

T.C.
FIRAT UNIVERSITY
INSTITUTE OF NATURAL AND APPLIED
SCIENCE



**DETERMINATION OF SHELF LIFE DURING COLD
STORAGE (+4±1 C°) OF PIKE BARBELL (*Luciobarbus
esocinus* Heckel, 1843) FISH FINGERS COATED
ADDITION OF GOJI BERRY**

Master Thesis
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JULY – 2019

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In this study, it was aimed to investigate the chemical, microbiological and sensory changes that occur during the storage of Pike Barbell (*Luciobarbus esocinus* Heckel, 1843) fish fingers coated addition of goji berry at $4\pm1^{\circ}\text{C}$ and to determine the shelf life of this product.

The Success of any kind in any person's life is the outcome of the sincere and combined efforts of a group of persons.

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SUMMARY

In this study, the shelf life of Pike Barbell (*Luciobarbus esocinus* Heckel, 1843) fish fingers, that ordinarily packed and stored at $4\pm 1^{\circ}\text{C}$. For this purpose, the moisture, ash, fat, protein, pH, TVB-N and TBA count, total aerobe microorganism (at 5°C and 30°C incubation temperature) and yeast-mold counts of both the material and the experimental groups were investigated. The sensory, chemical and microbiological changes of the products obtained in the research under cold storage conditions were also examined, analyses were performed every 3 days, and the study was conducted in two parallel analyses with two repetitions.

The material of the research consisted of Pike Barbell (*Luciobarbus esocinus* Heckel, 1843) obtained from the Keban Dam Lake. Together with the control group, three experimental groups fish finger were obtained with the addition of the goji berry extract into this content by 1% and 2%. Samples were then stored in styrofoam packs at $4\pm 1^{\circ}\text{C}$.

When the data obtained according to the results of nutrient composition analysis were analyzed statistically; the difference between fish finger groups was not significant ($p>0.05$) while the difference between the samples in terms of the amount of protein, fat and ash was found to be significant ($p<0.05$). The difference in TVB-N, TBARs values was significant in all groups during storage ($p<0.05$).

When the data obtained from microbiological analyzes were evaluated, it was found that the difference between the groups in terms of total aerobe mesophile and yeast-mold counts was significant in the control group and the samples with the addition of goji berry extract ($p<0.05$).

According to the results obtained from the sensory analysis, the difference between the groups in terms of general appreciation level was found to be significant ($p<0.05$).

The shelf life of fish fingers was determined as 9 days in group A (control group) and 12 days in group B and C with the addition of goji berry extract when the analyses of the study were assessed.

Finally, it was concluded when the data obtained from the analyses of the study were assessed that goji berry extracts added to fish fingers had a positive effect on the shelf life.

Keywords: *Luciobarbus esocinus*, Goji berry, Cold storage, Shelf life

ÖZET

Kurt Üzümlü İlaveli Kaplamalarla Kaplanmış Bıyıklı Balık (*Luciobarbus Esocinus* Heckel, 1843) Dilimlerinin Soğuk Muhafazada ($+4\pm 1$ °C) Raf Ömrünün Belirlenmesi

Bu çalışmada, *Luciobarbus esocinus* (Heckel, 1843)'tan hazırlanan balık çubukları (fish finger) adi paketlenerek 4 ± 1 °C'de muhafaza edilmiştir. Çalışmada ham materyalin ve deneysel grupların nem, kül, yağ, protein, pH, TVB-N, TBA sayısı, toplam aerob mikroorganizma (5 °C ve 30 °C inkübasyon sıcaklığında) ve maya-küf sayıları belirlenmiştir. Araştırmada elde edilen ürünlerin soğuk depolama koşullarında duyuusal, kimyasal ve mikrobiyolojik değişimleri incelenmiş, analizler 3 günde bir, çalışma iki tekerrürlü analizler iki paralelli olarak yapılmıştır.

Araştırmanın materyalini Keban Baraj Gölü'nden avlanarak elde edilen *Luciobarbus esocinus* teşkil etmiştir. %1 ve %2 oranlarında goji berry ekstraktı ilave edilerek kontrol ile birlikte üç deneysel balık çubuklarında oluşan grup elde edildi. Daha sonra örnekler strafor paketlerde 4 ± 1 °C'de muhafazaya alınmıştır.

Besin bileşimi analiz sonuçlarına göre edilen veriler istatistiksel olarak analiz edildiğinde; balık fish fingerlerinin gruplar arasındaki farklılığın önemli olmadığı tespit edilirken ($p>0.05$), protein, yağ ve kül miktarı bakımından ise örnekler arasındaki farklılıkların önemli olduğu belirlenmiştir ($p<0.05$). Muhafaza süresince tüm gruplarda TVB-N, TBA değerlerindeki farklılığın önemli olduğu belirlenmiştir ($p<0.05$).

Mikrobiyolojik analizler sonucu elde edilen veriler değerlendirildiğinde kontrol grubu ve goji berry ekstraktı ilaveli örneklerde toplam aerob mezofil ve maya-küf sayıları bakımından gruplar arasındaki farklılığın önemli olduğu tespit edilmiştir ($p<0.05$).

Duyuusal analizlerden elde edilen sonuçlara göre genel beğeni düzeyi bakımından gruplar arasındaki fark önemli bulunmuştur ($p<0.05$).

Çalışmada yapılan analizler değerlendirildiğinde balık çubuklarının raf ömrü A grubunda (kontrol grubu) 9 gün, goji berry ekstraktı ilaveli B ve C grubunda 12 gün olarak belirlenmiştir.

Sonuç olarak çalışmada yapılan analizler neticesinde elde edilen veriler değerlendirildiğinde balık fish fingerlerine ilave edilen goji berry ekstraktlarının raf ömrü üzerinde olumlu etki gösterdiği kanaatine varılmıştır.

Anahtar Kelimeler: *Luciobarbus esocinus*, Goji berry, Soğuk muhafaza, Raf ömrü

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1. INTRODUCTION

Nutrients come with an irreplaceable put in place human life. Nutrients that people consume in daily nutrition are necessary for treatment and repair of the body, achieving its need of energy and organizing metabolic activities. Nutrients generally consist of water, protein, fat, carbohydrate, minerals and vitamins. Some of the components in nutrients might be produced in the body, can be replaced with other materials or replaced by other issues. However, there are certain nutrients which do not can be found in our body at all or cannot be synthesized properly and can not be changed by another compound and they are essential compounds necessary for life. There are various fatty acids, amino acids, vitamins and minerals in this group (Gorga, 1998; Nettleton, 1992).

With more than 30,000 species (33,700 species, 322,900 names) is the biggest set of determined species in the animal kingdom. Very few of these (700 types of fish and 200 types of mollusks and shellfish) are used for making food (Fish Base, 2018). The production of seafood, shellfish, crabs, mollusks and other marine animals has been increasing constantly throughout the world, reaching 1709 million tons in 2016 (FAO, 2018). It has been verified by studies completed in various areas that individual feeding behaviors as well as what we eat essential factors in our health, which excontrols why people know about the necessity to eat healthy. Seafood, shellfish and other seafood are usually important in maintaining a healthy and nutritional diet.

