Quality assessment of fish burgers from deep flounder (*Pseudorhombus elevatus*) and brushtooth lizardfish (*Saurida undosquamis*) during storage at -18ºC

Mahmoudzadeh M. [1]; Motallebi A.A.[2]; Hosseini H.[1]; Haratian P.[1]; Ahmadi H.[1]; Mohammadi M.[1] and Khaksar R.[1*]

Abstract

Microbiological, chemical and sensory changes of fish burgers prepared from deep flounder (*Pseudorhombus elevatus*) and brushtooth lizardfish (*Saurida undosquamis*) were determined during storage at -18ºC for 5 months. Microbiological counts were including total plate count (TPC), total coliform (TC), *Staphylococcus aureus*, Psychotropic and *Escherichia coli* decreased throughout the frozen storage. Reduction of microbial load in brushtooth lizardfish was higher than that in deep flounder, except for *Staphylococcus aureus* counts that was almost equal in both groups. There was a significant increase in pH value in both groups ($P<0.05$) in first and second months of storage only. Moisture content increased in both groups at the end of 5th month, with increase of moisture in deep flounder fish burgers being higher than that in brushtooth lizardfish burgers. TVB-N values in both groups increased significantly ($P<0.05$ and $P<0.008$ for deep flounder and brushtooth lizardfish burgers, respectively) at the end of the second month, however, there was a decrease or no significant change afterward. TBA value of deep flounder fish burgers had a significant decrease ($P<0.05$) as storage time continued, however, it increased significantly in brushtooth lizardfish burgers at the end of second month ($P<0.006$) following by a decrease at the end of storage period. Peroxide value (PV) in both groups increased significantly ($P<0.05$ and $P<0.002$ in deep flounder and brushtooth lizardfish burgers, respectively) during storage time but a significant decrease was observed at the end of third and fourth months ($P<0.005$ and $P<0.001$ in deep flounder and brushtooth lizardfish burgers, respectively). Sensory parameters (color, texture, taste and general acceptability) for two groups decreased significantly ($P<0.003$ for all parameters in 2 groups) during storage with deep flounder fish burgers receiving higher scores than brushtooth lizardfish burgers at the beginning and end of the storage period.

Keywords: *Pseudorhombus elevatus*, *Saurida undosquamis*, Fish burger, Frozen storage, Quality

1- National Nutrition and Food Technology Research Institute, Faculty of Nutrition Science and Food Technology, Shahid Beheshti University, P.O.Box: 19395-4741 Tehran, Iran
2- Iranian Fisheries Research Organization, P.O.Box: 14155-6116 Tehran, Iran
* Corresponding author's e-mail: r.khaksar@sbmu.ac.ir

Introduction

Fish and fishery products contain high quality protein and other necessary nutrients; they are low in saturated fatty acids and contain high content of unsaturated fatty acids (Venugopal & Shahidi, 1995). Fish meal could be a major protein source in food diets. There are essential amino and fatty acids that are present in fish meal but not present in tissue from plants or animals (Francis-Floyd., 2002).

A well-balanced regime that includes a variety of fish and fish products can contribute to heart health and children's proper growth (Hui, 2007). In recent years, the increases of civilization have led to consumer's preference to ready-to-eat foods (Cakli *et al*., 2005). Fish burgers are one of the secondary minced fish based products and are popularly distributed frozen for longer shelf life (Jamilah, 2004). During frozen storage fish quality may be declined as a result of several factors. Bacterial growth is the main cause of fish spoilage so it is logical to use bacteria counts as an index of quality for frozen storage (Suvanich *et al*., 2000). However microbiological analyses are not extensively used for rapidly determining commercial quality (Ruiz-Capillas & Moral, 2001).

Oxidation of highly unsaturated lipids is other factor which highly related to the production of off-flavors and odors (Aubourg *et al*., 2002) and also as influencing, protein denaturation and texture changes (Orak & Kayisoglu, 2008).

Deep flounder (*Pseudorhombus elevatus*) and brushtooth lizardfish (*Saurida undosquamis*) are marine fish species which are distributed in Persian Gulf and Oman

Sea. They are important resources in the Far East and annually tons are eaten mainly in the forms of fish cakes and fish burgers (Nielsen, 1984; Van der elst, 1993). These fishes are not sold commercially in Iran and usually are part of by-catch products. use of less valuable fishes is a serious contrain in Iranian fisheries (Zareh Gashti, 2002).

Brushtooth lizardfish has average length about 20cm with undesirable figure also color and figure of deep flounder is not favorable for marketing as whole fish. So use of these fish for production of value added products could be benefit economically. The aim of the present study was to produce fish burgers from deep flounder and brushtooth lizardfish and to evaluate chemical, microbial and sensory attributes during storage at -18ºC for 5 months.

Materials and methods

Deep flounders and brushtooth lizardfish were caught in June 2008 from Persian Gulf. About 8kg of each fish was used for production of fish burger samples. The fish samples were frozen at -18ºC and transported to the Marine Industrial Unit (Nazarabad, Tehran).

Produced fish burgers were prepared from unthawed fishes. Frozen fish were deheaded, gutted and deboned with a Lima model deboner using 0.1mm diameter holes (France). Prepared minced meat was ground to form paste using a CFS model meat grinder with 5mm pore sizes (Netherlands).

