesophile counts above 6 log cfu/g were reached after 28 days of storage b groups. The psychrotroph growth was lower when the heat treatment and sauced were more severe (Fig. 1.c). The psychrotroph counts were below 3 log cfu/g in the carp fillets that received a heat treatment after 0 days of storage at all groups. However, the psychrotroph counts increased between day 0 and day 45 in the 10 ºC on the storage temperature, while in the carp that received a heat treatment was very slow, 2 ºC on the storage periods in the same period. Significant differences ($p<0.05$) were found between a, c groups and b, d 56 days of storage. After the sous vide treatment, Enterobacteriaceae were detected in b and d groups after 14 days of storage at 10 ºC (2.27 and 2.13 log cfu/g). This values were increased the storage until 56 days. The groups a nd c no dedected all storage periods. The initial contamination by lactic acid bacteria of the raw carp

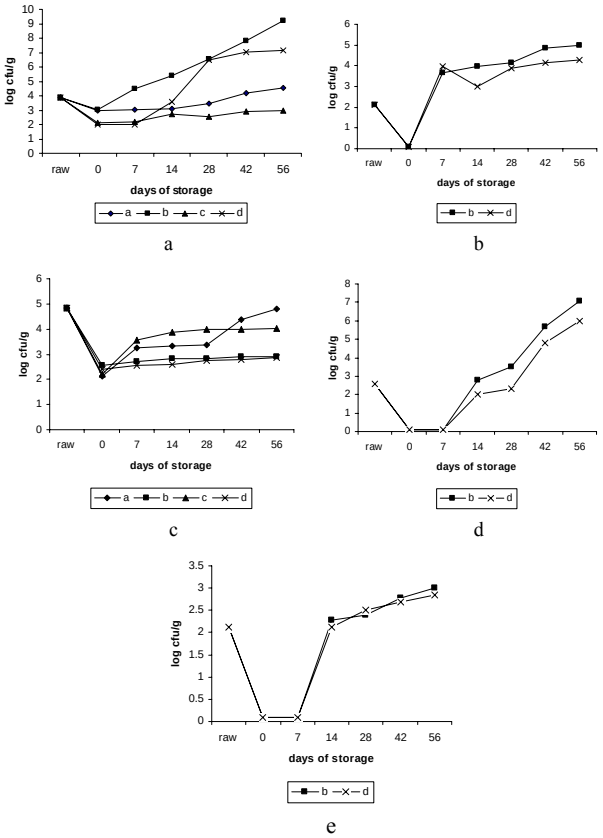

Fig.1. Impact of the processing conditions and storage temperature on the (a) mesophile counts, (b) anaerobe counts, (c) psychrotrophs counts, (d) lactic acid bacteria counts and (e) enterobacteriaceae conts in carp fillets processed by sous vide method. Control and 2 ºC for storage (a), control and 10 ºC (b), sauced and 2 ºC (c) and sauced and 10 ºC (d). Each point is the mean of three samples taken from three replicate experiments (n= 3x3). Error bars show SD.

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1 log cfu/g. After the heat treatment, the level was below the detection limit (1 log cfu/g) a and c groups (Fig.1. e). However, these bacteria reached a level above 6 log cfu/g in b an d groups on day 56. S. aureus, Bacillus cereus, Clostridium perfringens and Listeria monocytogenes were not detected in any sample. No strictly anaerobic bacteria were isolated from Reinforced Clostridial Agar.

B. Chemical Quality

The TBA value of raw material was found to be 0.4 mg/1000 g. The TBA value in the fillets of both b and d significantly increased from 0.98 to 0.81 mg/1000 g and from 4.2 to 2.7 mg/1000 g during storage time, respectively ($p < 0.05$). TBA for a and c groups (Fig.2.f) showed a very slow trend throughout the entire storage period ($p>0.05$). TVB-N values (Fig.2. g) showed an increasing trend for all samples throughout the entire storage period, with the b groups samples attaining the higher values (40.7 mg N/100 g on day 56 of storage).