Fish meat is essential due to its unique composition and therefore considered quality food for individual consumption. High-protein material, as well as essential amino acid profile and less stroma, make it easily digestible. It really is an important raw material to make different foods (Akkuş et al., 2004). Unsaturated essential fatty acids, vitamins and minerals are regarded as abundant in these meats (Aitken et al., 1982; Gülyavuz and Ünlüsayın, 2002). Seafood products are extremely important for human nourishment, however, its financial value isn't that much higher based on the current situation.

The literature on fishery fast foods is quite new. Concerns regarding the safety of synthetic food additives and the toxicity of such chemicals have lately been on a rise. The essential ingredients of coating materials largely consist of flour and water. Also taking into consideration the increasing demand for natural products, the protective effects of these materials and the shelf life of frozen products are needed to be studied. In line with this, the

current study aimed at the production of fish finger from deep flounder with and without coating and the evaluation of the chemical and sensory properties of these products during storage at -18 C for five months (Vareltzis et al., 1997).

Fishes are brought to our tables being processed in different ways in order to be able to maintain their quality and to be evaluated in different ways during the period which they are hunted. In addition, fishes become foodstuffs sought by such places as processed fishery products offer different aroma and alternative diversity for hotels and restaurants (Baygar et al., 2002).

The increasing population of the country and the desire consuming fish meat in a much wider regions than in all seasons and production regions has made it obligatory to process aquaculture. Meanwhile, nutritional habit and processed food sector are consistently developing in parallel with increasing urbanization and industrialization in our country, and raising level of income (Gökoğlu, 1994).

Fish-based or other seafood-based foods including cakes, crackers, burgers, fish fingers and marinated products are in large demand by consumers throughout the world. The stability of the quality of these fast foods have been studied before (Siaw et al., 1985; Ihm et al., 1992; Lazos et al., 1996).

It is known that microbial and chemical reactions lead to the quality deterioration of fish. Fish fingers are made from fish slices as battered and breaded foods. They are usually stored and marketed as frozen. However, frozen storage does not completely inhibit said reactions (Reddy & Srikar, 1996). These products, including fish burgers, fish fingers, fish balls, frankfurters and sausages, may be subject to undesirable changes during frozen storage, and these changes may cause deterioration and limit the storage time.

These undesirable changes derive from protein denaturation (Fijuwara et al, 1998; Benjakul et al, 2005), and lipid oxidation. Polysaccharides, proteins, and lipids edible coatings can lengthen the shelf life of foods by working as a solute, gas, and vapor barriers. Generally, meats and other foodstuffs are protected with dried out particles (breaded) or dipped in water solutions of the particles which called battering (Kilincceker et al, 2009; Ojagh et al, 2010).

Extracts are antiseptic, antioxidant, digestive stimulant antimicrobial and enzymatic effects as natural products used in food preservation. Plant extracts are a very large part of the GRAS list Goji berry (*Lycium barbarum*) or wolfberry in the world known as wolfberry but in our country, little-known Wolf is one of the highest nutritional value rape fruits. This

is a very powerful antioxidant fruit has been used for 2.000 years in the medical field in China. Goji berry high antioxidant activity of β -carotene and phenolic components have been found to have originated from. Other natural antioxidants present in Goji berries are tannin, lignin, and flavonoids (Kabara, 1991).

Goji berry is among the most nutritious fruits (Figure 1.1). It is a very strong antioxidant due to its β -carotene and phenolic compounds and other natural antioxidants in it such as tannin, lignan and flavonoids. In addition, China has been using goji berries in medicine for 2000 years.

Goji berries have the attraction of their red-yellow color, along with a group of lipophilic compounds (carotenoids). Previous works have marked carotenoids to be effective in preventing chronic diseases including cardiovascular and skin diseases (Kohlmeir and Lam, 1999; Qian et al., 2004).

Goji berries are used to produce juice, wine and tincture and in combination with meats and vegetables (Potterat, 2010).



Figure 1.1. Goji berry (*Lycium barbarum*)

Prosperity diseases have been on a rise in the last decades. This has necessitated intensified studies on the substances used in the prevention and treatment of such diseases. Most of these studies have demonstrated that lifestyle changes including a well-balanced diet and physical exercise may be effective in preventing many of such diseases (Amagase & Farnsworth, 2011; Marrazzo et al., 2014; Shashirakha et al., 2015). Current food analyses and clinical and epidemiological studies aim to look for foods with better health-promoting properties, as well as raw materials to be used to produce properly-engineered foods (Ahmad et al., 2015; Kmiecik et al., 2015). Bioactive food is another new field in health and nutrition

that is emphasized by many. According to the American Dietetic Association (ADA), bioactive ingredients are physiologically active components of foods or dietary supplements that are provided from animals and plants, that are safe for human consumption and that are shown to play a role in health (Saldanha, 2004). Studies show that bioactive compounds may satisfy many of the health-promoting functions in humans (Castro et al., 2005; Cho et al., 2013; Kris-Etherton et al., 2002;). In addition to lowering cholesterol levels and blood pressure, they can boost the immune system, help the regulation of blood glucose levels and hormonal balance, reduce weight, slow down ageing and can help neutralize cancer (Mao, 2011).

Goji yields an orange-red berry about 2 cm deep with an ellipsoid shape and a sweet-and-tangy flavor. Goji berry is the fruit of *L. barbarum* and *L. chinense* and belongs to the family of Solanaceae. This family ranges from tomato and potato to eggplant. The species of *L. barbarum* was named by Carl Linnaeus in 1753. The species of *L. chinense*, also known as “Chinese wolfberry” due to Chinese farmers seeing wolves eating them, was named by Philip Miller in 1768. These two species are native to Asia and mainly grow in the central north region called the Ningxia Hui Autonomous Region. Moreover, wolfberries grow in commercial volumes in the Xinjiang, Shaanxi, Gansu, Hebei and Inner Mongolia regions and in Japan, Korea, and Taiwan. China is the main supplier of wolfberry, where 95,000 tons of Goji fruits grow in areas totaling in 82,000 hectares. The rich chemical composition and medical properties of Goji berries have given them a key role in the traditional medicine of Chinese for over 2000 years. In addition, dried wolfberries have been traditionally cooked and used as food and herbal tea.

Goji berries are classified as superfruits due to their many nutrients and bioactive compounds. Goji berries are used to produce juice, wine and tincture and in combination with meats and vegetables (Gross, 2006; Potterat, 2010). The introduction of Goji to Europe is dated around the 18th century (Sopher, 2013). *L. barbarum* grows better in well-drained and slightly alkaline soils and sunny locations. *L. barbarum* has a germination temperature of 21–23°C, which is 15–25°C for *L. barbarum*. The plant has a high resistance to low temperatures. It typically bears fruit in 3 (Marczyński & Piotrowski, 2013).

