

Dept. of Food Hygiene, Animal Health Research Institute  
Agricultural Research Center, Alexandria, Egypt.

## **CHEMICAL INDICATORS OF SEA FOOD SPOILAGE** (With 10 Tables)

**MERVAT KAMAL IBRAHIM RAGAB**

(Received at 8/9/2005)

### **الدلائل الكيماوية لفساد المأكولات البحرية**

**ميرفت كمال إبراهيم رجب**

لقد أجريت الدراسة على ١٠٠ عينة من سمك الماكريل والهورس ماكريل والمكرونه والجمبرى الطازج المجمع من الأسكندرية. ولقد تم تخزينها عند درجة الصفر المئوى فى الثلج ودرجة ٤ درجة مئوية على رف الثلجة وعند -١٠ درجة مئوية فى الفريزر حتى الوصول إلى درجة التحلل والفساد. وتشير النتائج إلى الآتى:- ظهرت مؤشرات الفساد على سمك الماكريل المخزن فى الثلج بعد ٦ أيام وبعد ٣ أيام من التخزين عند ٤ درجة مئوية وبعد ١٢ شهر من التخزين عند -١٠ درجة مئوية. وفى حالة سمك الهورس ماكريل ظهرت مؤشرات الفساد بعد ٦,٥ أيام من التخزين عند درجة صفر و٤ درجة مئوية على التوالى وبعد ١٣ شهر من التخزين عند -١٠ درجة مئوية. وقد تم التحلل لسمك المكرونه بعد ٥,٦ أيام من التخزين عند ردرجة الصفر و٤ درجة مئوية على التوالى. وفى حالة الجمبرى ظهرت دلائل التحلل بعد ٦ أيام وبعد يوم واحد من التخزين عند درجة الصفر و٤ درجة مئوية على التوالى. وبعد شهر من التخزين عند -١٠ درجة مئوية. أسفرت نتائج التحاليل الكيماوية للأسماك والجمبرى إلى الآتى:- كان أعلى مستوى لمعدل النتروجين الكلى المتصاعد (١١٨,٣ مللى جم / ١٠٠ جم) فى الجمبرى المخزن لمدة شهر عند -١٠ درجة مئوية بينما أقل مستوى كان فى سمك المكرونه الطازج. وفى حالة مؤشرات تحلل الدهون كان أعلى مستوى للأحماض الدهنية الحرة (٤,٦٣ مللى/كجم) وأعلى مستوى للرقم الحمضى (٩ مللى/جم). فى سمك المكرونه المخزن لمدة ١٤ شهر عند -١٠ درجة مئوية. وكان أعلى مستوى لحامض الثيوباربيتيورك (٦ مللى جم/كجم) فى الجمبرى المخزن لمدة شهر عند -١٠ درجة مئوية.

### **SUMMARY**

A total of 100 samples (25 of each) of Mackerel fish (*Scomber scombrus*), Horse mackerel (*Treacherous tracharus*), Silver hake (*Merluccinus bilinearis*) and Shrimp (*Penus kerathurus*) were purchased from fishermen in Alexandria and stored at 0°C (held on ice), 4°C (refrigerator) and -10°C (home freezer), for as long as acceptable to

study how time and temperature affect the quality and acceptability of fish and shellfish. Changes in the quality of fish were determined by chemical analyses which were correlated with sensory evaluation made by an experienced panel. Results from this study were compared to chemical analytical and sensory results from a corresponding study to follow changes in the quality of fish and shellfish during cold storage after harvest. Our results indicated that the iced mackerel fish (held on ice) retained its good quality characteristics for about one day post harvest. After four days, Mackerel fish quality decreased and reached the limit of unacceptability at sixth day of storage. Sensory score results of refrigerated and frozen fish showed that, it was considered organoleptically putrid (completely spoiled), after three days of storage at 4°C and after 12 months of storage at -10°C. Concerning Horse mackerel fish, it was of excellent quality at the fifth day of storage at 0°C, at first day of storage at 4°C and after one month of frozen storage, and it became spoiled, at the 15<sup>th</sup> day, 6<sup>th</sup> day and 13<sup>th</sup> month of storage at 0, 4 and -10°C, respectively. However, in case of Silver hake, sensory score results showed that at the second day of storage at 0 and 4°C, it began to lose its quality (border line) and became completely spoiled at sixth and fifth days of storage at 0 and 4°C, respectively, and at the 14<sup>th</sup> month of frozen storage (-10°C). Regarding, Shrimp quality, it was of excellent quality for one day of storage at 0°C and became completely spoiled at 6<sup>th</sup> day of storage, at the mean time, shrimp stored at 4 and -10°C showed complete signs of spoilage after one day and one month., respectively. Results of chemical analyses of the examined fish and shellfish indicated that the mean highest total volatile nitrogen (TVN) (118.30 mg/100gm) was found in shrimp stored at -10 °C for one month while the lowest (18.78mg/100gm) was in fresh Silver hake fish. Concerning lipid spoilage parameters, Silver hake fish stored for 14 months at -10 °C showed the highest mean values of free fatty acids (FFA) (4.63 ml/gm), acid value (AV) (9.260ml/gm), fresh mackerel fish had the highest level of conjugated dienes (CD) (0.122n.mole/mg), while Shrimp stored at -10 °C for one month had the highest mean values of thiobarbituric acid value TBA (6.542mgMA/kg). At the same time, fresh Horse mackerel fish showed the lowest mean values of FFA (0.554 mg/ml), TBA (0.339 mgMA/kg and AV (1.108ml/gm), while Mackerel stored at -10 °C for 12 month showed the lowest value of CD (0.009n mole/mg). Results of shelf life of the examined fish during cold storage indicated that iced Mackerel was inedible after 6 days, refrigerated one was putrid after 3 days and frozen mackerel had spoiled after 12 months. Regarding horse mackerel

fish, shelf life was 15,6 days for iced and refrigerated storage and 13 months for frozen storage. However, Silver hake had shelf life of 6,5 days for iced, refrigerated storage and 14 months of frozen storage. In case of Shrimp, complete spoilage began after 6,1 days, 1 month of storage at 0,4 and -10 °C, respectively. Statistical analytical results indicated the presence of strong correlation between sensory scores and chemical parameters in all the examined fish and shellfish during storage period.

*Key words: Sea food, fish, shellfish, fish spoilage, mackerel, shrimp.*

## INTRODUCTION

Fish spoilage was defined as a change in odor, taste and consistency of fish or fish product that makes it unsafe, less acceptable to the consumer for its original intended purpose (Olley and Quarmby, 1981). Upon death, certain enzymes are inactivated and bacteria that are normally kept in check in the live fish now thrive on the proteins and fats, with the increase of the bacterial flora, decomposition of fish is rapid. Spoilage is therefore, the result of a combination of two factors enzyme activity (autolytic break down) and bacteria. The rate of spoilage depends largely on temperature, which controls to a large extent enzyme (autolytic) and bacterial break down. The rate also varies with species, amount of food in the gut, seasonality, gender age, geographical location. A part from intrinsic factors such as moisture content, pH, onset of rigor mortis, chemical composition and redox potential may influence spoilage in fish. Literally every stage from harvest to consumption affects spoilage in one way or another (Botta, 1995).

Fish are much more perishable than any other high protein muscle foods. The high degree of perishability is primarily due to the large amounts of non protein nitrogen (NPN) for example free amino acids, volatile nitrogen bases (ammonia), trimethylamine and histamine. The readily non protein nitrogen compounds support the post mortem bacterial growth. Therefore, the microbiological spoilage has been noted as the main cause of fish deterioration, followed by non microbiological namely oxidative rancidity and then chemical or enzymatic denaturation of protein (autolytic spoilage). Generally in microbiological spoilage, bacteria are characterised both by their dominance in the microflora of spoiling fish and their ability to produce spoilage compounds. Accumulated metabolic products of bacteria are the primary causes of

the organoleptic spoilage of raw fish. The microbial activity on reduced compounds, produced the characteristic fishy, ammoniac odours and change the texture to the slimy. Microbiological spoilage causes the formation of volatile bases principally ammonia, trimethylamine. These bases can be distilled, collected and made to neutralise acids. The amount of acid used is a measure of the total bases distilled and it corresponds with the level of spoilage or freshness indirectly. There are several procedures used for the determination of total volatile basic content and each procedure gives a different result for the same fish sample. The difference has been attributed to the way bases are released and distilled from the fish. (Kraft, 1992 and Botta, 1995).