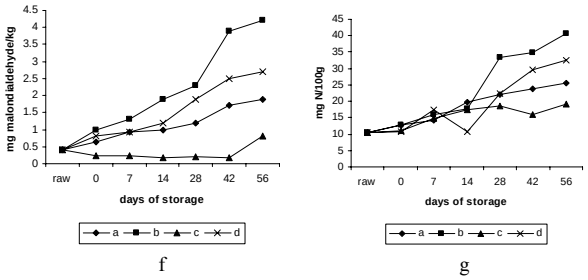

Fig.2. Impact of the processing conditions and storage temperature on the (f) Thiobarbituric acid value and (g) TVB-N values in carp fillets processed by sous vide method. Control and 2 ºC for storage (a), control and 10 ºC (b), sauced and 2 ºC (c) and sauced and 10 ºC (d). Each point is the mean of three samples taken from three replicate experiments (n= 3x3). Error bars show SD.

C. Sensory Quality

Changes detected during the storage of carp fillets in determined total assesment values are given in figure 3. The storage of carp fillets another values (texture, taste, color, smell and appearance) were given table 1.

TABLE I
RESULT OF SENSORY ANALYSİS OF CARP FİLET SAUCED AND SOUS- VİDE METHODS

		0	7	14	28	42	56
texture	A	3.56±0.08^{x,a}	3.22±0.14^{x,a}	3.43±0.09^{x,a}	3.35±0.3^{x,a}	3.42±0.27^{x,a}	3.25±0.3^{x,a}
	B	3.45±0.05^{x,a}	3.25±0.3^{x,a}	3.25±0.3^{x,a}	*	*	*
	C	4.81±0.08^{x,a}	4.81±0.08^{x,a}	4.75±0.3^{x,a}	4.56±0.2^{x,a}	4.4±0.1^{x,a}	4.56±0.1^{x,a}
	D	4.72±0.05^{x,a}	4.46±0.1^{x,a}	3.13±0.02^{x,a}	*	*	*
Color	A	2.43±0.4^{x,a}	2.43±0.9^{x,a}	2.18±0.09^{x,a}	1.56±0.07^{x,b}	1±0.01^{x,b}	1±0.01^{x,b}
	B	2.18±0.7^{x,a}	2.09±0.08^{x,a}	1.16±0.05^{x,b}	*	*	*
	C	4.81±0.2^{y,a}	4.68±0.2^{y,a}	4.37±0.1^{y,a}	4.24±0.2^{y,a}	4.5±0.03^{y,a}	4.37±0.03^{y,a}
	D	4.06±0.	3.19±0	2.15±0.	*	*	*

		3 y,a	.02 y,a	5 x,b			
Smell	A	2.56±0.08 x,a	2.56±0.08 x,a	2.81±0.08 x,a	2.74±0.17 x,a	2.51±0.14 x,a	2.43±0.09 x,a
	B	2.56±0.5 x,a	1.99±0.17 x,b	1.62±0.88 x,b	*	*	*
	C	4.68±0.26 y,a	4.68±0.26 y,a	4.56±0.26 y,a	4.87±0.17 y,a	4.56±0.26 y,a	4.5±0.35 y,a
	D	4.56±0.43 y,a	3.99±0.17 y,a	3.06±0.43 y,a	*	*	*
Aparence	A	2.68±0.2 x,a	2.68±0.2 x,a	2.56±0.2 x,a	2.56±0.43 x,a	2.24±0.6 x,a	2.18±0.5 x,a
	B	2.24±0.17 x,a	1.99±0.5 x,a	1.43±0.2 x,b	*	*	*
	C	4.75±0.01 y,a	4.68±0.09 y,a	4.61±0.05 y,a	4.31±0.5 y,a	4.31±0.5 y,a	4.21±0.03 y,a
	D	4.75±0.3 y,a	3.52±0.5 y,a	3.43±0.5 y,a	*	*	*
Taste	A	2.75±0.7 x,a	2.75±0.7 x,a	2.75±0.3 x,a	2.56±0.2 x,a	2.32±.02 x,a	1.16±0.1 x,b
	B	2.75±0.3 x,a	1.19±0.3 x,b	1±0.5 x,b	*	*	*
	C	4.6±0.3 y,a	4.6±0.3 y,a	4.56±0.2 y,a	4.43±0.09 y,a	4.31±0.03 y,a	4.16±0.3 y,a
	D	4.6±0.02 y,a	4.1±0.1 y,a	3.13±0.1 y,b	*	*	*

* not analyzed, a, b : Means within a column lacking a common superscript letter are different (P<0.05).
x, y : Means within a row lacking a common superscript letter are different (P<0.05). Values are means (±SD) for three trials at each groups (n=3x3). Control and 2 ºC for storage (a), control and 10 ºC (b), sauced and 2 ºC (c) and sauced and 10 ºC (d).