Wolfberries are rich in nutrients with high biological activity, including polysaccharide complexes, carotenoids, phenylpropanoids, etc. Among these, polysaccharide complexes are the most important and the most commonly found compounds, usually present in the water-soluble form of highly branched *L. barbarum*

Polysaccharides (LBP), which have a molecular weight of 8–214 kDa and constitute 5–8% of the total dry form of Goji fruits. The fruit has a composition of six types of monosaccharides (arabinose, rhamnose, xylose, mannose, galactose, glucose), galacturonic acid and eighteen amino acids (Amagase & Farnsworth, 2011). Reis *et al.* (2014) reported the fermentation of goji berries to give a 87% decrease in sugars in 24 hours and with no loss in polyphenol content. They also reported a decrease in carotenoid content by 17%. Carotenoids are the second most significant biologically- active compounds having health-promoting properties, being responsible for the orange-red color and constituting 0.03–0.5% of the dry form of the fruits. Zeaxanthin is one of the most abundant carotenoids in Goji constituting 31–56% of the total carotenoid pool and is found in the form of dipalmitin zeaxanthin. Goji has been reported as the best natural source of dipalmitin zeaxanthin known to humankind. Goji also contains beta-carotene, neoxanthin and cryptoxanthin, albeit at lower concentrations (Peng *et al.*, 2005; Wang *et al.*, 2010). Being bioactive compounds with very high antioxidant capacity, phenylpropanoids are reported to be present in Goji berry at 22.7 mg/100 g (gallic acid equivalent/g extract) (Guo *et al.*, 2008). Goji berries have free amino acids at 1.0–2.7%, and the most common of these is proline. Goji has also been reported to contain taurine and betaine (Potterat, 2010). It was found to contain vitamin C at 42 mg/100 g (Donno *et al.*, 2015). Llorent-Martínez *et al.* (2013) found Goji to have many mineral elements, including potassium (1.46 g/100 g), sodium (550 mg/100 g), phosphorus (184 mg/100 g), magnesium (90 g/100 g) and calcium (50 mg/100 g). Curiously, it has an iron concentration at 5.5 mg/100 g, which is quite high in relation to Dietary Reference Intake (DRI). Finally, Goji contains organic acids (citric acid, malic acid, fumaric acid and shikimic acid), with the most abundant fatty acids being palmitic acid, linoleic acid and myristic acid (Mikulic-Petkovsek *et al.*, 2012). The literature reports that Goji berries contain 370 kcal in dried fruits of 100 g (Gross, 2006).

Research has shown that polysaccharides, beta-carotene, vitamin E, phenolic, zeaxanthin and flavonoids are the main antioxidant components in goji berry (Potterat, 2010; Zhang, 2013). In the scholarly study, polysaccharides (*Lycium barbarum* polysaccharides, LBP), one of the bioactive ingredients of goji berry fruit peel, improve bone marrow function and cellular immune indicators; it might significantly enhance the energetic content of superoxide dismutase (SOD) in your body's blood, muscle, and liver tissue, which is conducive to the scavenging of free radicals. Research has even shown that LBP protected

frog eggs from oxidative harm from free radicals and increased the number of antibody forming cells to improve the antibody effect (Zhang, 2013).

In another study, goji flavonoids or TFL (total flavonoids of Lycium) was shown to protect red blood cells and mitochondria from oxidative damage. Similarly, TFL was also shown to protect certain white blood cells from oxidative damage. Zeaxanthin, one of the antioxidants that goji berry contains, may even help promote eye health. Studies have certainly shown that zeaxanthin from goji is well absorbed in humans (Zhang, 2013).

Goji berry contains phenolic constituents which helping in retarding oxidation of lipids and thereby improve the quality and nutritional value of food (Huang et al., 1999). Phenolics are natural antioxidants which have powerful effects in inhibiting the oxidation process. Antioxidant action of phenolic compounds is due to their tendency to chelate metals (Potterat, 2010; Zhang, 2013).

According to the literature, most of the studies on goji berry that have been carried out and fully explored are about its medical value. This study was conducted to gain more information about goji berries, especially antioxidants it contains. The study could help to determine the antioxidant properties and antioxidant activity of goji berries such as total phenolic content and radical scavenging activity (Zhang, 2013).

Luciobarbus esocinus (Figure 1.2) reaches a maximum length of up to 2. 3 m and weight as high as 140 kilograms. It is considered the largest extant cyprinid and may live for 17 years. It includes a large head, with a toothless mouth surrounded by four barbels. The silvery body is certainly covered with small scales. There is only one dorsal fin, a set of pectoral and ventral fins. The anal fin and tail they possess yellowish tones. The pike barbell takes place in the drainage basins of the Euphrates and Tigris rivers, including Iran, Iraq, Syria, and Turkey. Adults keep to larger bodies of water such as large rivers and reservoir, migrating to smaller inflows to spawn (Geldiay and Balık, 2007). *Luciobarbus esocinus* is a benthopelagic species living huge rivers and dams, but details of environmental requirements unidentified (Stone, 2007; Coad, 2011; Fishbase, 2011). The species are known as *Barbus esocinus* until 2007 was revised as *Luciobarbus esocinus* from 2007 (Fricke et al., 2007).



Figure 1.2. *Luciobarbus esocinus* used in this study

Keban Reservoir is one of the important areas where this species lives and it was reported that nearly 8.7% of the fish caught from Keban Reservoir consisted of *L. esocinus* (Celayir et al., 2006). This can be evaluated as a low rate, but it is the most valuable fish caught from the Keban Reservoir. So this rate is an important economical measurement.

The increase in the world population necessitates alternative food production while leading to decrease in earth-based food sources. The need for animal protein has also increased due to hunger or insufficient nutrition resulting from the enhancing population in recent years. Therefore, it is of utmost importance to utilize the available natural resources at the maximum level, to evaluate the obtained product as well as possible and to make it available for human consumption (Gündüz et al., 2018).

As a result of increasing urbanization and industrialization in our country and forcing working conditions, especially working people show great interest in the foodstuff used practically and ready for service and the interest aquaculture products has risen substantially. Smoked, canned, lakerda and marinated-processed and fried, cooked fish products are the products ready to be consumed without further preparation. Fish products that are packaged after cleaning and stored in a cold or frozen way are ready-to-cook products (Erol, 2013).

Processed products made from fish include various products such as smoked fish, fish sausage, fish salami and soujouk, fish burgers, fish crackers and fish meatballs. Some of these products are also preferred for the purpose of enriching people's taste and tables in addition to being ready for consumption. Nowadays, people are in search of new tastes and flavored tables instead of usual consumption. This has led to the creation of different fish products.

People are tended to produce ready-to-serve foods since women who do not have the opportunity to cook at home are tended to ready-to-serve foods with the increasing number

of working women today, working people have short time for lunch; places such as schools, hospitals, cafeterias and restaurants want to serve in a short time. This service is carried out in a short while with developments in World food technology. Ready to serve foods are the products which are made food by processing either directly or heated and consumed by itself or with some nutrients, having a certain shelf life and on which favourable processing techniques and methods are applied (Gökoğlu, 1994).

In this study, it was aimed to determine the chemical, microbiological and sensory changes that occur during the storage of Pike barbell (*Luciobarbus esocinus* Heckel, 1843) fish fingers coated addition of goji berry at $4\pm1^{\circ}\text{C}$ and to investigate the shelf life of this product.

2. MATERIAL and METHOD

2.1. Material

2.1.1. *Luciobarbus esocinus*, HECKEL 1843

Luciobarbus esocinus which are caught from in Keban Dam Lake (Figure 2.1) and have economic value is used in this thesis.



Figure 2.1. Keban Dam Lake where fish samples were obtained.