Formulation of fish burgers were composed of 80% fish meat and 20% other ingredients including: 5% crumble powder,

3.45% onion, 3% starch, 3% gluten, 1.2% salt, 1% garlic, 1% isolated soy protein, 1% sodium caseinate, 0.5% lime juice, 0.5% blend spice (white pepper, red pepper, thyme, cumin), 0.3% sodium tripoly-phosphate, 0.05% sodium ascorbate. Further stages were contained of forming into fish burgers and coating with batter and breading materials. These materials were purchased from N.P.O international company (Taiwan). The batter was put into a blender with a cold water/batter flour ratio of 1.1:1 (w/w) then it was covered with breading crumbs, this instruction was according to guidelines of N.P.O international company. After coating process, they were pre-fried at 180ºC for 30s in the blend of palm and cotton seed oil with a ratio of 2:1, and then were quick frozen individually at -40ºC for 45 minutes (CFS model, Netherlands). After frozen stage, thickness and diameter of produced fish burgers were about 0.5 and 10cm, respectively. Produced fish burgers were put in bags manually then packaged automatically in bags from PVC/PE polymers and stored at -18ºC for 5 months. The polymers for packaging were purchased from Delta Company (Turkey). Weight of each fish burger was less than 100g, and every 10 pieces were put in one package.

Analyses for the determination of the proximate composition (crude protein, total lipid, crude ash and moisture) for fish paste and fish burgers were performed on the production day. Fish burgers were analyzed for microbiological, chemical and sensory attributes periodically at the end of every month. Proximate composition, chemical analysis and microbiological counts were carried out triplicate. Thus for each analysis (chemical or microbial) two packages of fish burgers were taken out randomly from frozen storage and stored at 4ºC over night. About 20 burgers of each treatment were used for chemical or microbial analyses. To determine chemical and microbiological attributes, coating materials were removed.

Proximate composition of fish pastes and fish burgers were determined as crude protein by the Kjeldahl method AOAC (2005), total lipids ISO 1443 (1973) moisture content ISO 1442 (1997) and crude ash content ISO 936 (1998).

For all microbiological counts, 10g of sample were transferred in 90ml distilled water 9/1000 (9g NaCl/1000ml distilled water) (ICMSF, 1978). From the 1/10 dilution other decimal dilutions were prepared. Total plate count (TPC) was determined by using pour plate method and Plate Count Agar as medium according to ISO 8443 (2003). For total coliform (TC) pour plate procedure and Violet Red bile Agar medium were used according to ISO 4832 (1991). *Staphylococcus aureus* and Psychrotrophic were determined by surface plate method using Baird Parker Agar and Total Plate Count Agar mediums according to ICMSF (1978) and ISO 17410 (2001), respectively. For *Escherichia coli*, Most Probable Number method and Lauryl Sulfate broth as medium was used. Confirmation test was made in Pepton Water and EC broth according to ISO 7251 (2005).

pH value was measured by using a Metrohm model pH meter (Switzerland).

Samples were homogenized in distilled water in ratio of 1:1 (w/v) according to AOAC (2005). Thiobarbituric acid value (TBA, mg malonaldehyde/kg) was determined by a distillation method, according to this method 5ml TBA reagent (0.2883g/100ml of 90% glacial acetic acid) were added to 5ml distillate that is collected from distillation 10g of fish burger with 4M hydrochloric acid. After that, it was shaken and heated in boiling water for 35 min. A blank was prepared using 5ml water with 5ml reagent. Then were cooled in water for 10 min and measured the absorbance against the blank at 538nm using 1cm cell (Pearson, 1981).

TBA number (mg malonaldehyde per kg sample) = 7.8 D

Total volatile base nitrogen (TVB-N, mg N/100g) was determined according to Safari and Yosefian (2006). For measuring peroxide value (PV), fish oil was extracted by shaking 100g of samples with chloroform. After 2 hours of automatic shaking, samples and chloroform blends were filtered then chloroform were removed from clarified by using a rotary evaporator(Buchi, Switzerland) (Parvane, 1998). PV of obtained oil was determined according to AOAC (2005).

Sensory evaluation of group A and group B was assessed by 11 experienced panelists. Fish burgers were deep fried with frying oil until they were cooked. Panelists scored for color, texture, taste and general acceptability parameters using an 8 point hedonic scale according to Khalil (2000) (1, dislike extremely to 8, like extremely).

For statistical analysis, independent sample t-test and Mann-Whitney U using the SPSS 11.5 for windows were performed to determine the differences between two groups of chemical and sensory data, respectively. Paired sample t-test and Wilcoxon were used to find significant differences between storage periods.

Results

The contents of crud protein, total lipid, moisture and crude ash of fish pastes and fish burgers from deep flounder (group A) and brushtooth lizardfish (group B) were shown in Table 1. According to statistical findings, there were significant (P<0.05) differences in the crude protein of both fish pastes but no significant (P>0.05) differences was found in the fish burgers prepared from them. Total lipid and moisture contents were significantly (P<0.05) different between both fish burger groups, whereas no significant (P>0.05) difference was found between fish paste groups. Similarly, there were no significant (P>0.05) difference between crude ash content in group A and group B fish burgers and fish pastes but crude ash content between fish pastes and fish burgers were significantly different (P<0.05).

Microbiological analysis of fish pastes at production day was demonstrated in Table 2. Microbial load of fish paste from brushtooth lizardfish (B) was lower than the values of fish paste from deep flounder (A), so because of high microbial load especially in TPC and *Staphylococcus aureus* counts, quality of deep flounder paste was lower than that in brushtooth lizardfish paste.