As a result of sensory evaluation of samples, when the scores they received in terms of total assessment, it can be seen that the lowest scores belonged to b group, whereas the highest scores were represented by group c. Samples were evaluated in terms of color, smell, taste, appearance and total assessment; as a result, it was detected that the difference between b and d group and groups a and c was statistically significant ($p<0.05$), whereas in terms of texture the difference between groups remained insignificant ($p>0.05$).

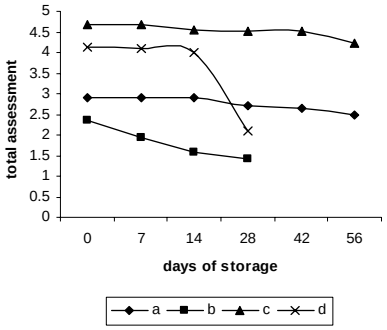

Fig. 3 Impact of storage temperature on the total sensory assessment of reheated sous vide carp fillets. Control and 2 ºC for storage (a), control and 10 ºC (b), sauced and 2 ºC (c) and sauced and 10 ºC (d). Each point is the mean of three samples taken from three replicate experiments (n= 3x3). Error bars show SD

VI. Discussion

The evolution of microbial growth in raw and treatment carp fillets along 2 and 10 ºC at storage in the shown in Fig. 1. Contamination of raw fish is well documented, being very variable depending on the water conditions and temperature. Fish from cold waters generally yield counts of 102–104 cfu/cm2 on the skin and gill surface, while its intestinal content ranges from 102 to 108 cfu/g [24]. Mesophiles counts were significantly lower ($p<0.05$) in carp stored at 2 °C compared to those stored at 10 °C for all processing treatments, this fact emphasizes the importance of storage temperature to ensure the quality and safety of minimally processed food products. Besides the little information on carp sous vide, some microbiological discrepancies have been found in the literature. Meshophiles counts of this study were higher than those reported by Rosnes, [25]. Microbiological safety of two sous vide fish based meals. These authors studied the microbiological quality of sous vide salmon processed at 70 °C for 15 min and storage at 4 and 10 °C. These researchers observed that mesophile counts were below 1 log cfu/g after 42 days of storage at 4 °C and above 6 and 8 log cfu/g after 17 and 42 days of storage at 8 °C respectively [25]. The higher counts found for us in salmon processed at 65 °C/10 min after 45 days of storage at 2 °C (7.49 ± 0.52 cfu/g) could be explained by the lower temperature and time applied. However, even when heat treatments of 90 °C/15 min were applied, we found the same as mesophiles counts after 45 days of storage at 2 °C (4.66 and 4.16 log cfu/g respectively). But, heat treatments of 90 °C/15 and sauced (groups c) applied, we found higer mesophiles counts after 42 days of storage at 2 ºC(4.66 and 2.88 log cfu/g respectively). Since, the heat treatment was more severe, these higher counts could be due to the higher counts in raw fish and to some protective factor such as the different species. Nevertheless, on day 45 the counts were higher in our study than those reported by Rosnes et al. [25]. They found higher mesophiles counts in salmon stored at 4 °C on day 7 (approximately 3 log cfu/g) than in any of the samples of the present study after 14 days of storage at 2 °C. With regard to microbial levels of raw fish, it must be considered that they vary according to water conditions, temperature and handling.