The fish required for the study was obtained from the fishermen hunting in the region in March 2017. Newly hunted fish were brought to the Faculty of Fisheries Fırat University under suitable conditions and were processed on the same day.

The study was carried out as two repeats and two parallel in the research. Information related to fish used in the study is submitted in the Table 2.1.

Table 2.1. Information related to fish used in the study

Length	100±10 cm
Weight	17±1 kg
Gender	Female

2.1.2. Goji berry

Goji berry extract (Figure 2.2) was obtained from a commercial company (Xi'a Xin Sheng Bio-chem Co.,Ltd, China) to add to coverings prepared in the study. This extract used is for food purpose, and has ISO 22000, GMP, FDA and halal certificates. Some features of the extract are presented in Table 2.2.



Fig. 2.2. Extract of Goji berry

Table 2.2. Specific features of Goji berry extract used in covering

Colour	Brown red
Shape	Powdery
Aroma and Flavour	Characteristic
Solubility	Soluble in water
Active ingredients	50% polysaccharide
Content of heavy metal	≤ 1.0 ppm
General number of living beings	<1000 kob/kg
Yeast-mold	<100 kob/kg
<i>E.coli</i>	Negative
<i>S. aureus</i>	Negative
<i>Salmonella</i>	Negative
<i>Shelf life</i>	In a cool and dry place, closed, away from high temperature and light, 2 years
Usage	For food purposes

2.2. Method

2.2.1. Preparation and Packaging of Experimental Fish Fingers

Blood and dirt were removed after the fish of which head was cut and internal organs were taken was washed to remove fresh fish fillets brought to the laboratory. Afterwards, fish skin was excoriated by using suitable tools and knives, the fillets obtained from the fishes of which the bones picked out were sliced in the length of 9 ± 1 cm, in the width of 2 ± 1 cm and in the height of 2 ± 1 cm, and fish meat was prepared for covering (Figure 2.3).



Figure 2.3. The fish slices preparation



Figure 2.4. The salted fish slices

Sliced obtained were hanged on the fridge for 1 hour salting in proportion as 1.5% (Figure 2.4). Liquid and solid coverings were prepared to cover the fish slices according to the content stated in the Table 2.3.

Table 2.3. Covering ingredients used in preparation of experimental samples

Coverings			
Liquid (%)		Solid (%)	
Egg white	75	Onion powder	2
Carbonate	3	Garlic powder	2
Crumbs bread	15	Red pepper	1
Wheat starch	5	Crumbs bread	40
Salt	1	Corn flour	30
Sugar	1	Wheat flour	25

Firstly, salted fish slices were covered with fluid, and then covered with simple solid covering, solid covering with additional 1% goji berry extract and solid covering with additional 2% goji berry extract as 3 groups (Table 2.4, Figure 2.5).

Table 2.4. Study of the experimental groups to be created

A	Simple coating prepared by the experimental group
B	1% goji berry extract with a coating made with experimental groups
C	2% goji berry extract with a coating made with experimental groups



Figure 2.5. The fish finger preparation

All of the fish fingers were fried for 1 min in oil at 180 C°. In this way the samples would be pre-cooked. After that samples of fish fingers would be packaged in styrofoam packages and would be stored at $+4\pm 1$ C°. The analyzes would be carried out in three stages, before fish processing, after processing and under storage conditions. In order to determine the shelf life of experimental fish finger, chemical, microbiological and sensory analyzes were performed with 3 replications every three days. Protein, fat, ash and moisture were determined before processing fish meat and after making fish finger. In addition, in the examples, pH, Thiobarbituric Acid Number (TBA), Total Volatile Basic Nitrogen (TVB-N) in certain days of the storage (days 0, 3, 6, 9, 12 and 15) and sensory analyzes were performed.

2.2.2. Nutritional Composition

- a) **Moisture:** The moisture content of fish samples were measured according to standard method 950.46 (AOAC, 2002a).
- b) **Crude protein:** Crude protein of samples were determined according to the micro kjeldahl method (method 928.08) (AOAC, 2002b).
- c) **Crude oil:** Soxhlet method would be used (method 960.39) to determine oil content (AOAC, 2002c).
- d) **Crude ash:** It would be determined according to method (920.153) (AOAC, 2002d).

2.2.3. Chemical Analyzes

- a) **pH Analysis:** The pH values of samples would be determined with a pH meter (Thermo Scientific Orion 3-Star). The sample was homogenized in distilled water in the ratio 1:10 (wt/vol), and the measurement was done with a pH meter (AOAC, 2002e).
- b) **Determination of Total Volatile Basic Nitrogen (TVB-N) :** Amounts of TVB-N in samples were made according to the method stated by Varlık et al. (1993). Volatile bases were kept in the H_3BO_3 solution of 3% with the water vapour distillation after that MgO was added to homogenized sample. Separated bases were standardized with 0,1 N HCl acid in company with Tashiro Indicator, and TVB-N amount of the samples was calculated as mg/100g.
- c) **Determination of Thiobarbituric Acid Number :** The method stated by Tarladgis et al. (1960) was applied. Absorbance of red colour at 538 nm given by malondialdehydes formed by oil oxidation, with thiobarbitruic acid at glacial acetic acid environment was read on spectrophotometer. Malondialdehyde value was calculated by multiplying the read absorbance value with 7,8 factor.

2.2.4. Microbiological Analyses

Samples were scaled as 10 kg in the special bag of a blender (Stomacher 400), and it was homogenized in the blender by adding 45 ml from the sterile 0.1% peptoned water on it

for the microbiological analyses. Thereby, sample dilution of 10^{-1} (1/10) was prepared. Other dilutions were made by means of using the same diluting agent from this dilution e.g. up to 10^{-6} . Two series of cultures were made by using 1 ml of each dilution of the samples with the pour plate technique, and plates consisting of 30-300 colonies were evaluated at the end of the incubation period (Varlık et al., 1993; Harrigan, 1998).

- a) **Count of Total Aerobic Psychophile Bacteria:** Plate Count Agar (PCA Merck1.05463) medium was used for count of total aerobic psychophilic microorganisms in samples. Plates were evaluated after incubated for 7 days at $\pm 5^{\circ}\text{C}$ (Harrigan, 1998).
- b) **Count of Total Aerobic Mesophile Bacteria:** Plate Count Agar (PCA Merck1.05463) medium was used for count of total aerobic mesophilic bacteria in samples. Colonies consisted after cultured plates were incubated for 72 hours at $30 \pm 1^{\circ}\text{C}$ were counted (Harrigan and McCance, 1976; ICMSF, 1986).
- c) **Count of yeast and mold:** Plates using Potato Dextrose Agar (PDA Merck1.10130) medium, of which pH was reduced to 3.5 by adding 10% tartaric acid (Meck1.00802) for the count of yeast-mold in the samples, was evaluated after 5 days incubation at $\pm 21^{\circ}\text{C}$ (ICMSF, 1986).

2.2.5. Sensory Analyses

The samples were analyzed in terms of sensorial on the 0., 3., 6., 9., 12., and 15. days of storage. The samples were evaluated by 10 panelists in terms of colour, appearance, odor, flavor and texture, and 5 points using 5-point hedonic scale was considered very good, 3 points were considered normal, and below 2 points were considered as defective. Form used for sensory analysis is demonstrated in Table 2.5 (Kurtcan and Gönül, 1987).