Table 1: Chemical composition of the fish paste and fish burgers

Proximate Composition (%)	Fish paste (A)	Fish paste (B)	Deep flounder fish burger (Group A)	Brushtooth lizard fish burger (Group B)
Crude protein	19.48±0.18 [a]	21.08±0.37 [b]	19.01±0.23 [ac]	18.69±0.19 [c]
Total lipid	2.13±0.18 [a]	1.62±0.18 [a]	6.73±0.33 [b]	5.45±0.11 [c]
Moisture	77.23±0.04 [a]	77.09±0.35 [a]	65.58±0.46 [b]	67.55±0.39 [c]
Crude ash	1.37±0.1 [a]	1.40±0.08 [a]	2.71±0.07 [b]	2.87±0.06 [b]

Within the same row having different superscripts are significantly different at P<0.05.

Table 2: Total plate count (TPC), total coliform (TC), *Staphylococcus aureus*, psychrotrophic and *Escherichia coli* counts for fish paste.

Analysis	TPC (log cfu/g)	TC (log cfu/g)	Psychrotrophic (log cfu/g)	*Staphylococcus aureus* (log cfu/g)	*Escherichia coli* (MPN/g)
Fish paste (A)	6.30±0.21	4.70±0.00	4.78±0.16	3.30±0.13	10±0.00
Fish paste (B)	5.59±0.13	4.57±0.07	4.59±0.23	2.74±0.00	5±0.00

Data are expressed as means± standard deviation (n=3).

Table 3 shows TPC, TC, *Staphylococcus aureus*, psychrotrophic and *Escherichia coli* counts during 5 months frozen storage. TPC and psychrotrophic counts of group B were higher than those in group A. The microbial count in fish burgers was found lower than fish paste due to using additive ingredients in this product. TPC count of group A highly reduced rather than its fish paste (6.30log cfu/g to 5log cfu/g) whereas this reduction was very low for Group B (5.59 log cfu/g to 5.48 log cfu/g). Decrease of microbiological count was related to the freezing time and highest decrease was found during the first month of storage for both groups. All microbiological counts were decreased by freezing at -18ºC but group A had higher TPC, TC and sychrotrophic counts at the end of the storage period.

The changes in pH, moisture, TVB-N and TBA values during 5 months frozen storage are shown in Table 4. group A had lower pH value rather than group B at the beginning and end of storage period. The pH value of group A significantly (P<0.05) increased at the end of second month then decreased at the end of frozen storage but in group B increased significantly at the end of second month afterwards no significantly (P>0.05) different was observed at the end of storage period (Table 4).

Table 3: Microbiological counts of fish burgers during storage at -18ºC

Analysis	Group	Storage time (month)					
		0 (control)	1	2	3	4	5
TPC (log cfu/g)	A	5.00±0.00	3.79±0.06	3.78±0.11	3.73±0.05	3.64±0.22	3.32±0.09
	B	5.48 ±0.04	2.63±0.02	2.60±0.1	2.70±0.00	2.74±0.07	2.26±0.03
TC (log cfu/g)	A	2.60±0.00	2.40±0.03	2.52±0.03	2.40±0.02	2.40±0.04	2.52±0.03
	B	2.60±0.01	2.10±0.13	2.00±0.00	2.20±0.01	2.11±0.12	2.20±0.00
Psychrotrophic (log cfu/g)	A	3.99±0.05	2.73±0.06	2.73±0.08	2.38±0.1	2.30±0.00	2.00±0.00
	B	4.30±0.24	3.00±0.00	3.00±0.14	2.90±0.09	3.08±0.05	1.30±0.03
Staphylococcus aureus (log cfu/g)	A	3.00±0.01	2.52±0.05	2.00±0.00	2.70±0.07	2.48±0.03	< 1.00
	B	2.41±0.09	2.23±0.07	1.95±0.03	1.95±0.06	2.00±0.00	< 1.00
Escherichia coli (MPN/g)	A	1.5±0.16	0.93±0.03	<0.3	<0.3	<0.3	<0.3
	B	1.1±0.05	<0.3	<0.3	<0.3	<0.3	<0.3

Data are expressed as means± standard deviation (n=3).

Produced fish burgers were lost moisture content during production process from fish pastes; nevertheless group B had higher moisture content than group A at the beginning and end of the storage. This factor significantly (P<0.05) increased in both of the groups during frozen storage (Table 4).

The TVB-N values of group A and group B were not different significantly (P<0.36) at the beginning of storage time whereas there were significant differences between two groups at the end of this period. TVB-N value of both group A and group B significantly (P<0.006 and P<0.001 for group A and group B, respectively) increased from 11.66 to 20.97 and 10.68 to 20.97 at the end of second and first months, respectively, but then significantly decreased in group A at the end of storage

whereas in group B no significant (P>0.05) changes were observed between end of the first month and end of the frozen storage.

The TBA values of both groups significantly decreased from 1.01 to 0.22 and 0.70 to 0.26 at the end of storage, respectively and there were no significant differences between values of two groups at the end of 5th month (P>0.05) (Table 4).

Peroxide values (PV) of both of the groups significantly (P=0.004 and P=0.002 for group A and B, respectively) increased at the end of second month then decreased at the end of 4th month. PV suddenly increased at end of 5th month and there were highly significant differences (P=0) between values at the end of 4th and 5th months. PV values of group A were higher

and significantly (P<0.05) different at the beginning and the end of the storage period.

To determine the sensory quality of fish burgers, color, texture, taste and general acceptability parameters were measured (Table 5). In both of the groups all of these parameters decreased significantly (P<0.05) during storage but group A was received significantly higher scores rather than group B during all of the storage periods except for taste and texture at the beginning and the end of the second month, respectively.