In the present study, the mesophile counts in raw fillets were almost one log unit lower than the counts reported by Gonzalez and Fandos [26]. Since carp is not usually eviscerated at the capture stage, low counts can be found in the raw product depending on the storage and handling conditionsand species diversty. But, mesophile counts Grobantes and Gomez [27] as same. Tokur [28], the determination of the initial freshness quality of carp fingers before frozen storage, aerobic plate count, E. coli, total coliforms, and staphylococcus aureus were analyzed. This study total bacterial count was found to be 2 -8 cfu/g. This value higer than our resuls. The results concur with the Schafaitle [29] where sous vide chicken ballotine, chicken à la king and courgette samples stored at 0–3°C for 21 days had maximum total plate counts of only 8×10^2, 9×10^1 and <20 CFU/g, respectively. In the same study, fish samples had a maximum total count of 4×10^4 CFU/g after two weeks at 0–3°C.

However, other authors have reported higher mesophiles counts. Bergslien [30] observed mesophiles population in sous vide salmon processed at 65 °C for 10 min after 7 days of storage at 2 °C above 5 log cfu/g.

Simpson [31] studied the shelf life of sous vide spaghetti and meat sauce subjected to a heat processing at 65 °C (71 and 105 min) and 75 °C (37 and 40 min). They also observed a gradual increase in total aerobic, anaerobic and lactic acid bacteria counts throughout storage. They found that products stored at 5 °C had a shelf-life of >35 days irrespectively of the processing treatment. However, for products stored at 15 °C, packages were visibly swollen after 14 or 24 days, depending on the severity of the heat

processing treatment. This fact could be explained since minimally processed foods may contain a large proportion of thermally injured cells which are able to undergo repair throughout storage, particularly at temperature abuse conditions and reach levels of public health concern.

Counts of anaerobic bacteria were lower than total aerobic counts. But, Carlin [32], who found similar counts of anaerobic and aerobic bacteria in sous vide vegetables, being most of the anaerobes isolated capable of growing in air. On the other hand, after vacuuming of "sous vide" products, there is usually 1–5% oxygen left in the package at the beginning of the process, this allows facultative anaerobic bacteria to grow. This mechanism explains why clostridia, as obligate anaerobic microorganisms, can only be found after an extend storage period.

The main bacterial groups isolated in raw fish are: Pseudomonas, Moraxella and even Aeromonas [33]. Although the initial counts were significantly lower in the products subjected to a more severe heat treatment, the mesophile and psychrotroph populations increased gradually during the storage, particularly at 10 ºC. The International Commission on Microbiological Specifications for Food [24] recommend that the flesh total aerobic bacteria count should not exceed 10^6/g wet weight. This recommendation was met by our results. This fact could suggest that the micro-organisms were not totally inactivated at the temperatures tested; rather, they were only thermally injured and were capable of recovering throughout storage, particularly under temperature abuse conditions (10ºC) and when subjected to mild treatments, reaching levels of public health concern. Our results are in agreement with those reported by Simpson [31], who studied the evolution of aerobic, anaerobic and lactic acid bacteria in other sous vide products. Simpson [31] also observed a gradual increase in the total aerobic, anaerobic and lactic acid bacteria counts throughout the storage. They concluded that products stored at 5ºC had a shelf-life lower than 35 days irrespectively of the processing treatment. However, for products stored at 15ºC, the packages were visibly swollen after 14 or 24 days, depending on the severity of the heat treatment.

The psychrotrophs counts were lower than the mesophile counts and were only detectable after 56 days of storage except in carp that received a heat treatment. These low psychrotroph counts had also been observed by other authors [25]. Psychrotrophs grow, although slowly at good refrigeration temperatures (5ºC) and had an optimum of about 25ºC.

Our results are in line with those reported by [34], who studied the evolution of the lactic acid bacteria (LAB) in sous vide meat products. These authors also observed that LAB were undetectable immediately after the sous vide treatment in meat products, but that they could recover after storage. However, these authors reported that the lactic acid bacteria growth only occurred sporadically in a few samples. In contrast, Rosnes [25], did not detect viable lactic bacteria or aerobic/anaerobic spores during a storage time of 42 days. Lactic acid bacteria are capable of growing in microaerophilic/anaerobic environments and could be associated with the spoilage of sous vide products, involving the swelling of the packs and/or the development of off-flavors and off-odors [32].

The number of Enterobacteriaceae found in raw fish in this study compared with the results of Gonzalez et [35] almost at the same. Gonzalez and Fandos [26] reported by Enterobacteriaceae end the storage time values, our result lower(2.84-3.01 log/cfug,respectively).