Table 2.5. Point Scoring Form for Sensory Analysis (Kurtcan and Gönül, 1987).

GROUPS	COLOUR	ODOR	APPEARANCE	FLAVOUR	TEXTURE
A					
B					
C					

Point Scoring: 5- Very Good 4- Good 3- Normal 2- Bad 1- Very Bad

2.2.6. Statistical Analyses

In this study, microbiological, chemical and sensory values obtained during the production of fish finger which were prepared experimentally were subjected to statistical analysis. SPSS® 22.0 computer package statistical program was used for statistical analysis. Kruskal-Wallis test was applied to determine whether there is a meaning difference between the groups or not, and the difference between the groups was determined with Duncan test (Özdamar, 2001).



3. RESULTS

In this study, Pike barbell (*Luciobarbus esocinus*) which are caught from Keban Dam Lake was used. Fresh fish was brought to the laboratory in aseptic and cold conditions. Heads were cut, eviscerated, washed, and then a slice of fillets of fish and boneless fish will be prepared. The fillet slices were coated with contents of 1% and 2% goji berry extract. Samples of fish fingers were packaged in styrofoam packages were stored at 4 ± 1 °C

Moisture, protein, fat, ash and salt analyses of the prepared fish finger samples were performed. pH, TVB-N, TBA values, microbiological and sensory qualities of the samples prepared in the study on the specific days of the storage were examined (0th, 3rd, 6th, 9th, 12th and 15th days).

3.1. Chemical and microbiological quality of the crude material

The chemical and microbiological quality of *Luciobarbus esocinus* used in the study is presented in Table 3.1.

Table 3.1. The findings about the fish meat used in the study

Parameters	Values $\bar{X} \pm S_x$
Moisture (%)	70.35 $\pm$ 1.85
Crude Fat (%)	11.68 $\pm$ 1.66
Crude Ash (%)	0.93 $\pm$ 0.17
Crude Protein(%)	15.66 $\pm$ 0.83
pH	6.26 $\pm$ 0.35
TVB-N (mg/100 g)	7.59 $\pm$ 0.58
TBA (mg MDA/kg)	0.90 $\pm$ 0.22
Aerobic Mesophile Bacteria Count (log cfu/g)	1.88 $\pm$ 0.01
Aerobic Psychrophile Bacteria Count (log cfu/g)	2.69 $\pm$ 0.30
Yeast-Mold Count (log cfu/g)	1.87 $\pm$ 0.18

X: Arithmetic mean

S_x: Standard deviation

3.2. Nutritional Composition

The results of chemical composition analysis belonging to simple samples (group A) and fish finger samples with the addition of goji berry extract 1.0% and 2.0% (group B and C) are presented in Table 3.2 and Figure 3.1.

As a result of the analyses, 63.80% moisture, 15.74% protein, 14.82% fat and 3.01% ash were calculated in Group A on average. In other samples with the addition of goji berry extract, 61.96% moisture, 16.39% protein, 15.73% fat, 3.42% ash were determined in the samples of group B, 61.99% moisture, 16.76% protein, 16.32 % fat and 2.92% ash were found in the samples of group C.

When the data obtained as a result of the study were analyzed statistically, this difference between the groups was found significant in terms of moisture, protein, fat and ash amounts determined in control fish finger samples and the samples prepared with the addition of extract ($p<0.05$).

Table 3.2. Average nutrient composition of fish finger samples (%)

Group	Moisture	Ash	Protein	Fat
A	63.80±0.33	3.01±0.02	15.74±0.73	14.82±0.28
B	61.96±0.27	3.42±0.16	16.39±0.68	15.73±0.75
C	61.99±0.26	2.92±0.55	16.76±0.36	16.32±0.60

A: Control group, **B:** Fish finger sample with the addition of 1% goji berry extract, **C:** Fish finger sample with the addition of 2% goji berry extract

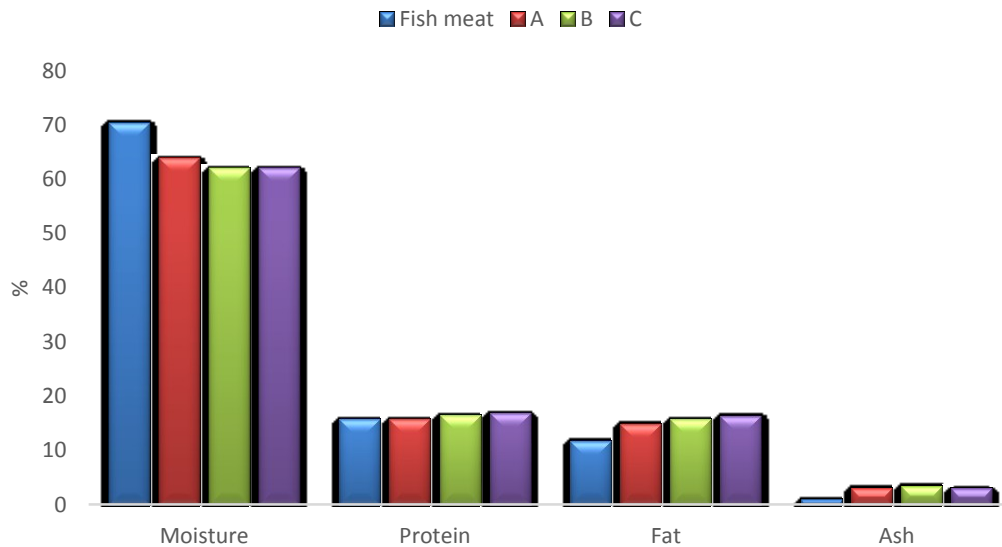


Figure 3.1. Nutrient composition of experimental fish finger samples (%)

3.3. Chemical Changes

The pH, TVB-N and TBA amounts of the experimental fish finger samples in group A, B, C, during the storage were determined. These determined values are presented in Table 3.3.

Table 3.3. Chemical quality parameters determined during the storage of fish finger samples ($\bar{X} \pm S_x$)

Chemical Parameters	Group	Storage Days						
		0.	3.	6.	9.	12.	15.	18.
pH	A	6.19±0.06	6.13±0.01	6.22±0.10	5.93±0.42	5.13±0.18	N.A.	N.A.
	B	6.09±0.06	6.26±0.03	6.25±0.19	6.31±0.22	6.30±0.23	5.86±0.36	5.72±0.05
	C	6.24±0.04	6.54±0.23	5.89±0.35	6.48±0.35	6.46±0.10	5.92±0.09	5.72±0.05
TVB-N mg/100g	A	8.11±0.13	12.92±1.19	21.26±0.74	30.13±1.33	32.78±0.52	N.A.	N.A.
	B	5.99±1.40	6.04±1.12	8.99±0.78	18.38±1.60	20.88±0.88	22.49±1.84	26.06±2.76
	C	3.76±0.51	4.16±0.23	4.16±0.24	13.29±2.80	22.16±0.07	15.94±0.5	16.94±0.54
TBA mgMDA/kg	A	1.26±0.08	2.80±0.12	3.14±0.11	4.31±0.15	5.45±0.64	N.A.	N.A.
	B	1.58±0.08	2.31±0.33	2.53±0.17	3.74±0.12	3.76±0.34	4.12±0.15	4.21±0.16
	C	2.09±0.16	2.07±0.35	2.05±0.10	2.35±0.25	2.87±0.17	3.22±0.04	3.14±0.11

N.A: No analysis

3.3.1. pH values

The pH values of the experimentally prepared fish finger samples measured throughout the storage are presented in Table 3.3 and Figure 3.2.