Table 4: Chemical analysis parameters during frozen storage (-18ºC)

Analysis	Group	Storage time (month)					
		0 (control)	**1**	**2**	**3**	**4**	**5**
pH	A	$6.53\pm0.05^{a*}$	6.78 ± 0.02^{b}	7.03 ± 0.05^{c}	6.93 ± 0.02^{c}	$6.80\pm0.00^{b*}$	$6.83\pm0.02^{bc*}$
	B	6.70 ± 0.00^{a}	6.88 ± 0.07^{ab}	7.00 ± 0.00^{b}	7.00 ± 0.00^{b}	7.03 ± 0.02^{b}	7.00 ± 0.00^{b}
Moisture (g/100g)	A	$65.58\pm0.46^{ab*}$	66.74 ± 0.27^{ac}	$65.86\pm0.14^{ab*}$	66.23 ± 0.18^{bd}	$66.92\pm0.1^{c*}$	$66.86\pm0.25^{dc*}$
	B	$67.55\pm0.0.39^{a}$	67.52 ± 0.44^{ab}	67.92 ± 0.04^{ab}	67.07 ± 0.85^{ab}	67.90 ± 0.26^{ab}	67.93 ± 0.28^{b}
TVB-N (mg N/100g)	A	11.66 ± 0.98^{a}	$16.88\pm0.56^{bd*}$	$20.97\pm0.49^{c*}$	$15.25\pm0.56^{b*}$	$17.54\pm0.49^{d*}$	$14.60\pm0.49^{b*}$
	B	10.68 ± 0.00^{a}	20.97 ± 0.49^{b}	17.86 ± 1.13^{c}	22.44 ± 0.00^{d}	19.99 ± 0.00^{bc}	19.50 ± 0.49^{bc}
TBA(mg malonaldehyde/kg)	A	$1.01\pm0.00^{a*}$	0.68 ± 0.00^{b}	$0.72\pm0.02^{b*}$	$0.35\pm0.00^{c*}$	$0.27\pm0.00^{de*}$	0.22 ± 0.02^{e}
	B	0.70 ± 0.01^{ac}	0.64 ± 0.05^{a}	0.95 ± 0.03^{b}	0.71 ± 0.04^{c}	0.33 ± 0.03^{d}	0.26 ± 0.01^{d}
Peroxide (meq O2/kg)	A	$1.27\pm0.03^{a*}$	$2.67\pm0.12^{b*}$	$3.06\pm0.12^{c*}$	1.24 ± 0.03^{a}	0.70 ± 0.02^{d}	$4.10\pm0.07^{e*}$
	B	0.35 ± 0.03^{a}	0.65 ± 0.01^{b}	2.56 ± 0.25^{c}	1.20 ± 0.05^{d}	0.56 ± 0.09^{ab}	3.15 ± 0.14^{c}

- **Data are expressed as means± standard deviation (n=3).**
- **Means within the same column shown in * are satisfactory different at p<0.05.**
- **Means within the same row having different superscripts are significantly different at p<0.05.**

Table 5: Changes in sensory attributes of fish burgers during storage at -18ºC.

Analysis	Group	Storage time (month)					
		0 (control)	**1**	**2**	**3**	**4**	**5**
Color	A	$7.18\pm0.40^{a*}$	$7.54\pm0.52^{b*}$	$5.27\pm0.64^{c*}$	$5.18\pm0.6^{c*}$	$4.27\pm0.78^{d*}$	$3.18\pm0.87^{e*}$
	B	5.90 ± 1.04^{a}	5.54 ± 1.03^{a}	4.00 ± 1.09^{b}	3.90 ± 1.04^{b}	2.27 ± 0.90^{c}	1.36 ± 0.50^{d}
Texture	A	$7.72\pm0.46^{a*}$	$7.63\pm0.50^{a*}$	6.18 ± 0.98^{b}	$6.27\pm1.00^{b*}$	$4.09\pm0.70^{c*}$	$3.27\pm0.64^{d*}$
	B	6.27 ± 1.42^{a}	5.27 ± 1.42^{ab}	5.27 ± 1.42^{ab}	5.18 ± 0.75^{b}	2.09 ± 0.30^{c}	1.54 ± 0.52^{d}
Taste	A	7.27 ± 0.90^{a}	$7.63\pm0.50^{a*}$	$5.54\pm0.68^{b*}$	$5.45\pm0.68^{b*}$	$4.09\pm0.30^{c*}$	$3.18\pm0.75^{d*}$
	B	7.27 ± 1.00^{a}	6.18 ± 1.53^{a}	4.45 ± 0.82^{b}	4.45 ± 0.82^{b}	2.09 ± 0.70^{c}	1.36 ± 0.50^{d}
General Acceptability	A	$7.72\pm0.46^{a*}$	$8.00\pm0.00^{a*}$	$5.63\pm0.92^{b*}$	$5.54\pm0.93^{b*}$	$3.72\pm0.64^{c*}$	$3.00\pm0.77^{d*}$
	B	5.90 ± 0.70^{a}	5.81 ± 0.75^{a}	4.45 ± 0.93^{b}	4.27 ± 1.00^{b}	2.18 ± 0.60^{c}	1.54 ± 0.52^{d}

- **Data are expressed as means± standard deviation (n=11).**
- **Means within the same column shown in * are satisfactory different (P<0.05).**
- **Means within the same row having different superscripts are significantly (P<0.05) different.**

Discussion

Produced fish burgers had higher total lipid content than related fish pastes, because of frying stage in production process. Ravindranathan *et al*. (1982) indicated that peroxide and TBA values were affected by the lipid content in fish meat. Protein and lipid oxidation can account for the toughened texture, off-flavor and unappealing odor of frozen stored sea foods (Thanonkaew *et al*., 2006). Group A and group B had higher ash content than fish paste due to addition of ingredients to the fish burger formulation.