However in chemical spoilage, after death fish lipids are subjected to two major changes, lipolysis and auto-oxidation. The main reactants in these processes involves atmospheric oxygen and fish lipids but the reactions are initiated and accelerated by heat, light (especially ultra violet light) and several organic and inorganic substances like copper and iron ions (Mayer and ward, 1991). The end products of lipolysis was liberation of free fatty acids and the primary products of auto-oxidation of lipids was peroxides. Almost immediately after peroxides are formed, the non conjugated double bonds ( $C=C-C-C=C$ ) that are present in the natural unsaturated lipids are converted to conjugated double bonds ( $C=C-C=C$ ). Conjugated dienes absorb ultra violet radiation strongly at 233nm. Thus oxidation can be followed by dissolving the lipids in a suitable organic solvent and measuring the change in its absorbance with time using ultra violet spectrophotometer (Santiago *et al.*, 1997). During storage, In later stages of lipid oxidation, secondary oxidation products will usually be present and thus be indicative of a history of autoxidation. These products comprise aldehydes, ketones, short chain fatty acid and others, many of which have very unpleasant odours and flavours, and which in combination yield the fishy and rancid character associated with oxidized fish lipid. Some of the aldehydic secondary oxidation products react with thiobarbituric acid, forming a reddish coloured product that can be determined spectrophotometrically. Using this principle, a measure of thiobarbituric acid-reactive substances (TBA-RS) can be obtained. The results are expressed in terms of the standard (di-) aldehyde used, malonaldehyde, and reported as micromoles malonaldehyde present in 1 g of fat. (A note of caution: Sometimes the TBA-results may be expressed as mg malonaldehyde in 1 g of fat, or as amount of malonaldehyde ( $\mu\text{mol}$  or  $\mu\text{g}$ ) in relation to amount of tissue analyzed.) In the autolytic spoilage, the enzymes and

other related chemical reactions do not immediately cease their activities in the fish muscle. Their continuation initiates other precursor processes like rigor mortis, which is a basis for autolytic spoilage. The autolytic changes are responsible for the early quality loss in fresh fish but contribute very little to spoilage of chilled fish and fish products. However under frozen conditions, autolytic enzymes cause autolytic tissue damage which tends to be more pronounced in heavily feeding fish than petite feeders.

The bacterial and autolytic enzymes degradation are the parameters used to identify and quantitate sea food quality. Therefore, methods for evaluation of the degree of spoilage are essential. Of course, sensory methods are useful for identifying products of good or poor quality. A characteristic sensory changes in fish post mortem vary considerably depending on fish species and storage method. A characteristic pattern of the deterioration of fish stored in ice can be divided into four phases: phase one in which fish is very fresh and has a sweet, seaweedy and delicate taste. While in phase two, there is a loss of the characteristic odour and taste, the flesh become neutral but has no off flavours. The texture is still pleasant. In phase three, there is a significance of spoilage and a range of volatile unpleasant- smelling substances is produced depending on the fish species and type of spoilage (aerobic, anaerobic). In phase four, the fish can be characterized as spoiled and putrid (Nielsen, 1995). Chemical methods may best be used in resolving issues regarding products of marginal quality. In addition, chemical indicators have been used to replace more time consuming microbiological methods. Such objective methods should however correlate with sensory quality evaluations (Zdzislaw, *et al.*, 1990, Gram, 1992 and Hultin, 1992). Since, Chemical and biological changes, which begin immediately after fish are harvested affect the quality of the product and affected by time and temperature. Therefore the objective of this work is to explain how time and temperature ultimately affect the quality (and thus acceptability) of fresh fish and shell fish and to follow changes in quality that occur during typical conditions of preservation and storage.

## **MATERIALS and METHODS**

Sample preparation: A total of 100 samples of fresh fish and shellfish: Mackerel (*Scomber scombrus*), Horse mackerel (*Treacherous trachurus*), Silver hake (*Merluccius bilinearis*) and Shrimp (*Penus*

kerathurus) were random chosen from fishermen in Alexandria (25 samples for each). Samples were divided into three groups: the first group was held in ice (at 0 °C) and the second group was stored in a refrigerator (at 4 °C) and the third was stored in home freezer (at -10 °C) for several periods of time until the fish were noticeably in an advanced stage of decomposition (as determined by off odour and slimy texture). Samples at each storage temperature at each storage time were evaluated by sensory and chemical methods.

**Sensory quality evaluation:**

Fish quality in sensory evaluation was assessed by trained panel consisting of 10 persons. Shelf life was established by the duration between the time of capture and the time when the cooked quality score dropped below the limit of acceptability (Singh, 2000). Samples were cut into 20gm pieces, boiled in aluminium boxes at 98 °C for five minutes and served to each panellist. Samples were served in duplicates (Winger, 2000).

A scale from 0-10 was used samples retaining odours and flavours typical for the species were given scores above 6. Fish with slightly off odours and off flavours were given scores 4-5. Scores below 4 indicated objectionable strong unpleasant off-odours and off flavours (Meilgaard, *et al.*, 1991).

**Chemical quality evaluation:**

**Protein spoilage indicator:**

Total volatile basic nitrogen: it was determined by steam distillation method (Malle and Pourmeyrol 1998).

**Lipid spoilage indicators:**

Free fatty acids and acid value were determined according to Pearson (1970).

Conjugated dienes were determined by using UV -visible spectrophotometer. (Sanitago, *et al.*, 1997).

Thiobarbituric acid reactive substances were determined according to Li, *et al.*, 2001.

## RESULTS

Sensory and chemical quality of the examined fish and shellfish are recorded in Tables 1-10.

**Table 1:** summary of sensory and chemical quality evaluation of mackerel fish during cold storage

Storage temperature (degrees)	Holding time	Sensory scores (mean values $\pm$ S.E)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)
			T.V.N.(mg/100gm)	FFA(ml/gm)	AV(ml/gm)	CD(n mole/mg)	TBA(mgMA/kg)
0	0 (days)	9.5 $\pm$ 0.159	24.84 $\pm$ 0.796	1.194 $\pm$ 0.0164	2.388 $\pm$ 0.033	0.122 $\pm$ 0.004	0.806 $\pm$ 0.018
0	1	7.54 $\pm$ 0.194	27.4 $\pm$ 1.515	1.602 $\pm$ 0.046	3.204 $\pm$ 0.108	0.097 $\pm$ 0.007	1.324 $\pm$ 0.016
0	2	4.28 $\pm$ 0.116	32.84 $\pm$ 1.510	1.868 $\pm$ 0.025	3.736 $\pm$ 0.062	0.057 $\pm$ 0.004	2.532 $\pm$ 0.021
0	4	4.44 $\pm$ 0.136	47.62 $\pm$ 1.323	2.360 $\pm$ 0.163	4.720 $\pm$ 0.273	0.030 $\pm$ 0.002	3.614 $\pm$ 0.013
0	6	2.32 $\pm$ 0.139	64.14 $\pm$ 1.371	2.706 $\pm$ 0.017	5.412 $\pm$ 0.018	0.018 $\pm$ 0.001	4.194 $\pm$ 0.053
Total	6	5.616 $\pm$ 0.149	39.368 $\pm$ 1.303	1.945 $\pm$ 0.053	3.892 $\pm$ 0.099	0.065 $\pm$ 0.004	2.494 $\pm$ 0.024
4	0 (days)	9.5 $\pm$ 0.159	24.84 $\pm$ 0.796	1.194 $\pm$ 0.0164	2.388 $\pm$ 0.033	0.122 $\pm$ 0.004	0.806 $\pm$ 0.018
4	1	7.54 $\pm$ 0.178	28.74 $\pm$ 0.654	1.64 $\pm$ 0.038	3.280 $\pm$ 0.164	0.095 $\pm$ 0.002	1.532 $\pm$ 0.013
4	2	4.32 $\pm$ 0.146	34.82 $\pm$ 0.396	1.862 $\pm$ 0.047	3.724 $\pm$ 0.198	0.041 $\pm$ 0.003	2.918 $\pm$ 0.016
4	3	2.42 $\pm$ 0.153	61.54 $\pm$ 0.702	2.100 $\pm$ 0.080	4.200 $\pm$ 0.021	0.016 $\pm$ 0.001	4.786 $\pm$ 0.017
Total	3	5.945 $\pm$ 0.159	37.49 $\pm$ 0.637	1.699 $\pm$ 0.045	3.396 $\pm$ 0.104	0.003 $\pm$ 0.007	2.511 $\pm$ 0.016
-10	0 (months)	9.5 $\pm$ 0.159	24.84 $\pm$ 0.796	1.194 $\pm$ 0.0164	2.388 $\pm$ 0.033	0.122 $\pm$ 0.004	0.806 $\pm$ 0.018
-10	3	7.76 $\pm$ 0.093	30.50 $\pm$ 0.490	2.790 $\pm$ 0.013	5.580 $\pm$ 0.031	0.055 $\pm$ 0.001	1.254 $\pm$ 0.011
-10	6	4.46 $\pm$ 0.121	49.17 $\pm$ 1.424	3.146 $\pm$ 0.050	6.292 $\pm$ 0.092	0.010 $\pm$ 0.002	3.266 $\pm$ 0.013
-10	12	2.26 $\pm$ 0.169	69.28 $\pm$ 1.990	3.36 $\pm$ 0.040	6.720 $\pm$ 0.049	0.009 $\pm$ 0.001	6.348 $\pm$ 0.026
Total	12	5.995 $\pm$ 0.136	43.448 $\pm$ 1.175	2.623 $\pm$ 0.030	5.246 $\pm$ 0.051	0.049 $\pm$ 0.002	2.919 $\pm$ 0.017