According to Table 3.3, the mean pH value of the simple fish finger (group A) sample was found as, 6.19±0.06 on the first day of the storage; a decrease was observed as 5.13±0.19 on the 12th day of the storage. In the fish finger samples with the addition of goji berry (B group), pH was determined as 6.09±0.06 on the first day of the storage and as 5.81±0.05 on the 18th of the storage. The initial pH of group C was 6.24±0.04 on the 18th day of the storage it was discovered as 5.61±0.09 on the 18th day of the storage.

When the data obtained as a result of the study were analyzed statistically, it was found that the difference of the pH values between the groups during the storage was significant in the simple samples and the samples with the addition of extract ($p<0.05$).

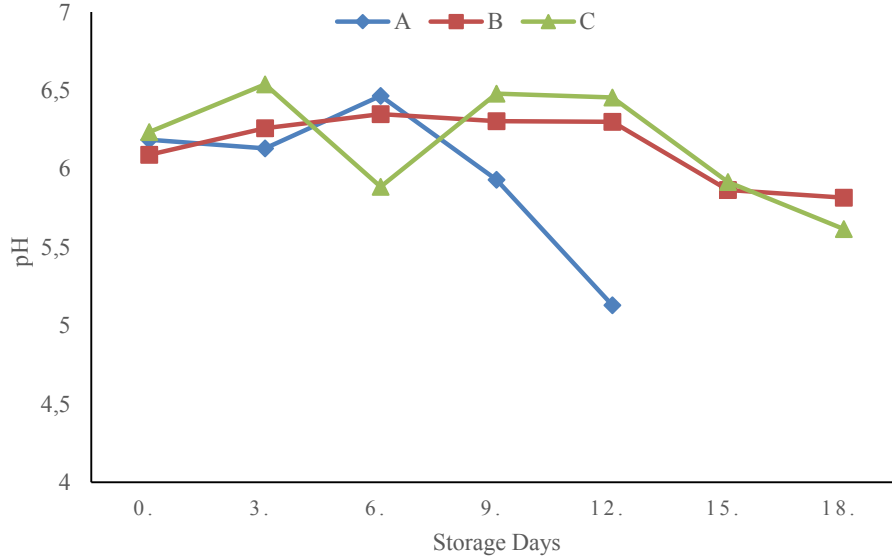


Figure 3.2. pH values of the experimental fish finger samples during the storage

3.3.2. TVB-N Value

The mean TVB-N values of the pike barbell samples, which were prepared experimentally in the study, during the storage are shown in Table 3.3 and Figure 3.3.

When the findings were examined, the mean TVB-N value of the group A fish finger sample, which was prepared controllably, was discovered as $8.11 \pm 0.13 \text{ mg/100g}$ on the first day. In other fish finger samples prepared with the addition of goji berry, the mean TVB-N value was found as 5.99 ± 1.40 for group B and $3.76 \pm 0.50 \text{ mg/100g}$ for group C. These values increased during the storage duration of the samples.

When the data obtained in the research were analyzed statistically, the difference between the simple samples and the samples with the addition of goji berry in terms of the TVB-N amount during the storage was found to be significant ($p<0.05$). In addition, the difference between the days of analysis in terms of the amount of TVB-N determined during the storage of the samples was also significant ($p<0.05$).

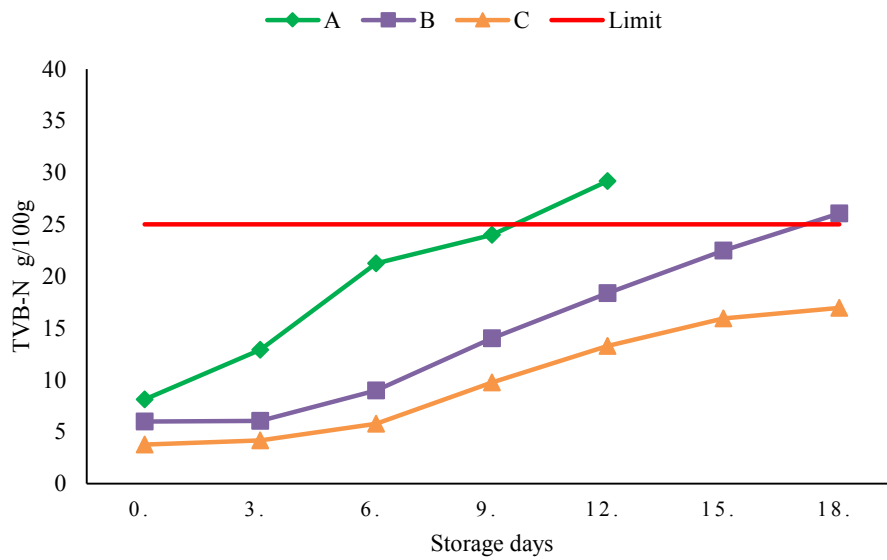


Figure 3.3. The TVB-N (mg/100g) values of the experimental fish finger samples detected during the storage

3.3.3. Thiobarbituric Acid (TBA) Value

The mean TBA values of the samples during the storage are given in Table 3.3 and the TBA changes during the storage are given in Figure 3.4.

When the findings were examined, they were found to increase during the storage. When the data obtained as a result of the study were analyzed statistically, it was found that the difference in the TBA amounts of the control samples and the samples with extract addition between the groups and days during the storage was significant ($p < 0.05$).

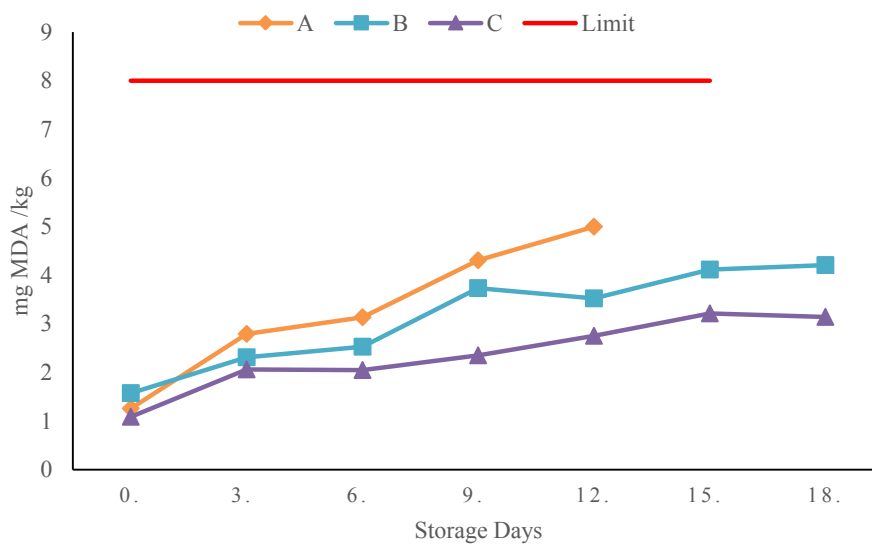


Figure 3.4. TBA (mg MDA/kg) values of the experimental fish finger samples during the storage

3.4. Microbiological Changes

The aerobic mesophile bacteria, aerobic psychrophile bacteria counts and yeast-mold counts of the control fish finger samples and the samples with the addition of goji berry extract in certain proportions were determined during the storage at 4°C. These determined values are presented in Table 3.4.