TPC analyses of fish pastes and fish burgers did not exceed the maximum limits (7logcfu/g) of microbiological criteria for fresh and frozen fish given by the ICMSF (1978). TPC is considered a quality indicator for food samples; there is not direct correlation between this and the presence of pathogenic microorganisms (Arvanitoyannis *et al*., 2005).

TPC is an indicator of the shelf-life of products, and also the potential for growth of the microorganism that is present (Arvanitoyannis *et al*., 2005). Coliform counts should be present in numbers less than 100 per gram of meat (Nollet, 2007), so TC of group A was not between recommended levels above during all storage time, however, maximum level of TC in fish products was given as 400cfu/g by ICMSF (1978).

Escherichia coli is one of the microbiological hazards occurring in fish. Confirmation tests of this bacterium in both groups were negative after 2 months of frozen storage so according to MPN procedure was reported as MPN/g <0.3.

Staphylococcus aureus was eliminated in both groups with an equal amount during first month of storage. The presence of *Staphylococcus aureus* in foods is often resulting of improper handling by personnel, who are frequently contaminated with this microorganism (Gundogan *et al*., 2005).

Viability of psychrotrophic bacteria depends on several factors such as aerobic and anaerobic conditions or level of existent salt (Lawrie & Ledward, 2006) so probably because of variable conditions in both groups; psychrotrophic bacteria count in group B was higher than those in group A during storage time. But the decrease in group B during last month was higher than those in group A. This result may be attributed to high lipid content and other compounds of group A that had maintenance effect on death rate of these bacteria. Psychrotrophic bacteria are very important among different bacteria causing spoilage, because they are mostly related to the changes in sensory attributes such as odor, texture and flavor and could produce different metabolic compounds such as ketones, aldehydes, volatile sulfides and biogenic amines (Safari & Yosefian, 2006). A proposed limit of psychrotrophic bacteria is 103 to 104cfu/g which is consistent with other studies (Pons-Sanchez-Cascado *et al*., 2006).

According to Grigorakis *et al*. (2003) post mortem pH can vary from 5.4–7.2, depending on fish species. Several authors have reported different results about decreasing or increasing of pH in various fish species (Orak & Kayisoglu, 2008). The highest pH value in both groups was 7.03 that group A and group B were obtained this value after 2 and 4 months, respectively. pH value between 6.8-7 were proposed as acceptance limit of fish, and values above 7 were considered to be spoiled (Kose *et al*., 2006, Ozyurt *et al*., 2007). However pH value is not a suitable index and it can be useful as a guideline for quality control of fish (Ruiz-Capillas & Moral, 2001). The increase in the pH value in both groups during initial months of storage is caused by the enzymatic degradation of the fish muscle content (Vareltzis *et al*., 1997) whereas decrease or constant levels of pH might be attributed to increasing solubility of CO_2 at last months, effecting on aerobic microflora growth (Adams & Moss, 2000).

Moisture content is one of the factors affecting microbial growth in foods (Adams & Moss, 2000). Also moisture content is one of the factors that will be affected in functional differentiation of muscles (Lawrie & Ledward, 2006). According to Biswas *et al*. (2004) deceased textural scores during storage might be related to release of moisture. Increasing of moisture content in group A was higher than those in group B during storage time.

Total volatile basic nitrogen (TVB-N) that is mainly composed of ammonia,

trimethylamine (TMA) and dimethylamine (DMA), is widely used as an indicator of meat deterioration (Fan *et al*., 2008). TVB-N value is not stable during frozen storage and could be changed according to species, processing methods, and storage temperature (Tokur *et al*., 2006). The results of Castro *et al*. (2006) study in European sea bass indicated that prevention of false negative interpretations with the TVB-N method beyond the normal shelf life for this fish type is important. The increasing of TVB-N value during storage is related to bacterial spoilage and activity of endogenous enzymes (Chomnawang *et al*., 2007). Irregular changes on TVB-N values in the study of Kilinc *et al*. (2003) during the storage period of Sardine (*Sardina pilchardus*) were due to the elimination of dissolved volatile constituents through drip. Leaching out phenomena of volatile bases could be caused decreasing of TVB-N values if samples were stored in loose closed bags (Özogul & Özogul, 2000) also decreasing of TVB-N value may be result of hypothesis that mentioned for reduction of pH value. We believe, at least one of the above is responsible for the reduction of the TVB-N values.

Several lipid oxidation indices were assessed to follow up the development of oxidation in frozen state. Peroxide value and thiobarbituric acid reactive substances showed primary and secondary oxidation, respectively (Losada *et al*., 2007). TBA test is based on the reaction of malonaldehyde with TBA reagent to obtain a red pigment,

which results from the condensation of two molecules of TBA with one molecule of malonaldehyde and the probable elimination of two molecules of water (Fernandez *et al*., 1997). TBA test should not be considered as general reference of rancid odor, because factors such as specie, dietary status, age and raw or cooked meats could be influenced the numbers of TBA (Fernandez *et al*., 1997). The results of Nakayama and Yamamoto (1977) study on TBA values in Dogfish, Pollock and Grey cod during frozen storage showed that increasing of TBA values at the surface of blocks were different between species, also this study showed that there were no clear relationship between TBA values and taste panel results. Kolakowska and Deutry (2006) found that in the presence of interfering substances the TBA test should be considered as a "freshness test" rather than a strictly rancidity one. High lipid content of deep flounderfish paste could be accelerated formation of TBA substances in group A fish burgers so group A had higher TBA value at the beginning of period but then decreased gradually. The decrease of TBA values in both groups could be attributed to the interaction of decomposition products of protein with malonaldehyde to give tertiary products (Hernández-Herrero *et al*., 1999).