Ascale from 0-10 was used .samples retaining odours and flavours typical for the species were given scores above6. Fish with slightly off odours and off flavours were given scores 4-5. Scores below 4 indicated objectionable strong unpleasant off-odours and off flavours.

**Table 2:** summary of sensory and chemical quality evaluation of horse mackerel fish during cold storage

Storage temperature (degrees)	Holding time	Sensory scores (meanvalue±S.E)	Chemical parameter changes(meanvalue±S.D)	Chemical parameter changes(meanvalue±S.D)	Chemical parameter changes(meanvalue±S.D)	Chemical parameter changes(meanvalue±S.D)	Chemical parameter changes(meanvalue±S.D)
			T.V.N.(mg/100gm)	FFA(ml/gm)	AV(ml/gm)	CD(n mole/mg)	TBA(mgMA/kg)
0	0 (days)	8.56±0.157	19.89±0.602	0.554±0.0134	1.108±0.010	0.050±0.001	0.339±0.003
0	2	8.68±0.146	28.94±0.321	1.182±0.002	2.264±0.018	0.061±0.002	0.862±0.002
0	5	8.60±0.192	36.06±0.934	1.466±0.006	2.932±0.016	0.065±0.005	1.838±0.022
0	8	4.5±0.173	43.88±1.712	3.026±0.011	6.052±0.025	0.034±0.001	4.63±0.007
0	15	2.4±0.141	64.34±1.64	3.800±0.016	7.60±0.020	0.019±0.002	6.936±0.005
Total	15	6.55±0.162	38.622±1.042	2.006±0.010	4.012±0.015	0.046±0.002	2.921±0.008
4	0 (days)	8.56±0.157	19.89±0.602	0.554±0.0134	1.108±0.010	0.050±0.001	0.339±0.003
4	1	8.28 ±0.218	24.26±0.680	1.296±0.011	2.592±0.016	0.068±0.0004	0.926±0.006
4	3	4.50±0.159	32.18±0.832	1.588±0.013	3.176±0.010	0.073±0.002	2.354±0.008
4	4	4.36±0.150	59.916±1.116	2.02±0.016	4.040±0.010	0.045±0.006	2.729±0.006
4	6	2.520±0.154	71.800±0.719	2.525±0.006	5.050±0.010	0.035±0.002	4.762±0.001
Total	6	5.644±0.167	41.609±0.790	1.597±0.012	3.194±0.011	0.054±0.003	2.222±0.005
-10	(months) 0	8.56±0.157	19.89±0.602	0.554±0.0134	1.108±0.010	0.050±0.001	0.339±0.003
-10	1	7.240±0.051	32.620±1.100	1.032±0.001	2.064±0.011	0.057±0.009	0.926±0.005
-10	3	4.100±0.045	45.99±0.740	1.608±0.013	3.216±0.002	0.017±0.004	2.354±0.006
-10	6	4.64±0.108	68.00± 0.796	2.300±0.002	4.600±0.010	0.018±0.002	3.729±0.003
-10	13	2.820±0.037	69.800±0.960	3.30±0.010	6.600±0.003	0.010±0.003	4.969±0.004
Total	13	5.472±0.080	59.08±0.838	1.759±0.008	3.518±0.007	0.030±0.001	2.463±0.004

Ascale from 0-10 was used .samples retaining odours and flavours typical for the species were given scores above6. Fish with slightly off odours and off flavours were given scores 4-5. Scores below 4 indicated objectionable strong unpleasant off-odours and off flavours.



**Table 3:** summary of sensory and chemical quality evaluation of silver hake fish during cold storage

Storage temperature (degrees)	Holding time	Sensory scores (mean values $\pm$ S.E)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)	Chemical parameter changes (mean values $\pm$ S.D)
			T.V.N.(mg/100gm)	FFA(ml/gm)	AV(ml/gm)	CD(n mole/mg)	TBA(mgMA/kg)
0	0 (days)	8.52 $\pm$ 0.177	18.78 $\pm$ 0.740	0.818 $\pm$ 0.005	1.636 $\pm$ 0.002	0.052 $\pm$ 0.001	0.924 $\pm$ 0.003
0	1	8.18 $\pm$ 0.341	24.84 $\pm$ 0.639	0.954 $\pm$ 0.009	1.908 $\pm$ 0.013	0.057 $\pm$ 0.002	0.962 $\pm$ 0.004
0	2	4.36 $\pm$ 0.150	32.62 $\pm$ 1.073	1.268 $\pm$ 0.011	2.536 $\pm$ 0.015	0.066 $\pm$ 0.004	2.214 $\pm$ 0.021
0	4	4.40 $\pm$ 0.105	39.70 $\pm$ 0.926	1.288 $\pm$ 0.013	2.576 $\pm$ 0.025	0.037 $\pm$ 0.006	2.630 $\pm$ 0.007
0	6	2.16 $\pm$ 0.108	66.76 $\pm$ 1.101	1.400 $\pm$ 0.007	2.800 $\pm$ 0.025	0.026 $\pm$ 0.001	2.936 $\pm$ 0.005
Total	6	5.524 $\pm$ 0.176	36.540 $\pm$ 0.896	1.146 $\pm$ 0.009	2.292 $\pm$ 0.016	0.048 $\pm$ 0.003	1.858 $\pm$ 0.008
4	0 (days)	8.52 $\pm$ 0.177	18.78 $\pm$ 0.740	0.818 $\pm$ 0.005	1.636 $\pm$ 0.002	0.052 $\pm$ 0.001	0.924 $\pm$ 0.003
4	1	8.200 $\pm$ 0.352	24.04 $\pm$ 0.365	0.847 $\pm$ 0.002	1.694 $\pm$ 0.019	0.062 $\pm$ 0.002	0.956 $\pm$ 0.002
4	2	4.440 $\pm$ 0.191	29.720 $\pm$ 1.028	0.898 $\pm$ 0.005	1.796 $\pm$ 0.010	0.069 $\pm$ 0.004	2.186 $\pm$ 0.001
4	4	4.480 $\pm$ 0.183	57.720 $\pm$ 0.687	1.00 $\pm$ 0.003	2.000 $\pm$ 0.006	0.043 $\pm$ 0.006	2.356 $\pm$ 0.006
4	5	2.320 $\pm$ 0.146	64.50 $\pm$ 0.740	1.420 $\pm$ 0.004	2.840 $\pm$ 0.010	0.022 $\pm$ 0.002	2.729 $\pm$ 0.003
Total	5	5.592 $\pm$ 0.210	38.952 $\pm$ 0.712	0.997 $\pm$ 0.004	1.994 $\pm$ 0.010	0.050 $\pm$ 0.003	2.762 $\pm$ 0.003
-10	0 (months)	8.52 $\pm$ 0.177	18.78 $\pm$ 0.740	0.818 $\pm$ 0.005	1.636 $\pm$ 0.002	0.052 $\pm$ 0.001	0.924 $\pm$ 0.003
	1	8.200 $\pm$ 0.352	34.100 $\pm$ 0.652	1.074 $\pm$ 0.002	2.148 $\pm$ 0.004	0.067 $\pm$ 0.003	0.989 $\pm$ 0.006
-10	3	4.500 $\pm$ 0.173	57.18 $\pm$ 1.193	1.672 $\pm$ 0.010	3.344 $\pm$ 0.009	0.071 $\pm$ 0.002	2.145 $\pm$ 0.001
-10	6	4.46 $\pm$ 0.181	64.620 $\pm$ 0.792	3.673 $\pm$ 0.001	7.346 $\pm$ 0.027	0.052 $\pm$ 0.001	3.244 $\pm$ 0.002
-10	14	2.52 $\pm$ 0.143	77.52 $\pm$ 0.893	4.630 $\pm$ 0.005	9.260 $\pm$ 0.001	0.038 $\pm$ 0.003	5.560 $\pm$ 0.004
Total	14	5.640 $\pm$ 0.205	50.440 $\pm$ 0.854	2.373 $\pm$ 0.005	4.746 $\pm$ 0.009	0.056 $\pm$ 0.002	2.352 $\pm$ 0.003

Ascale from 0-10 was used .samples retaining odours and flavours typical for the species were given scores above6. Fish with slightly off odours and off flavours were given scores 4-5. Scores below 4 indicated objectionable strong unpleasant off-odours and off flavours.