Table 3.4. Microbiological changes detected in the samples examined during the storage (log cfu/g)

Microbiological Parameters	Group	Storage Days						
		0.	3.	6.	9.	12.	15.	18.
Aerobic Mesophile Bacteria Count	A	2.10±0.06	3.16±0.51	5.47±0.34	5.94±0.43	6.40±0.62	N.A .	N.A .
	B	2.09±0.15	2.74±0.57	3.56±0.47	4.27±0.14	4.88±0.85	5.85±0.14	6.71±0.12
	C	2.26±0.02	2.45±0.03	2.80±0.14	3.30±0.42	4.27±0.26	4.50±0.58	4.19±0.15
Aerobic Psychrophile Bacteria Count	A	2.13±0.18	3.53±0.14	6.38±0.60	6.27±0.11	6.60±0.49	N.A .	N.A .
	B	2.83±0.57	2.73±0.67	4.33±0.21	4.83±0.66	5.59±0.09	6.41±0.05	8.07±0.16
	C	3.16±0.16	3.12±0.07	3.34±0.19	3.59±0.80	4.59±0.17	4.94±0.66	5.64±1.19
Yeast-Mold	A	2.54±0.54	3.05±0.14	3.39±0.12	3.89±0.18	5.51±0.33	N.A .	N.A .
	B	2.35±0.21	2.64±0.48	2.69±0.41	3.01±0.28	3.57±0.60	3.92±0.78	6.28±0.27
	C	1.68±0.13	2.67±0.32	1.77±0.26	2.18±0.31	3.33±0.16	3.58±0.11	4.58±0.59

N.A.: No analysis

3.4.1. Aerobic Mesophile Bacteria Count

Total aerobic mesophile bacteria counts (log cfu/g) determined by incubation of control fish finger samples and the samples prepared with the addition of goji berry at 30 °C are given in Table 3.4 and Figure 3.5.

The aerobic mesophile bacteria count of group C with the addition of goji berry extract (2%) was identified as 2.26 ± 0.02 log cfu/g on the first day of the storage, it reached 4.19 ± 0.15 log cfu/g on the 18th day.

An increase was observed in the aerobic mesophilic bacteria counts depending on spoilage throughout the storage. When the data obtained as a result of the study were analyzed statistically, it was determined that the intergroup difference between all the experimental groups in terms of aerobic mesophilic bacteria counts was statistically significant ($p < 0.05$).

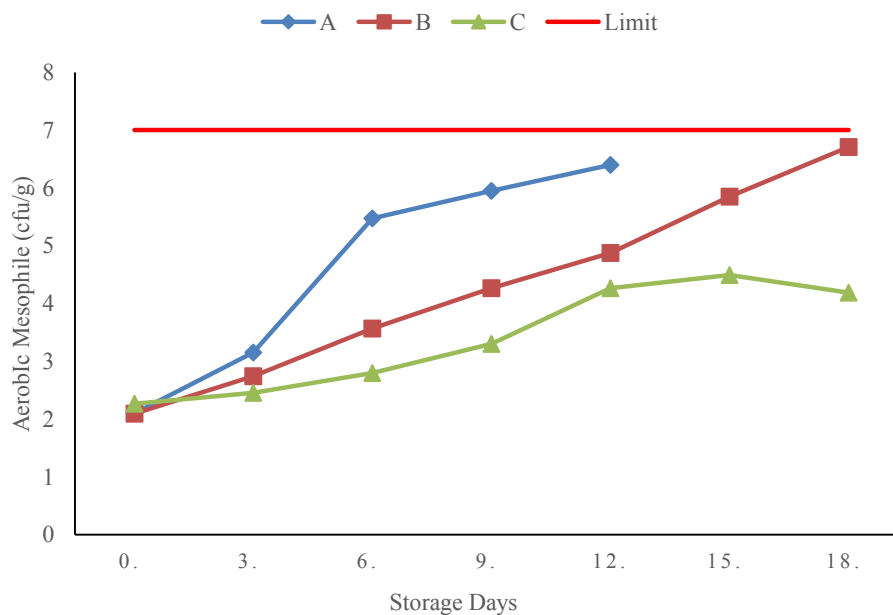


Figure 3.5. Changes in the number of aerobic mesophilic bacteria detected during storage of experimental fish finger specimens

3.4.2. Aerobic Psychrophile Bacteria Count

Total aerobic psychrophile bacteria counts (log cfu/g) determined by incubation of control fish finger samples and the samples prepared with the addition of goji berry at $\pm 5^{\circ}\text{C}$ are presented in Table 3.4 and Figure 3.6.

When the data obtained as a result of the study were analyzed statistically, the difference between the groups in terms of the aerobic psychrophilic bacteria count was not significant in control samples and samples with goji berry addition ($p < 0.05$). In addition, the differences between the total counts of aerobic psychrophilic bacteria within days during the storage were found significant ($p < 0.05$).

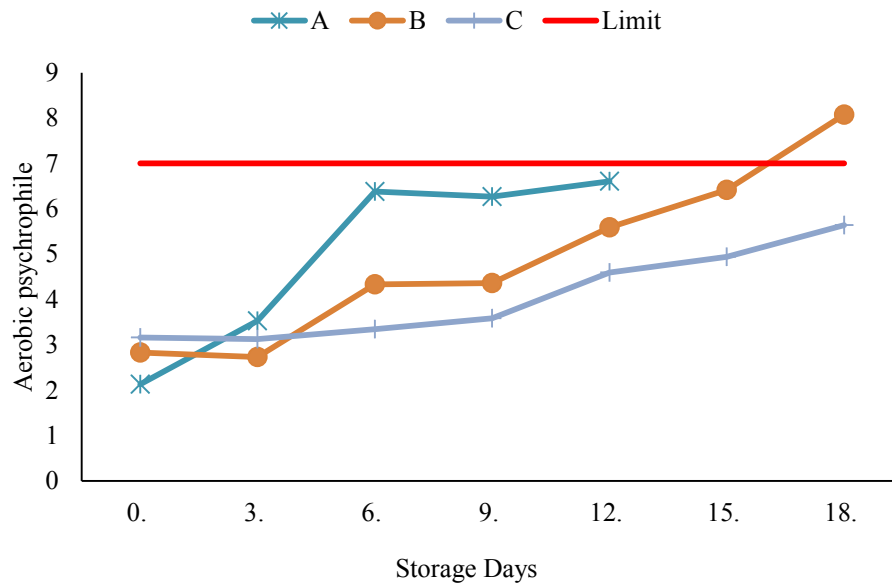


Figure 3.6. Changes in total aerobe psychrophilic bacteria counts (log cfu/g) of the experimental fish finger samples determined during the storage

3.4.3. Mold-yeast counts

Mold-yeast count (log cfu/g) of the control fish finger samples and the samples with goji berry addition determined by incubation at 21°C is given in Table 3.4'de and Figure 3.7.