Lipids in cooked meat could be more easily oxidized than those in raw meat (Widayaka *et al*., 2001). The PV of two groups increased during two months of frozen storage, indicating an increase in lipid oxidation, and then it decreased the end of 4th month. Decrease of PV could be attributed to different rates of lipid oxidation (Arvanitoyannis *et al*., 2005) or predominance of lactic acid bacteria. Several genera of lactics including lactobacilli, lactococci and pediococci have been identified as producing hydrogen peroxide (Vandenbergh, 1993). Lipid oxidation compounds may be reacted with nucleophilic biological constituents (proteins, peptides, free amino acids) and caused the formation of interaction compounds (Aubourg, 1999) so measuring them are always not useful.

Group A was received higher scores at the beginning of period in terms of color, texture and general acceptability that show fish burgers prepared from brushtooth lizardfish were not favored by consumers. However, except taste parameter, loss of sensory quality was almost at the equal level for group A and group B during the storage. According to Orak and Kayisoglu (2008) the decrease in the values of sensory analyses was faster than chemical changes during frozen storage. Appearance of off-flavor may be attributed to WOF (Warm Over Flavor). WOF is usually associated with reheated meats and includes odors and flavors commonly described as stale or rancid (Brewer, 2007) also interaction of ketones, aldehydes, alcohols, hydrocarbons, acids, and epoxides with proteins may be produced off-colors (Thanonkaew *et al*., 2006).

Group A had lower TVB-N and pH values at the end of storage but increase of moisture content in group A was higher than group B during storage time. TBA and PV of group A because of high total lipid content were higher at the beginning of period and increasing in PV was almost at the equal level for both groups. For micro-biological analyses except *Staphylococcus aureus* and *Escherichia coli* counts that were equal in group, TPC, TC and psychrotrophics in Group A were higher than those in group B. According to Kilcast (2000) microbiological testing must be carried out prior to sensory testing and if there are samples with questionable microbiological quality should be submitted for sensory testing. Results of this study showed that pH, TVB-N and TBA are not correlated with the organoleptic characters. Fagan *et al*. (2003) have been reported that important predictors of acceptance are fish flavor, off flavor and toughness for fish minces, and off flavor and fish flavor for fish fingers. So considering sensory results, at the same production conditions, fish burgers from deep flounder (group A) had better quality characteristics than fish burgers from brushtooth lizardfish (group B) at the end of 5th month. Minimum scores by groups were 3-4. So according to these criteria maximal holding time for deep flounder and brushtooth lizardfish burgers is the end of the fourth and third month, respectively. However, to determine more precise shelf life, it is advisable that other chemical methods such as FFA (Free Fatty Acid) and AFR (Fluorescence Ratio in the Aqueous phase) also to be used to assess spoilage and shelf life of fish burgers from these two fish.

Acknowledgement

References

Adams, M.R. and Moss, M.O., 2000. Food Microbiology. Page.51. Cambridge. Published by the Royal Society of Chemistry.

AOAC, 2005. Association of Official Analytical Chemists. 18th edition. Gaithersburg, Maryland, USA.

Arvanitoyannis, I.S., Tsitsika, E.V. and Panagiotaki, P., 2005. Implementation of quality control methods (physicochemical, microbiological and sensory) in conjunction with multivariate analysis towards fish authenticity. International Journal of Food Science and Technology, **40:**37-263.

Aubourag, S.P., 1999. Lipid damage detection during the frozen storage of an underutilized fish species. Food Research International, **32:**497-502.

Aubourg, S.P., Lehmann, I. and Gallardo, J.M., 2002. Effect of previous chilled storage on rancidity development in frozen horse mackerel (*Trachurus trachurus*).

Journal of the Science of Food and Agriculture, **82:**1764–1771.

Biswaz, A.K., Keshri, R.C. and Bisht, G.S., 2004. Effect of enrobing and antioxidants on quality characteristics of precooked pork patties under chilled and frozen storage conditions. Meat Science, **66:**733-741.

Brewer, M.S., 2007. What is warmed-over flavor? American Meat Science Association Fact Sheet.

Cakli, S., Taskaya, L., Kisla, D., Celik, U., Ataman, C.A., Cadun, A., Kikinc, B. and Haji Maleki, R., 2005. Production and quality of fish fingers from different fish species. European Food Research and Technology, **220:**526-530.

Castro, P., Padron, J.C.P., Cansino, M.J.C., Vela´zquez, E.S.N. and Larriv, R.M.D., 2006. Total volatile base nitrogen and its use to assess freshness in European sea bass stored in ice. Food Control, **14:**245–248.

Chomnawang, C., Nantachai, K., Yongsawatdigul, J., Thawornchinsombut, S. and Tungkawachara, S., 2007. Chemical and biochemical changes of hybrid catfish fillet stored at 4ºC and its gel properties. Food Chemistry, **103:**420–427.

Fagan, J.D., Gormley, T.R. and Mhuircheaetaigh, M.U., 2003. Effect of freeze-chilling, in comparison with fresh, chilling and freezing, on some quality parameters of raw whiting, mackerel and salmon portions. Food Science and Technology, **36:**647–655.

Fan, W., Sun, J., Chen, Y., Qiu, J., Zhang, Y. and Chi, Y., 2008. Effects of chitosan coating on quality and shelf life of silver carp during frozen storage. Food Chemistry, pp.66-70.

Fernandes, J., Perez-Alvaez, J.A. and Fernandez-Lopez, J.A., 1997. Thiobarbituric acid test for monitoring lipid oxidation in meat. Food Chemistry, **59:**345-353.

Francis-Floyd, R., 2002. Fish Nutrition: Institute of Food and Agricultural Sciences.