**Table 4:** summary of sensory and chemical quality evaluation of shrimp during cold storage

Storage temperature (degrees)	Holding time (days)	Sensory scores (meanvalues± S.E)	Chemical parameter changes(meanvalues± S.D)	Chemical parameter changes(meanvalues± S.D)	Chemical parameter changes(meanvalues± S.D)	Chemical parameter changes(meanvalues± S.D)	Chemical parameter changes(meanvalues± S.D)
			T.V.N.(mg/100gm)	FFA(ml/gm)	AV(ml/gm)	CD(n mole/mg)	TBA(mgMA/kg)
0	0	8.26±0.357	30.96±1.036	1.030±0.0160	2.060±0.016	0.063±0.002	0.734±0.006
0	1	8.20±0.352	60.22±0.554	1.200±0.009	2.496±0.011	0.067±0.002	1.536±0.013
0	2	4.46±0.181	32.84±1.515	1.616±0.011	3.230±0.013	0.072±0.001	2.640±0.008
0	4	4.66±0.103	69.98±0.646	2.016±0.011	4.032±0.011	0.045±0.002	2.926±0.017
0	6	2.44±0.175	79.60±0.561	2.396±0.011	4.792±0.016	0.022±0.002	3.190±0.007
Total	6	5.604±0.234	54.72±0.862	1.652±0.012	3.322±0.013	0.054±0.004	2.205±0.010
4	0	8.26±0.357	30.96±1.036	1.030±0.0160	2.060±0.016	0.063±0.002	0.734±0.006
4	1	2.34 ±0.157	93.18±0.559	2.420±0.009	4.840±0.018	0.014±0.001	3.234±0.006
Total	1	5.3±0.257	62.07±0.798	1.725±0.013	3.450±0.017	0.039±0.002	1.984±0.006
-10	0	8.26±0.357	30.96±1.036	1.030±0.0160	2.060±0.016	0.063±0.002	0.734±0.006
-10	21	8.68±0.146	64.74±1.092	2.396±0.011	4.792±0.016	0.065±0.003	4.664±0.015
-10	30	2.64±0.121	118.30±3.010	4.467±0.010	8.934±0.061	0.024±0.002	6.542±0.005
Total	30	6.527±0.208	71.30±1.713	2.631±0.013	5.262±0.031	0.051±0.002	3.980±0.009

Ascale from 0-10 was used .samples retaining odours and flavours typical for the species were given scores above6. Fish with slightly off odours and off flavours were given scores 4-5. Scores below 4 indicated objectionable strong unpleasant off-odours and off flavours.

**Table 5:** Shelf life of the examined sea food during cold storage

Sea food	Storage Temperatures (degrees)	Shelf life
<b>Fish:</b>		
Mackerel	0	6 days
	4	3 days
	-10	12 month
Horse mackerel	0	15 days
	4	6 days
	-10	13 month
Silver hake	0	6 days
	4	5 days
	-10	14 month
Shrimp	0	6 days
	4	1 day
	-10	1 month

**Table 6:** Correlation coefficient between sensory scores and total volatile nitrogen of the examined sea food during cold storage

Type of sea food	Storage temperatures (degrees)	Sensory scores (mean values)	Total volatile nitrogen (mean values) (mg/100gm)	Correlation coefficient(r )
Mackerel	0	5.616	39.408	0.971
	4	6.550	37.490	0.916
	-10	5.780	37.240	0.938
Horse mackerel	0	6.550	36.622	0.875
	4	5.644	41.609	0.892
	-10	5.472	47.300	0.826
Silver hake	0	5.524	36.540	0.937
	4	5.592	38.952	0.942
	-10	5.640	50.440	0.971
Shrimp	0	5.604	67.080	0.934
	4	5.300	62.460	0.975
	-10	6.527	68.00	0.987

**Table 7:** Correlation coefficient between sensory scores and free fatty acids of the examined sea food during cold storage

Type of sea food	Storage temperatures (degrees)	Sensory scores(mean values)	Free fatty acid content (mean values) (ml/gm)	Correlation coefficient(r )
Mackerel	0	5.616	1.945	0.927
	4	6.550	1.698	0.942
	-10	5.780	2.622	0.881
Horse mackerel	0	6.550	2.257	0.845
	4	5.644	1.597	0.946
	-10	5.472	3.300	0.823
Silver hake	0	5.524	1.146	0.916
	4	5.592	2.007	0.824
	-10	5.640	2.370	0.851
shrimp	0	5.604	1.652	0.879
	4	5.300	1.860	0.893
	-10	6.527	2.631	0.906

**Table 8:** Correlation coefficient between sensory scores and acid number of the examined sea food during cold storage

Type of sea food	Storage temperatures (degrees)	Sensory scores(mean values)	Acid number (mean values) (ml/gm)	Correlation coefficient(r )
Mackerel	0	5.616	2.890	0.899
	4	6.550	3.396	0.937
	-10	5.780	5.244	0.879
Horse mackerel	0	6.550	4.514	0.925
	4	5.644	3.194	0.905
	-10	5.472	3.518	0.844
Silver hake	0	5.524	2.292	0.803
	4	5.592	4.014	0.798
	-10	5.640	4.740	0.700
shrimp	0	5.604	3.304	0.970
	4	5.300	3.720	0.940
	-10	6.527	5.262	0.874

**Table 9:** Correlation coefficient between sensory scores and conjugated dienes of the examined sea food during cold storage

Type of sea food	Storage temperatures (degrees)	Sensory scores(mean values)	conjugated dienes (mean values) (n mole/mg)	Correlation coefficient(r )
Mackerel	0	5.616	0.065	0.966
	4	6.550	0.069	0.944
	-10	5.780	0.049	0.908
Horse mackerel	0	6.550	0.046	0.905
	4	5.644	0.054	0.807
	-10	5.472	0.030	0.904
Silver hake	0	5.524	0.048	0.971
	4	5.592	0.050	0.877
	-10	5.640	0.056	0.840
shrimp	0	5.604	0.054	0.888
	4	5.300	0.039	0.873
	-10	6.527	0.051	0.887

**Table 10:** Correlation coefficient between sensory scores and thiobarbetic acid number of the examined sea food during cold storage

Type of sea food	Storage temperatures (degrees)	Sensory scores (mean values)	thiobarbetic acid number (mean values) (n mole/mg)	Correlation coefficient(r )
Mackerel	0	5.616	2.494	0.931
	4	6.550	2.524	0.876
	-10	5.780	2.919	0.908
Horse mackerel	0	6.550	2.921	0.891
	4	5.644	2.222	0.922
	-10	5.472	2.463	0.889
Silver hake	0	5.524	1.858	0.797
	4	5.592	2.762	0.898
	-10	5.640	2.352	0.891
shrimp	0	5.604	2.205	0.882
	4	5.300	1.984	0.807
	-10	6.527	3.980	0.758

## DISCUSSION

In general, spoilage can be described as both a process (all deteriorative processes) and a result (result in products becoming unacceptable for human consumption). Fish and other sea food spoil very quickly, if they are not handled and treated properly. On the other hand it is possible to preserve fresh raw fish, so that it remains fresh just like when it caught for a long period of time, the four key factors to monitor for preserving the freshness of fish are time, temperature, care and hygiene.

Our results indicated that Mackerel fish stored at 0 °C (held on ice), for one day had good quality. However, at the second and fourth days of icing it showed a decrease in quality but was otherwise of acceptable quality, while at sixth day of storage, fish had completely spoiled. At the same time, mackerel fish stored at 4 °C in a refrigerator showed excellent quality at first day of storage, while it was at a border line quality at the second day and it became putrid, at the third day.