Further research on Clostridium botulinum type E is needed as this micro-organism is capable of growing at low temperature and can survive lower heat treatments. The lowest temperature limit established for the growth and toxin production by strains of psychrotrophic Cl. botulinum is 3.3 ºC [36]. Due to the problems to keep the cold chain and the common temperature abuses during the distribution, retailing and consumption, additional hurdles should be included [37] , [38].

The TBA value is widely used as an indicator of the degree of lipid oxidation. In the present study, the TBA value in the fillets of both b and d groups significantly increased during 10 ºC storage ($p < 0.05$). The increasing of the TBA value during storage has been demonstrated by Gelman and Benjamin [38], for minced pond-bred flesh of silver carp, and by Tokur [39] for fish burgers made from tilapia. The development of the TBA value was very slow in fillets during the 56 days 2 ºC storage. The TBA values were very slowly group c. This situation is caused by the effects of sauce. Bozkurt [40], carp fillets was sauced and cooked. The number of TBA increased very slowly sauce groups. The study resuls was smilar to this study.

TVB-N are most useful indices for spoilage in fresh and lightly preserved seafood [41]. A TVB-N value, of 35 mg N/100 g has been proposed as an upper acceptability limit for spoilage initiation for fresh fish, by the European Commision [42]. The present study, the value of TVB-N was low sauced and heat treatment product and c groups belongs TVB-N value after 42 day 19.2 mg/100g. The heat treatment, sauced, vacuum packaging and cooked at 2 °C did not cause much increase in TVB-N amount. Kaya [43] found TVB-N amount at cured trout and bonito preserved at room temperature to be 48.6 mg/100 g on day 5; the findings for salmon was 45.2 mg/100 g. The same researcher [43] fund out that if preserved in refrigerator for 50 days, trout produced 58.4 mg/100 g, bonito produced 57.6 mg/100 g and salmon produced 55.8 mg/100 g TVB-N. This results were higer than our study storage 10 ºC. Chouliara [44], reported that TVB-N amount of cured and vacuum-packaged Atlantic salmons which were preserved at +4 °C for 2-3 weeks was found as 22.4 mg/100 g.

It can be concluded that, in this study, the heat treatment and sauced were the most effective one to extend the shelf-life of carp. The storage temperature plays a key role to ensure the quality and safety of sous vide products, together with the heat treatment. Temperature abuses at 10ºC decrease the shelf-life.

REFERENCES

[1] G. E. Karmas and R.S. Harris, "Nutritional Evaluation of Food Processing" 3 rd ed. V. N. Reinhold, Ed. New York: 1987, pp. 37-53.
[2] P. G. Creed, "Sensory and nutritional aspects for sous vide processed foods" in Sous vide and cook-chill processing for food industry, S. Ghazala, Ed. Gaithersburg: 1998, pp. 57–88.
[3] D. L. Hrdina-Dubsky,"Sous vide finds its niche," Food Engineering International, vol.14, pp. 40-48, 1989.
[4] Y. Özoğul, F. Özoğul, E. Kuley," Effects of combining of smoking and marinating on the shelf life of anchovy stored at 4 ºC," Food Science and Biotechnology, vol.19, no.1, pp.69-75, 2010.
[5] M. Schellekens, T. Martens," Sous Vide State of the Art" in Conf Rec N1 EUR 15018 EN 1992 Commission of the European Communities Directorate General XII, Research and Development.
[6] C. L. Hatherway, "Clostridium botulinum and other clostridia that produce botulinum neurotoxin,". in Clostridium botulinum: Ecology