Grigorakis, K., Taylor, K.D.A. and Alexis, M.N., 2003. Seasonal patterns of spoilage of ice-stored cultured gilthead sea bream (*Sparus aurata*). Food Chemistry, **81:**263–268.

Gundogan, N., Citak, S., Yucel, N. and Devren, A., 2005. A note on the incidence and antibiotic resistance of *Staphylococcus aureus* isolated from meat and chicken samples. Meat Science, **69:**807-810.

Hernandez-Herrero, M.M., Roig-Sagues, A.X., Lopez-Sabater, E.I., Rodriguez-Jerez, J.J. and Mora-Ventura, M.T., 1999. Total volatile basic nitrogen and other physicochemical and microbiological characteristics as related to ripening of salted anchovies. Journal of Food Science, **64:**344-347.

Hui, Y.H., 2007. General food quality factors. *In*: Nollet, L.M.L. (ed.) Handbook of Meat, Poultry and Seafood Quality. Blackwell Publishing.

ICMSF, 1978. Microorganisms in foods. The International Commission on Microbiological Specifications for Foods. Vol. 1, Toronto, Canada.

ISO, 936, 1998. Determination of total ash. International Organization for Standardization.

ISO, 1442, 1997. Determination of moisture content. International Organization for Standardization.

ISO, 1443, 1973. Determination of total fat. International Organization for Standardization.

ISO, 4832, 1991. General guidance for the enumeration of coliforms-colony count technique. International Organization for Standardization.

ISO, 7251, 2005. Detection and enumeration of presumptive escherichia coli-most probable number technique. International Organization for Standardization.

ISO, 8443, 2003. Horizontal method for the enumeration of microorganisms-colony count technique at 30ºC. International Organization for Standardization.

ISO, 17410, 2001. Horizontal method for the enumeration of psychrotrophic micro-organisms. International Organization Standardization.

Jamilah, B., 2004. Frozen seafoods. Hand book of frozen foods.

Khalil, A.H., 2000. Quality characteristics of low-fat beef patties formulated with modified corn starch and water. Food Chemistry, **68:**61-68.

Kilcast, D., 2000. Sensory evaluation methods for shelf-life assessment. In Kilcast, D. and Subramaniam, P. (eds.). The stability and shelf-life of food. Woodhead Publishing Limited and CRC Press LLC.

Kilina, B., Cakli, S. and Kisla, D., 2003. Quality changes of Sardine (*Sardina pilchardus* W., 1792) during frozen storage. E.U. Journal of Fisheries & Aquatic Sciences, **20:**139–146.

Kolakowska, A. and Deutry, J., 2006. Some comments on the usefulness of 2-thiobarbituric acid (TBA) test for the evaluation of rancidity in frozen fish. Molecular Nutrition & Food Research, **27:**513-518.

Kose, S., Boran, M. and Boran, G.K., 2006. Storage properties of refrigerated whiting mince after mincing by three different methods. Food Chemistry, **99:**129-135.

Lawrie, R.A. and Ledward, D.A., 2006. Lawrie's meat science, Woodhead Publishing Limited.

Losada, V., Barros-vela´zques, J. and Auborge, S.P., 2007. Rancidity development in frozen pelagic fish: Influence of slurry ice as preliminary chilling treatment. LWT, **40:**991–999.

Nakayama, T. and Yamamoto, M., 1977. Physical, chemical and sensory evaluations of frozen-stored deboned (minced) fish flesh. Journal of Food Science, **42:**900-905.

Nielsen, J.G., 1984. *Pseudorhombus elevatus* Bothidae. *In*: W. Fischer and G. Bianchi (eds.) FAO species identification sheets for fishery purpose. Western Indian Ocean fishery area51. FAO, Rome, Italy. Vol. 1.

Nollet, L.M.L., 2007. Handbook of meat, poultry and seafood quality. Wiely-Blackwell, 744P.

Orak, H.H. and Kayisoglu, S., 2008. Quality changes in whole, gutted and filletd three fish species (*Gadus euxinus*, *Mugil cephalus*, *Engraulis encrasicholus*) at frozen storage period (–26°C). Acta Scientiarum Polonorum Technologia Alimentaria, **7:**15-28.

Özogul, F. and Özogul, Y., 2000. Comparison of methods used for determination of total volatile basic nitrogen (TVB-N) in rainbow

trout (*Oncorhynchus mykiss*). Turkish Journal of Zoology, 24P.

Ozyurt, G., Polat, A. and Tokur, B., 2007. Chemical and sensory changes in frozen (-18ºC) wild sea bass (*Dicentrarchus labrax*) captured at different fishing seasons. International Journal of Food Science and Technology, **42:**887–893.

Parvane, V., 1998. Quality Control and Chemical Analyses of Food Products, Tehran University.

Pearson, D., 1981. The Chemical Analysis of Foods. 8th edn. London, UK.

Pons-Sanchez-Cascado, S., Vidal-Carou, M.C., Nunes, M.L. and Veciana-Nogues, M.T., 2006. Sensory analysis to assess the freshness of Mediterranean anchovies (*Engraulis encrasicholus*) stored in ice. Food Control, **17:**564–569.

Ravindranathan, N.P., Thankamma, R. and Opakumar, K., 1982. Biochemical changes of fish fingers held at frozen storage. Fishery Technology, **19:**19-23.

Ruiz-Capillas, C. and Moral, A., 2001. Correlation between biochemical and sensory quality indices in hake srored in ice. Food Research International, **34:**441-447.

Safari, R. and Yosefian, M., .2006. Changes in TVN (Total Volatile Nitrogen) and psycrotrophic bacteria in Persian sturgeon Caviar (*Acipenser persicus*) during processing and cold storage. Journal of Applied Ichthyology, **22:**416-418.