In case of mackerel fish stored at -10 °C it retained its excellent quality till three months and there was a decrease in quality at sixth month and complete spoilage was observed at 12<sup>th</sup> month (Table 1).

Mackerel fish is a fatty fish (a fat range through out the year is 6-23%) and is not normally gutted at the sea, therefore spoil quickly unless, they are chilled immediately after catching and kept chilled. The off odour of Mackerel with a fat content of 10%, will developed after 1-2 days, at 10 °C and will be putrid after 5-6 days, Fish with a high fat content spoil even faster. Mackerel of medium fat content stowed in ice or in refrigerated sea water immediately after capture will be in a good condition for 4-5 days (Smith, *et al.*, 1980, Suhendan, *et al.*, 2004).

Results of TBA values for Mackerel increased sharply between day 2 and day 3 and then fluctuated with rapid increases and decreases. Some of these changes may be due to sample variation and may reflect the usefulness of the test in the early stage of oxidation (Helen, 1989).

It was reported that mackerel fish stored at -18 °C for 10 months was rejected at the 10<sup>th</sup> month of the storage in the sensory evaluation (Alya and Ekim, 1999).

Storage of krill fish at 0-2 °C after 6-8 hours of storage, a dark color started in the head, legs and spread slowly to the tail within 24 hours. After 72 hours, krill fish became inedible due to ammoniacal odor and flavour (Connell, 1990).

The total volatile nitrogen contents of mackerel fish stored at 0 °C, 4 °C and -10 °C showed gradual increase from 24.84 mg/100gm to 64.14 mg/100gm until day six of icing, from 24.84 mg/100gm to 61.54 mg/100 gm until day three of refrigeration and from 24.84 mg/100gm to 69.28mg/100gm until tenth month of frozen storage, respectively (Table1).

It was found that total volatile nitrogen contents of Mackerel fish began to increase sharply around day 2 and continued rising around day 6 at 0-4 °C in a refrigerator (Helen, *et al.*, 1989).

It was reported that a large increase in TVBN in Mackerel after 10 days chilling (Gill, *et al.*, 1992).

Results of Table (1) indicated that mackerel fish stored at -10 °C for 12 months showed the highest mean concentration of free fatty acids and acid value (3.3600ml/gm, 6.72ml/gm), while fresh fish, had the lowest (1.194ml/gm, 2.388 ml/gm) (Table1). At the same time, fresh Mackerel fish showed the highest mean concentration of conjugated dienes (CD) (0.122 nmole/mg) while mackerel stored at -10 °C for 12 months had the lowest (0.009 nmole/mg). However, TBA mean concentration was increased from 0.806mgMA/kgm of fresh fish to 4.194 mgMA/kgm of mackerel stored at 0 °C for 6 days, to 4.786 mgMA/kgm of fish stored at 4 °C for three days and to 6.348mgMA/kgm of fish stored at -10 °C for 12 months, respectively (Table1).

Results of sensory analysis of the iced salmon indicated that both the pink and coho salmon retained their excellent quality (prime quality) characteristics for about 8 days post harvest. The chemical indicator for oxidative rancidity (TBARS) in premium grade iced pink and coho salmon do not exceed 2.5 micromoles MA./100gm. After 3 months in frozen storage at -23 °C, the pink and coho salmon were of acceptable quality after 3 and 6 months, respectively but less than premium quality because of rancidity. About 9 days into the experiments, the iced pink salmon showed loss of quality. After 14 days, it showed complete signs of spoilage (poor quality). In case of iced coho salmon, loss of quality appeared after 9 days on ice and complete spoilage after 13 days on ice. It was also found that between 9,11 days, there was an increase in TBA number of pink salmon (indicating that the additional time on ice created a window of susceptibility that permitted increased oxidation of fatty acids in the oils of pink salmon). Low TBA numbers found in the coho salmon indicated that, as expected, oxidation of lipids was not as much of a probable as it was with the pink salmon, at the same time, this was verified by taste panel evaluations which indicated no organoleptic

rancidity. By the end of three months, in frozen storage of pink salmon, obvious changes were detected in the quality, where the mean sensory score (mean rancidity score was higher (2.5) than that of the corresponding one at the beginning of storage (first 3 months (1.1). After 9 months, TBA was 9.8 micromoles MA/gm in iced pink salmon of low quality, indicating that rancidity influenced flavor and odor scores. The excellent quality had TBA of 2 micromoles/100gm or less (Barnett, *et al.*, 1991).

From the data presented in table (2), it was evident that the quality of iced Horse mackerel fish (held on ice) was considered excellent through the first 5 days and slightly decreased (at the border line quality) after five days, while at the fifteenth day of storage, it showed complete signs of spoilage. At four degrees of storage, Horse mackerel fish showed excellent quality after one day of storage, border line quality after three to four days and bad quality (complete spoilage) after sixth day. Meanwhile, at  $-10^{\circ}\text{C}$  of storage, fish started to lose its quality after three months and became putrid at 13<sup>th</sup> month of storage.

It was found that the frozen storage of Horse mackerel fish at  $-20^{\circ}\text{C}$  allows greatly increasing shelf life to 12 months (Eymard, *et al.*, 2003).

It was found that cod fish began to show organoleptic signs of spoilage on day four and was listed as unsatisfactory by day nine (Smith, *et al.*, 1980).

Sensory changes are those perceived with the senses, i.e., appearance, odour, texture and taste. The first sensory changes of fish during storage are concerned with appearance and texture. The characteristic taste of the species is normally developed the first couple of days during storage in ice. (Howgate, 1994).

It was indicated that Horse mackerel fish showed a gradual increase in total volatile nitrogen (T.V.N.) content from 19.89mg/100gm to 64.34mg/100gm at 15<sup>th</sup> day of ice storage, from 19.89mg/100gm at zero time of fish storage to 71.80mg/100gm at 6<sup>th</sup> day of storage at  $4^{\circ}\text{C}$  and from 19.89mg/100gm at zero time to 69.8mg/100gm at 13<sup>th</sup> month of storage at  $-10^{\circ}\text{C}$ . In case of lipid hydrolysis parameters (F.F.A), fresh Horse mackerel fish had the lowest mean value of 0.554ml/gm, while Horse mackerel fish stored at  $-10^{\circ}\text{C}$  for 13 months, showed the highest mean value of 3.3ml/gm (Table 2).

It was found that the total volatile basic nitrogen (TVBN) content of sardine fish did not differ during the 9 days of storage at  $0^{\circ}\text{C}$ . At the 16<sup>th</sup> days, an increase was observed indicating the end of lag phase of



microorganisms (Gracia and Careche, 2002). The free fatty acid content at 0 °C was increased after 3 days of storage of sardine and then remained fairly constant till the end of storage. It was also found that conjugated dienes increased after one day of storage at 0 °C and after that remained quite constant. It was concluded that CD measurements has been used satisfactory as a quality determination for the initial changes. At the same time, the TBA index (TBA -I) increased on day 3 at zero but decreased in sardine on day 16, TBA like CD are relatively unstable, capable of interacting with constituents (Santiago, *et al.*, 1997).

Several chemical and biochemical deteriorative reactions were reduced in super chilled fish (storage at 0 to -4 °C) including mackerel, sea bass, rainbow trout, carp (Aleman, *et al.*, 1982).

It was indicated that herring fish stored in pure ice only for 3 days, showed an increase in total volatile nitrogen (TVN) content from 20.1mg/100gm in fresh samples to around 50mg/100gm in iced fish at the third day of storage (Huss, 1995).

Levels of total volatile basic nitrogen (TVBN) in sardine, Atlantic horse mackerel, Chub mackerel and Atlantic mackerel during ice storage and storage at room temperature, were increased gradually in all the fish species as decomposition progressed, regardless of storage temperature (Mendes, 1999).

Results of Table (2) revealed that the acid value of Horse mackerel fish was ranged from 1.108ml/gm at zero time of storage to 7.60ml/gm at 15<sup>th</sup> day of storage at 0 °C it was ranged from 1.108ml/gm at zero time of storage to 5.050 ml/gm of storage for 6 days at 4 °C while acid value was ranged from 1.108ml/gm at zero time to 6.6 ml/gm at 13<sup>th</sup> month of storage at -10 °C. Concerning lipid oxidation parameters (CD) it decreased from 0.050 n mole/mg at zero time to 0.0190 n mole/mg at 15<sup>th</sup> day of storage at 0 °C from 0.050 n mole/mg to 0.045 n mole/mg at 6<sup>th</sup> day of storage at 4 °C and from 0.05 n mole/mg at zero time to 0.010 n mole/mg at 13<sup>th</sup> month of storage at -10 °C. The secondary lipid oxidation parameters (TBA) in Horse mackerel fish reached to the highest mean levels of 6.936mgMA/kg, 4.762 mgMA/kg, 4.969 mgMA/kg at time of spoilage during storage at different temperatures (0,4,-10 °C), respectively. It was reported that the thiobarbituric acid reactive substances decreased at the end of frozen storage of Horse mackerel fish, this was related to interaction and decomposition between lipid oxidation products and proteins (Eymard, *et al.*, 2003).