and control in foods, A. Hauschild and K. L. Dodds, Ed. New York, 1992, pp.8.
[7] E. Borch, M. L. Kant-Muermans, Y. Blixt, "Bacterial spoilage of meat and cured meat products," International Journal of Food Microbiology, vol.33, no.1, pp. 103–120,1996.
[8] H. Korkeala, K. J. BjÎrkroth, "Spoilage and contamination of vacuumpackaged cooked sausages: A review," Journal of Food Protection, vol.60, pp.724–731,1997.
[9] A. F. Egan, "Lactic acid bacteria of meat and meat products," Antony van Leeuwenhoek, vol.49, pp.327–336, 1983.
[10] P. Díaz, G. Nieto, M.D. Garrido, S. Bañón, "Microbial, physical–chemical and sensory spoilage during the refrigerated storage of cooked pork loin processed by the sous vide method," Meat Science, vol.80, pp.287–292, 2008.
[11] H. Nyati, "An evaluation of the effect of storage and processing temperature on the microbiological status of sous vide extended shelf-life products," Food Control, vol.11, pp.471–476, 2004.
[12] S. H. Wang, M. H. Chang, T. C. Chen, "Shelf-life and microbiological profiler of chicken wing products following sous vide treatment," International Journal of Poultry Science, vol. 3, no.5,pp. 326–332, 2004.
[13] ACMSF, "Report on Vacuum Packaging and Associated Processes," Advisory Committee on the Microbiological Safety of Food (ACMSF), HMSO, London,1992
[14] C. A. Genigeorgis, " Additional hurdless for sous vide products," In: First European Symposium on Sous-vide Proceedings, Leuven, Belgium, pp.57, 1993.
[15] ICMSF, "Microorganisms in Foods, Microbial Specifications of Commodities," International Commission on Microbiological Specifications for Foods (ICMSF) Blackie Academic & Professional, London, vol.6,1986.
[16] J. C. Man, M. Ragosa, M. Sharpe, "A medium for the cultivation of lactobacilli," J. Appl. Bacteriol., vol.23, pp.130–135, 1960.
[17] D. A. A. Mossel, J. E. L. Corry, C. B. Struijk, R. M. Baird, Essentials of the microbiology of foods, A Textbook for Advanced Studies, Wiley, Chichester, England, 1995.
[18] A. H. Varnam, M. G. Evans, Foodborne Pathogens, Manson Publishing, London,1996.
[19] C. Varlık, M. Uğur, N. Gökoğlu, H. Gün, Su Ürünlerinde Kalite Kontrol İlke ve Yöntemleri. Gıda Teknoloji Derneği, Ankara, 1993.
[20] B. G. Tarladgis, B. M. Watts, M.S. Younathan, L. Dugan, "A Distilation Method For The Quantitative Determination Of Malonaldehyde İn Rancid Foods," J. American Oil Chemical., vol.37, pp. 44-48, 1960.
[21] ISO 8586-1,Sensory analysis, General Guidance for Selection, Training and Monitoring of Assesor, Switherland, 1993.
[22] Ü. Kurtcan, M. Gönül M, "Gıdaların Duyusal Değerlendirilmesinde Puanlama (Scoring) Metodu," Ege Üniversitesi Mühendislik Fakültesi Dergisi, vol. 5, pp. 137-146, 1987.
[23] Anonim, Statistical Analysis System (Version 6.1). SAS Institute Inc., Cary, North Carolina. USD, 1996.
[24] ICMSF, "Microorganisms in Foods, Microbial Specifications of Commodities," International Commission on Microbiological Specifications for Foods (ICMSF) Blackie Academic & Professional, London, vol.3, 1998.
[25] J. T. Rosnes, H. Kleiberg, H. Bergslein, J. Vidvei, "Microbiological safety of two sous vide fish based meals," In: Third European Symposium on Sous-vide Proceedings, Leuven, Belgium, pp. 195–204, 1999.
[26] E. Gonzalez-Fandos, M. C. García-Linares, A. Villarino-Rodríguez, M. T. García-Arias, M. C. Garcia-Fernández, "Evaluation of the micrbiological safety and sensory quality of rainbow trout (Oncorhynchus mykiss) processed by the sous vide method," Food Microbiology, vol. 21, pp.193–201, 2004.
[27] I. Grobantes, R. Glomez, "Envasado de filetes de trucha acuicultuvada bajo atmosfera modificada," Aliment. Equipos Tecnol., vol.5, pp.115–121, 1999.