Suvanich, V., Marshall, D.L. and Jahncke, M.L., 2000. Microbiological and color quality changes of channel catfish frame mince during chilled and frozen storage. Journal of Food Science, **65:**151-154.

Thanonkaew, A., Benjakul, S., Visessanguan, W. and Decker, E.A., 2006. The effect of metal ions on lipid oxidation, color and physicochemical properties of cuttlefish (*Sepia pharaonis*) subjected to multiple freeze–thaw cycles. Food Chemistry, **95:**591-599.

Tokur, B., Çaklı, Ş. and Polat, A., 2006. The quality changes of Trout (*Oncorhynchus mykiss* W., 1792) with a vegetable topping during frozen storage (-18°C). E.U. Journal of Fisheries & Aquatic Sciences, **23:**345–350.

Vanderelst, R.,1993. A guide to the common sea fishes of Southern Africa.

Vandenbergh, P.A., 1993. Lactic acid bacteria, their metabolic products and interference with microbial growth. Federation of European Microbiological Societies, **12:** 221-238.

Vareltzis, K., Koufidis, D., Gavriilidou, E., Papavergou, E. and Vasiliadou, S., 1997. Effectiveness of a natural Rosemary (*Rosmarinus officinalis*) extract on the stability of filleted and minced fish during frozen storage. European Food Research and Technology, **205:**93-96.

Venugopal, V. and Shahidi, F., 1995. Value-added products from underutilized fish

species. Food Science and Nutrition, **35:**431-453.

Widayaka, K., Setyawardani, T. and Sumarmono, J., 2001. The effect of storage and cooking on lipid oxidation of raw and cooked beef and goat meat. Asia Pacific Journal of Clinical Nutrition, 10**(4):**548.

Zareh Gashti, G., 2002. Estimation of micro-biological and chemical variations in minced fish processing of Atlantic pollock (*Pollachius vireos*). The United Nations University. Final project, pp.1-30.

ارزیابی کیفیت فیش برگرهای تولید شده از کفشک پرلکه (*Pseudorhombus elevatus*) و کیجار منقوط (*Saurida undosquamis*)

طی شرایط نگهداری در ۱۸- درجه سانتیگراد

مریم محمودزاده'؛ عباسعلی مطلبی'؛ هدایت حسینی'؛ پریوش هراتیان'؛ حامد احمدی'؛ مهرداد محمدی' و رامین خاکسار'*

چکیده

تغییرات میکروبی، شیمیایی و حسی فیش برگرهای تهیه شده از ماهی کفشک پرلکه (گروه A) و ماهی کیجار منقوط (گروه B) طی نگهداری در ۱۸- درجه سانتیگراد به مدت ۵ ماه تعیین شد. شمارشهای میکروبی شامل شمارش کلی میکروبی (TPC)، شمارش کلی کلیفرم (TC) استافیلوکوکوس اورئوس، سرماگراها و اشرشیاکلی طی انبارداری انجمادی کاهش یافتند. میزان کاهش در کیجار منقوط بیشتر از کفشک پرلکه بود به جز کاهش در شمارش استافیلوکوکوس اورئوس که تقریباً در دو گروه برابر بود. میزان pH بطور معنیداری (P<0.05) طی ماه اول و دوم بترتیب در فیش برگرهای ماهی کیجار منقوط و کفشک پرلکه افزایش پیدا کرد سپس تغییر معنیداری (P>0.05) در میزان pH تا آخر دوره انبارداری مشاهده نشد. میزان رطوبت هر دو گروه تا انتهای دوره نگهداری افزایش پیدا کرد اما افزایش رطوبت در فیش برگرهای کفشک پرلکه بیشتر از کیجار منقوط بود. میزان TVB-N هر دو گروه بطور معنی داری (P<0.05) تا آخر ماه دوم افزایش پیدا کرد سپس کاهش یافت و تغییر معنیداری تا انتهای دوره انبارداری مشاهده نشد. میزان TBA فیش برگرهای کفشک پرلکه با پیشرفت دوره انبارداری کاهش معنی داری P<0.05 یافت در صورتی که در مورد کیجار منقوط تا انتهای ماه دوم افزایش معنی داری (P=۰/۰۰۶) داشت سپس کاهش نشان داد. میزان پراکسید در هر دو گروه بطور معنیداری (P<0.05) طی زمان انبارمانی افزایش پیدا کرد ولی کاهش معنیداری در انتهای ماه سوم و چهارم (P<0.05) بترتیب برای کفشک پرلکه و کیجار منقوط مشاهده شد. پارامترهای حسی رنگ، بافت، مزه و پذیرش

۱ - دانشکده تغذیه و صنایع غذایی، دانشگاه علوم پزشکی شهید بهشتی، تهران صندوق پستی: ۴۷۴۱-۱۹۳۴۵

۲ - موسسه تحقیقات شیلات ایران، تهران صندوق پستی: ۶۱۱۶-۱۴۱۵۵

* پست الکترونیکی نویسنده مسئول: r.khaksar@sbmu.ac.ir

کلی در هر دو گروه کاهش معنی‌دار ($P<0.05$) داشتند اما فی‌ش برگرهای کفشک پرلکه امتی‌ازات بهتری نسبت فی‌ش برگرهای کی‌جار منقوط در شروع و پای‌ان دوره انبارداری کسب نمودند.

کـلـمـات کـلـیـدی : کفشک پرلکه، *Pseudorhombus elevatus*، کی‌جار منقوط، *SauridaUndosquamis* فی‌ش برگ، انبارداری انجمادی، کی‌فی‌ت