Fish Scad (*Trachurus trachurus*) was stored at 4 °C after 7 days of storage, TVBN was 50.4 mg/100gm (Sengor, *et al.*, 2000).

The lipid oxidation products (CD, TBA) were decreased in Horse mackerel after 12 months of frozen storage. The decrease was results from decomposition and interactions of lipid oxidation products with proteins (Eymard, *et al.*, 2003).

Lipid hydrolysis (free fatty acid content) and oxidation (conjugated dienes formation; thiobarbituric acid index, TBA-i) showed an increase during the frozen storage (-20 °C) for 270 days of an under-utilised medium-fat fish species (Horse mackerel; *Trachurus trachurus*). (Santiago, *et al.*, 2002).

The sensory analysis of Silver hake fish showed that the excellent sensory quality was observed at the first day of storage at 0 and 4 °C while border line quality was at 2<sup>nd</sup> day of storage at 0,4 °C respectively, and it showed signs of complete spoilage at 6<sup>th</sup> day of storage at 0 °C and at 5<sup>th</sup> day of storage at 4 °C. In case of frozen storage, after 1 month of storage, it showed excellent quality, while after 6 months, it was on a border line and at 14<sup>th</sup> month, it became unfit for human consumption (Table 3).

Silver hake fish showed the highest mean concentrations of TVN (77.52 mg /100gm) and FFA (4.63ml/gm) and acid value (9.260ml/gm) and TBA (5.56 mgMA/kg) at the 14<sup>th</sup> month of frozen storage. At the same time, it had the highest mean concentrations of CD (0.071 n mole/mg) at third month of storage at -10 °C while the lowest mean concentrations of TVN, FFA, AV, CD and TBA (18.78mg/100gm, 0.818ml/gm, 1.636 ml/gm, 0.052 n mole/mg and 0.924mgMA/kg respectively) were found in fresh fish (Table 3).

TVBN of guitar fish stored at -18 °C for 6 months were in 3 months intervals ranged from 19.87mg/100gm -48.62 mg/100gm. (Ayse, and Yilmaz, 2003). Minced sardine flesh stored at -10 °C showed the highest values of free fatty acids at -10 °C during the second month of storage (12.6ml/mg). The TBA value in minced sardine flesh increased from 5.5 to 23 mg of MA/kg in fourth month (Ortiz, and Bllo, 1992).

It was indicated that increasing the temperature from 0 to 5 °C at least double the rate at which cod and similar species spoil. AT the same time storage at 0-5 °C for fish caused decrease of TVBN at the beginning of storage, then at the end (complete spoilage), TVBN increased sharply (Botta, *et al.*, 1984).

It was repored that the longer the fish were stored in ice, the more influence oxidative rancidity had an over all acceptability (Huss, 1995).

It was found that the increase in T.V.N. in fish stored in ice for different days was most likely caused by autolytic processes (which

produce volatile amine compounds) and bacterial spoilage. T.V.N. in fish mainly contribute volatile nitrogenous compound ammonia which is produced by deamination of protein, peptides and amino acids (Huss, 1995).

Refrigerated storage of Mahi mahi fish fillets at 7 °C for 3 days showed TVBN of 30mg/100 gm, sensory scores were 6 to 6.5 (10 very fresh, 1 very spoiled) for odor, appearance, texture and color (Anastasio, *et al.*, 2004).

It was reported that the average TVBN value of cod fish stored at 0-4 °C on day 4 (it is the first day showed some signs of spoilage like odor), was 64 mg TVBN/100 gm. (Gill, 1992).

It was reported that there are some factors affecting spoilage rate like size of fish (fast in small size), post mortum PH (fast in high PH), fat content (fast in fatty fish), skin (fast in thin skin) (Olley and Ouarmby, 1981).

The most predominant lipid damage in different hake zones (Three light muscle zones and the dark muscle) during frozen storage at –11 and –18°C was hydrolysis, at the end of storage reaching values of about 40% (for the light muscle zones) and 12% (for the dark muscle) of the total lipids at –11°C. Significant ( $P<0.05$ ) correlation value ( $r=0.67-0.85$ ) relationships between the frozen storage time and the free fatty acid content (FFA) were obtained for the four muscle zones at both temperatures. However, the dark muscle showed a higher oxidation development (TBA- formation) as a result of a higher lipid content (Santiago, *et al.*, 1999).

Storage of fresh Mediterranean hake (in Spain) in ice and under refrigeration at home resulted in slight accumulation of volatile and biogenic amines in hake. when it was stored at 6-8 °C a significant production of TVBN was observed. Analysis revealed that hake stored in ice was inedible after 29 days and the figure for refrigerated hake being 20 days (Malle and Poumeryrol, 1998, Baixas, *et al.*, 2003).

Generally, fish stored at zero degrees kept longer than fish kept at 5 °C irrespective of species although it was not always apparent. In essence, the warm water fish had kept longer than cold water fish at chilled temperatures. (ICMSF1998).

Table (4) presented the summary of sensory and chemical quality evaluation of shrimp during cold storage. It was found that iced shrimp (held on ice) showed signs of spoilage after 6 days of storage, while refrigerated shrimp at 4 °C became putrid after one day and frozen one after one month. Results of chemical quality tests indicated that the

highest levels of TVN (118.30 mg/100g) was found in Shrimp stored at 0 °C for 6 days while the lowest was in fresh shrimp 30.96mg/100gm). In case of free fatty acids, fresh Shrimp showed the lowest (1.03ml/gm) while the highest (4.467 ml/gm) was in shrimp stored at -10 °C for 10 months. Concerning acid value, fresh Shrimp had the lowest value (2.060 ml/gm), while Shrimp stored for one month at -10 °C showed the highest (8.934ml/gm). Regarding lipid oxidation parameters (CD, TBA) the conjugated dienes decreased with the progress of spoilage during storage at different temperatures and the vice versa was true for TBA values.

It was reported that the whole Shrimp samples stored in a refrigerator (+4 °C) were found to be of excellent (prime) quality at day 0, good quality (less than excellent) at day 1 and spoiled after 2 days, according to findings of sensory, chemical and physical analyses. The total volatile nitrogen (mg/100gm) ranged from 22.95 mg/100gm to 109.15 mg/100gm, during storage for 4 days (Candan, *et al.* 2000).

T.V.N of 50mg/100gm in peeled Shrimp represent such an objective lower limit if a comparison with organoleptic assesment is made. The maximum storage period in ice was 7 days (Solberg and Nesbakken, 1981).

It was reported that 30 mgTVBN/100gm in shrimp meat used as indicator limit for Shrimp acceptability (Solberg and Nesbakken, 1981). Thanae, *et al.*, (2003) found that the TVBN mean values of farm, fresh marine (brackish) and frozen shrimp (imported) were 2.542 mg/100 gm, 14.182 mg/100gm, 27.16mg/100gm., respectively. Otherwise, frozen Shrimp had the highest significant values which attributed to denaturation of protein during frozen storage. At the same time, TBA values were 0.801mg MA/kg for frozen shrimp, 0.308 for farm Shrimp and 0.182 mg MA/kg for marine fresh Shrimp (Thanae, *et al.*, 2003).

Triplicate sensory evaluations of each decomposition increment at 0,12,24,36 °C were conducted by three FDA sea food sensory experts. Sensory and chemical evaluation of Shrimp showed that decomposition more rapidly progressed at high temperatures than at low temperatures. It were considered to be decomposed by sensory evaluation on day 14 at 0 °C (Benner and Otwell, 2003).

Total volatile bases (TVB) of fish stored at low temperature, showed an increase, particularly at 5°C; at this temperature, values above 30 mgN/100g were reached after 5 days storage at 5°C (Shamsad, *et al.*, 1990, Huss, 1993, Holt, 1994,).