[28] B. Tokur, S. Ozkütük, E. Atici, G. Ozyurt, C. E. Ozyurt, "Chemical and sensory quality changes of fish fingers, made from mirror carp (Cyprinus carpio L., 1758), during frozen storage (-18 C)," Food Chemistry, vol. 99, pp.335-341, 2006.
[29] J. M. Schafheitle, N. D. Light, "Sous vide preparation and chilled storage of chicken ballotine," International Journal of Food Science and Technology, vol.24, pp.199–205, 1989.
[30] H. Bergslien, "Sous vide treatment of salmon (Salmon solar)," In: Second European Symposium on Sous Vide Proceedings, Leuven, Belgium, pp. 281–291,1996.
[31] M. V. Simpson, J. P. Smith, B. K. Simpson, H. Ramaswamy, K. L. Dodds, "Storage studies on a sous vide spaghetti and meat sauce product," Food Microbiol., vol.11, pp.5–14, 1994.
[32] F. Zoellin, M. H. Guinebretiere, C. Choma, P. Schmitt, C. Nguyen, "A fair collaborative programme: research on factors allowing a risk assessment of spore-forming pathogenic bacteria in cooked chilled foods containing vegetables," In: Third European Symposium on Sous-vide Proceedings, Leuven, Belgium, pp. 53–70, 1999.
[33] J. A. Sousa, J. L. Romalde, A. Ledo, J. C. Eiras, J. L. Barja, A. E. Toranzo, "Health status of two salmonid aquaculture facilities in North Portugal: characterization of the bacterial and viral pathogens causing notificable diseases," J. Fish Dis., vol.19, pp.83–89, 1996.
[34] M. E. Guerzoni, A. Gianotti, C. C. Lopez, "Effect of some process variables on safety and shelf-life of ''sous vide'' cooked foods," In: Third European Symposium on Sous-vide Proceedings, Leuven, Belgium, pp. 253–266, 1999.
[35] C. Gonzalez, T. Lopez, M. L. Garcıa, M. Prieto, A. Otero, "Bacterial microflora of wild brown trout (Salmon trutta), wild pike (Esox lucius) and aquacultured rainbow trout (Oncorhynchus mykiss)," J. Food Prot., vol. 62, pp.1270–1277, 1999.
[36] C. F. Schmidt, R. V. Lechowvich, J. F. Folinazzo, "Growth and toxin production by type E Clostridium botulinum below 40ºF," J. Food Sci., vol. 26, pp.626–630, 1961.
[37] A. Tolstoy, "Practical monitoring of the chill chain," Int. J. Food Microbiol., vol. 13, pp.225–230, 1991.
[38] A. Gelman, E. Benjamin, "Characteristics of mince from pond-bred silver carp (Hypophthalmichthys molitrix) and preliminary experiments on its use in sausages," Journal of the Science of Food and Agriculture, vol. 47, pp.225–241, 1988.
[39] B. Tokur, A. Polat, G. Beklevik, S. Özkütük, "Changes in the quality of fishburger produced from Tilapia (Oreochromis niloticus) during frozen storage (-18 °C)," European Food Research and Technology, vol.218, pp.420-423, 2004.
[40] Ö. P. Bozkurt, "Soslu aynalı sazan balıklarının raf ömrünün belirlenmesi," Phd Thesis, University of Fırat, 2006.
[41] P. Dalgaard, "Fresh and lightly preserved seafood," in Shelf-life evaluation of foods,2 nd ed. C.M.D. Man and A.A. Jones, Ed. Gaithersburg: MD, USA, 2000, pp. 110–139.
[42] Commission Decision 95/149/EC, (1995). Commission Decision 95/149/EC. (1995). Commission Decision of 8 March, 1995 fixing the total volatile basic nitrogen (TVB-N) limit values for certain categories of fishery products and specifying the analysis methods to be used. Official Journal of the European Communities, L 097, 29/04/1995. pp. 0084–0087.
[43] Y. Kaya, "Balık Dumanlama Teknolojisinde Çeşitli Faktörlerin Kalite ve Dayanma Sürelerine Etkileri," Yüksek Lisans Tezi. 19 Mayıs Üniversitesi, 1994.
[44] I. Chouliara, I. N. Savvaidis, N. Panagiotakis, M. G. Kontominas, "Preservation of salted, vacuum-packaged, refrigerated sea bream (Sparus aurata) fillets by irradiation: microbiological, chemical and sensory attributes," Food Microbiology, vol. 21, pp. 351–359, 2004.