It was found that white Shrimp (*penaeus setiferus*) stored on ice, at 4 °C in refrigerator at zero time showed TVN of 18mg/100gm. (Solbergan and Nesbakken, 1981)

Whole Shrimp raw, will keep in good condition in cold storage at -30 °C for at least 6 months, individually frozen whole shrimp will keep for 3-4 months in good condition at -20 °C and only one month at -10 °C (Fatima, *et al.*, 1988 and Candan, *et al.*, 2000).

In the present study, the results of table (5) showed that the shelf life of the examined sea food during cold storage, where the shelf life of Mackerel fish was 6days, 3 days for storage at 0,4 °C and 12 months for storage at -10 °C respectively. In case of Horse mackerel fish, the shelf life was 15,6 days for storage at 0 and 4 °C respectively. While storage of Horse mackerel at -10 °C had shelf life of 13 months. Regarding Silver hake fish, it had shelf life of 6 and 5 days of storage at 0 and 4 °C respectively. while storage at -10 °C Silver hake fish showed shelf life of 14 months. At the same time, Shelf life of Shrimp was 6 days, 1 day and one month for storage at 0,4 and -10 °C respectively. The shelf life of stuffed rain bow trout during cold storage, (refrigerator) was to be 5 days. (Baygar, *et al.*, 2002).

The microflora responsible for spoilage of fresh fish changes with changes in storage temperature. Storage of fish at temperatures between 0°C and -4°C is called superchilling or partial freezing. The shelf life of various fish and shellfish can be extended by storage at subzero temperatures. Superchilling extends the shelf life of fish products. The technique can be used, for example where productive fishing grounds are so far from ports and consumers that normal icing is insufficient for good quality products to be landed and sold. (Aleman, *et al.*, 1982).

It was found that the shelf life of Silver hake stored at 0 °C (held in ice) was 4-5 days (Hiltz, *et al.*, 1976).

Samples of lean fish, fatty fish, and black tiger shrimp were tested using sensory evaluation parameters. They were preclassified 1-3 by odor, color and texture of the muscle tissue (class 1 indicates excellent quality, class 2 has border line quality and class 3 showed bad quality. (Christopher, *et al.*, 1997).

Fish (especially the oily varieties) transported for four weeks at -18 °C will lose the equivalent of 16 weeks of shelf life due to the high transport temperature. Conversely, if the fish is transported for the same four weeks at -35 °C, it loses only two weeks of shelf life. Many times, frozen fish is stored at the destination for a considerable amount of time in anticipation of market price shifts. Extended shelf life at premium

(excellent) quality is an important factor in the business. Since it is possible that a product will stay in storage longer than originally intended, it is generally safer to use the lower recommended temperature (Jorgensen, *et al.*, 1988).

The shelf life study of Shrimp stored in ice was over a 9 days period (Candan, *et al.*, 2000).

Results of correlation between sensory scores and TVN value (Tables 7, 8, 9, 10) of the examined sea food during storage period at different temperatures (0,4,-10 °C) indicated the presence of a strong correlation ( $r=0.826-0.987$ ) (Table 6). however, there was a strong correlation with FFA ( $r=0.823-0.946$ ). sensory scores also correlated well with acid value ( $r=0.700-0.970$ ) In case of CD and TBA as a lipid oxidation products, they were positively correlated with sensory scores ( $r=0.807-0.971, r=0.758-0.931$ )

Several reports by Hovland and Taylor (1991) speak of some correlation between TBA-RS and sensory assessments, but other authors fail to find a correlation (Connell, *et al.*, 1975). Thus, caution is necessary in interpretation of TBA-RS values into measures of sensory quality. Examples of guidelines for TBA-RS-values: foods with TBA-RS above 1-2  $\mu\text{mol}$  MDA-equiv per g fat (Connell, *et al.*, 1975) or above 10,  $\mu\text{mol}$  MDA-equiv per 1 kg fish (Ke and Woyewoda, 1976) will probably have rancid flavours.

There was insignificant correlation between sensory rancid odor and thiobarbituric acid test results where there was an increase in TBA values in Mackerel, Horse mackerel and Silver hake without development of rancid odor (Kolakowska and Denty, 1983).

It was reported that the lipid oxidation parameters (TBA, FFA) were increased significantly in Chub mackerel (*Scomber japonicus*) stored at  $-18\text{ }^{\circ}\text{C}$  for 10 months. The sensory scores of the samples were in the same parallel with chemical results with respect to flavour and general acceptability (Alya and Ekim, 1999).

It was found that the mean sensory scores (flavor) and corresponding mean TBA numbers for the iced pink salmon correlated well ( $r=0.87$ ) suggesting that rancidity had as much to do with loss of quality as microbial and chemical degradation, at the mean time, low levels of TBA of coho salmon, correlated with results of sensory evaluation indicating that, the presence of low levels of chemical rancidity did not greatly influence the overall quality of the coho salmon.

The greatest increases in chemical parameters (TVN, TBA) were found between days 2 and 4 of flounder fish stored at  $5\text{ }^{\circ}\text{C}$ . After 4 days

of storage, the flounder was considered spoiled by organoleptic criteria. The results also showed that although the chemical and organoleptic measures of spoilage overlapped, organoleptic indication of spoilage lagged behind the chemical markers (Gill, 1992).

It was stated that TVBN of gutted mediterranean hake (*Merluccius merluccius* var *Mediterrancus*) stored for up to 20 days in ice in different seasons of the year, was not correlated with the time of ice storage and proved to be better as a spoilage index than a freshness index (Baixas, *et al.*, 2003).

## REFERENCES

- Aleman, M.P; Kaluda, K. and Uchiyama, H. (1982):* Partial freezing as a means of keeping freshness of fish. Bull. Tokai Reg. Fish. Res. Lab. 106, 11-26
- Alya, S. and Ekim, S. (1999):* Effect of glazing storage time on lipid oxidation of frozen chub mackerel (*scomber Japonicus*). Turk. J. Vet. Animal. Sci. 23, 575-584.
- Anastasio, A.; Vollano, L.; Visciano, P.; Miranda, E.; Antoine, F.R.; Weicl, Otwell, W.S.; Sims, C.A.; Little., Hogle, A.D. and Marshall, M.R. (2004):* Chemical and sensory evaluation of Mahi-Mahi(*Cryphaena hippurus*) during chilled storage. J. Food. Prot. 67(10): 2255-2262.
- Ayse, B. and Yilmaz, D. (2003):* Determination of proximate composition, quality changes in the common Guitar fish during cold storage. Turk. J. Vet. Animal Sci. 27: 207-212.
- Baixas, N.; Bover, S.; Veciana, N. and Vidal, M.C. (2003):* stability of volatile amines as freshness indexes for iced Mediterranean hake. J. Foods Sci., 68 (5): 1607-1610.
- Barnett, H.J.; Nelson, R.W. and POYSK, F.T. (1991):* A comparative study using multiple indices to measure changes in quality of pink and coho salmon during fresh and frozen storage. Utilization Research Division, North west fisheries center National marine fisheries services, National oceanic and Atmospheric Administration 2725 Mont lake Boulevard East, seattle, WA98112.PP::1-28
- Baygar, T.; Nuray, E.; Metin, S. and ozkan, O. (2002):* Determination of shelf life of stuffed rain bow trout during cold storage. Turk. J. Vet. Animal Sci. 26: 577-580.

- Benner, R.A. and Otwell, W.S. (2003):* Evaluation of putrescine, cadaverine and indole as a chemical indicator of decomposition in Penacid shrimp. *J. Food Science.* 86(7): 312-320.
- Botta, J.R.; Lauder, J.T. and Jewer, M.A. (1984):* Effect of methodology on total volatile basic nitrogen (TVBN) determination as an index of quality of fresh Atlantic cod (*Gadus morhua*). *J. Food Sci.* 49, 734-736, 750.
- Botta, J.R. (1995):* Evaluation of sea food freshness quality. VCH Publishers Inc.
- Candan, V.; Tacnur, B.; Ozhan, O. and Mentin, S. (2000):* Sensory evaluation and determination of some physical and chemical characteristics of shrimp during cold storage. *Turk. J. Vet. Animal. Science*, 24: 181-186.
- Christopher, P.; Ellis, Mary, L. (1997):* Statistical classification of sea food quality. *journal of A.O.A.C. Int.* 80(6):1347-1353.
- Connell, J.J. (1975):* Control of fish quality. Fishing News (Books) Ltd., Farnham, Surrey, UK.
- Connell, J.J. (1990):* Control of fish quality. Oxford: Fishing News Books 3 rd ed .
- Eymard, S.; Genot, C.; Rampon, V. and Chopin, C. (2003):* Detection of biochemical changes during frozen storage of horse mackerel surimi. *Trans. Atlantic. Fisheries conference, Reyk Javik, Iceland, june 10-14.*
- Fatima, R.; Khan, M.A. and Qadri, R.B. (1988):* Shelf life of shrimp (*Penaeus merguensis*) stored in ice (0&deg;C) and partially frozen (-3&deg; C). *J. Sci. Food Agric.* 42, 235-247.
- Gill, T.A.; Conway, J. and Evrovski, J. (1992):* Biochemical and chemical indices of seafood quality. In: H.H. Huss, M. Jacobsen and J. Liston (eds.) *Quality Assurance in the Fish Industry. Proceedings of an International Conference, Copenhagen, Denmark, August 1991.* Elsevier, Amsterdam, 377-388.
- Gram, L. (1992):* Evaluation of the bacteriological quality of seafood, *J. Food Microbiol.* 16, 25-39.
- Gracia, R. and Careche, M. (2002):* Influence of chilling methods on quality of sardines (*Sardina pilchardus*). *J. Food Prot.*, 65 (6):1024-32.



- Holt, J.G.; Krieg, N.R.; Sneath, P.H.A.; Staley, J.T. and Williams, S.T. (1994):* Bergey's manual of determinative bacteriology. Williams & Wilkins, Baltimore, 787p.
- Helen, T.; MacCarthy, P.; Christophe, E.; Marry, L. and Barbara, M. (1989):* Comparison of volatile acid number test with Enzymatic Acetic acid Assay for Assesment of sea food quality. J.A.O.A.C. 72(5): 828-833.
- Helen, T.; MacCarthy, P.; Christopher, E.; Marryl Hiltz, D.F.; Lall, B.S.; Lemon, D.W. and Dyer, W.J. (1976):* Deteriorative changes during frozen storage in fillets and minced flesh of Silver Hake (*Merluccius bilinearis*) processed from round fish held in ice and refrigerated sea water. J. Fish. Res. Board Can. 33, 2560-2567
- Hovland, D.V. and Taylor, A.D.J. (1991):* A Review of the Methodology of the 2-Thiobarbituric Acid Test. Food. Chem. 40, 271- 291.
- Howgate, P. (1994):* proposed draft guideline for the sensory Evaluation of fish and shellfish. Joint of FAO/WHO Food standard programes. Codex Committee on fish fishery products, twenty first session, Bergen, Norway.
- Hultin, H.O. (1992):* Biochemical deterioration of fish muscle. In Huss, H.H., Jakobsen, M and Liston, J.eds.
- Huss, H.H. (1993):* Assurance of seafood quality FAO-DANIDA, Rome, 169p.
- Huss, H.H. (1995):* Quality and quality changes in fresh fish. FAO Fisheries Technical paperNo. 348.Rome:FAO.
- ICMSF (1998):* International Comission on Microbiological Specifications for Foods. Micro-organisms in Foods.: Microbial Ecology of Food Commodities. London: Blackie, Academic professional.
- Jorgensen, B.R.; Gibson, D.M. and Hus, H.H. (1988):* Microbiological quality and shelf life prediction of chilled fish. Int. J. Food Microbiol. 6, 95-307.
- Ke, P.J. and Woyewoda, A.D. (1976):* Micro determination of thiobarbetic acid values in marine lipids by a direct spectrophotometric method with a monophasic reaction system. Anal. Chem. Acta. 106, 279-284.
- Kolakowsk, A. and Denty, J. (1983):* The usefluness of 2 thiobarbetic acid test for the evaluation of rancidity of frozen fish. Nahrung.27(5): 513-518.

- Kraft, A.A. (1992):* Psychrotrophic Bacteria in foods: Disease and spoilage, Ranton, Florida: CRC. press Inc.
- Li, C.T.; Wich, M. and Marriott, N.G. (2001):* Evaluation of lipid oxidation in animal fat. Bull.Ohio State university,Research and Reviews: meat special circular 172-199.
- Malle and Poumeryrol (1998):* Anew chemical Criterion for the quality control of fish: TMA/TVBN(%).J .Food.Prot. 52(6):419-423.
- Mayer, B.K. and Ward, D.R. (1991):* Microbiology of fin fish and fin fish processing.In Ward.D.R. and Hackney,C.eds.microbiology of marine food products, New york.Van Nostrand Reinhold.
- Meilgaard, M.; G.V. Civille and Carr, B.T. (1991):* Sensory Evaluation Techniques. 2<sup>nd</sup> ed. CRC Press, Bocan Raton, FA, USA.
- Mendes, R. (1999):* Changes in biogenic amines of major portuguese blue fish species during storage at different temperatures. journal of food biochemistry .23(1) :33-43.
- Nielsen, J. (1995):* Sensory changes. In Huss,H.H. (ed).Quality and Quality changes in fresh fish.FAO Fisheries Technical Paper, No.348, Rome, FAO.PP.35-
- Olley, J. and Quarmby, A.R. (1981):* Spoilage of fish from Hong Kong at different storage temperatures. 3. Prediction of storage life at higher temperatures, based on storage behaviour at 0&deg;C, and a simple visual technique for comparing taste panel and objective assessments of deterioration. Trop. Sci. 23, 147-153.
- Ortizh, Bello, R. (1992):* Composition and Stability of fatty acids from deboned sardine meat during freezer storage. Arch. Latnoam. Nutr.42(4):460-6.
- Pearson, D. (1970):* Chemical analysis of foods, chemical publishing CO., INC., New york, NY.
- Santiago, P.; Aubourg, Maurizio, U. and Carmen, G. (1997):* Quality assesment of sardine during storage by measurement of fluorescent compounds.European Food Research and technology. 62(2): 295-298.
- Santiago, P.; Aubourg, Maurizio, U. and Carmen, G. (1999):* Differential lipid damage in various muscle zones of frozen hake (Merluccius merluccius). European Food Research and Technology. 208( 3) :189 – 193.
- Santiago, P.; Aubourg, Maurizio, U. and Carmen, G. (2002):* Effect of brine pre-treatment on lipid stability of frozen horse mackerel (Trachurus trachurus) European Food Research and Technology., 215, (2): 91-95.

- Shamsad, S.I.; Nisa, K.; Riaz, M.; Zuberi, R. and Quadri, R.B. (1990):* Shelf life of shrimp. (*Penaeus merguensis*) stored at different temperatures. *J. Food Sci.* 55(5):1201-1205.
- Sengor, F.; Celik, U. and Sevilsen, A. (2000):* Determination of freshness and chemical composition of scad (*Trachurus trachurus*) stored in a refrigerator. *Truk. J. Vet. Animal. Sci.* 24:187-193.
- Singh, R.P. (2000):* Scientific principles of shelf life evaluation, pp.3-22, In D.Man and A.jones, (eds.), shelf life evaluation of foods, 2<sup>nd</sup> Edition, Aspen publishers, INC.Gaithers burg, M.D.
- Smith, J.M.; Hardy, R. and Younk, K.W. (1980):* Seasonal study of the storage characteristics of mackerel stored at chilled and ambient temperatures. In, Connell, J.J. ed. *Advances in fish science and technology. Jubilee conference of Torry Research 23-27 July 1979.* Oxford, England: Fishing News.
- Solberg, T. and Nesbakken, T. (1981):* Quality changes in iced shrimps (*pandalus borealis*): indole and Phin shrimps caught in the Barents sea compared with shrimp caught in the Far East. *Nord. Vet. Med.* 33(9-11): 446-453.
- Suhendan, M.O.L.; Ozkan, O. and Nuray, E. (2004):* Determination of the quality parameters of imported mackerel under different thawing conditions. *Turk.J.Vet.Anim.Sci.*,28:1071-1077.
- Thanae, M.; Amine., Mona. O.; Abu, El.Nile. and Azza, M. Aboul wafa. (2003):* Quality of fresh and frozen shrimp in alexandria city. The third international scientific conference. Mansoura, 29-30 April, 2003.
- Winger, R.J. (2000):* Preservation Technology and shelf life, pp.73-86 In Man, D.A., Jons, (eds), shelf life evaluation of foods, 2<sup>nd</sup> Edition, Aspen publishers, Inc. Gaithers burg, M.D.
- Zdzislaw, E.S.; Kolakowska, A. and Burt, J.R. (1990):* Post Harvest Biochemical and microbial changes. In Zdzislaw, E.S. ed. *Sea food resources nut. Composition and preservation*, PP.70-71. Academic Press.