When the data obtained as a result of the study were analyzed statistically, the difference between the groups in terms of yeast-mold count during the storage was found to be significant in the control samples and samples with extract addition ($p < 0.05$).

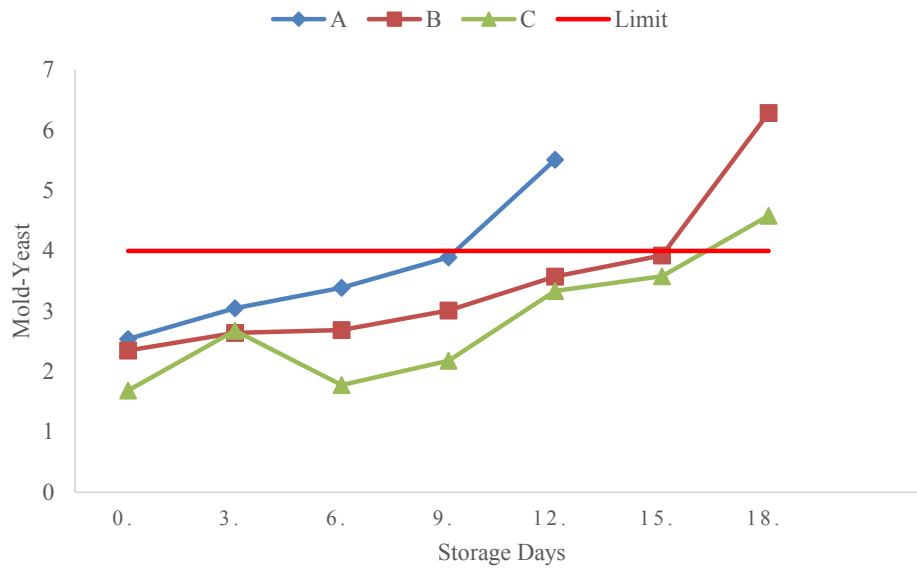


Figure 3.7. Changes in the yeast-mold counts of the experimental fish finger samples observed during the storage

3.5. Sensory Evaluation

During the sensory evaluation of the control fish finger samples and samples prepared with the addition of goji berry extract in different proportions, the appreciation of the samples' the color, smell, appearance, taste and texture were evaluated by 10 panelists using the hedonic scale. Throughout the storage, the panelists evaluated the control (without the addition of goji berry extract) fish finger samples and the samples with the addition of goji berry extract, which were made ready for consumption, according to their color, smell, appearance, taste and texture. The data obtained as a result of the panelists' scoring were evaluated and the change in the general appreciation levels of the samples during the storage were presented. The sample of group A lost its consumability on the 12th day of the storage and the samples of group B and C with fish finger samples containing extract on the 15th day; samples were not included in the analysis.

3.5.1. Overall acceptability

Overall acceptability of group A, B and C interm of storage days are given in Figure 3.8.

It was found that the effect of groups and duration on the overall acceptability level of the samples during storage was significant ($p<0.05$).

In terms of the overall acceptability scores in the evaluation between the groups, there was no statistically significant difference between the groups on the 0th day ($p>0.05$) while the difference between the control group and the groups with coating was significant during storage ($p<0.05$).

When the data obtained as a result of the study were analyzed statistically, it was found that the difference between the groups in terms of the general appreciation level of the control group A and group C sample from samples (group B and C) containing goji berry extract was significant ($p<0.05$).

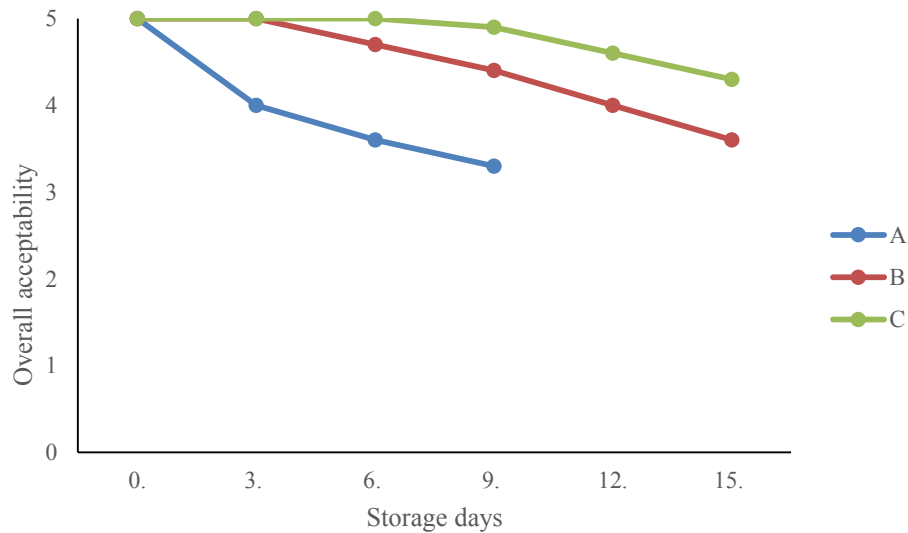


Figure 3.8. General appreciation scores of the experimental fish finger samples determined during the storage

4. DISCUSSION

In this study, the shelf life of fish fingers with different amounts of goji berry extract was investigated in cold storage. In this study, the moisture, ash, fat, protein, pH, TVB-N and TBA count, total aerobe microorganism (at 5 °C and 30 °C incubation temperature) and yeast-mold counts of both the material and the experimental groups were determined.

4.1. Nutrient Composition

In our study, average moisture, crude ash, crude fat, crude protein and carbohydrate amounts were determined in pike barbell fillet, which was used in the preparation of the experimental samples. The differences observed after the examination of the data in our study may be attributed to the difference in the seasonal and cultural conditions under which fish were obtained.

Tokur et al., (2006) respectively found the water, protein, fat and ash amounts in carp sticks as follows: 68.50%, 15.50% 6.00% and 2.20%. In their study, Gülyavuz and Timur (1991) obtained very similar amounts as a result of their investigation on the nutrient components of carp: 15.92% protein, 9.50% fat, 1.13% ash and 73.02% moisture. In Kara's (2007) study, protein values obtained at the end of storage according to the groups A, B, C and D were discovered as $15.16 \pm 0.01\%$, $15.65 \pm 1.12\%$, $14.88 \pm 0.84\%$, $15.68 \pm 0.77\%$, respectively, and crude fat values were found as A $6.68 \pm 0.03\%$, B $6.47 \pm 0.03\%$, C $6.27 \pm 0.19\%$ and D $6.30 \pm 0.18\%$.

In a different study, water, protein, fat, ash and carbohydrate amounts were investigated in sardine sticks (52.04%, 14.39%, 16.16%, 2.61% and 14.80%), in the whiting sticks (63.01%, 6.71%, 15.98%, 3.33% and 10.97%) and the sander sticks (69.73%, 4.28%, 15.75%, 2.75% and 7.49%) (Çaklı et al., 2005). In Yanar and Fenercioglu's (1999) studies, the amounts of 16.67 crude protein, 8.45 lipid, 73.04 moisture and 1.18 crude ash were found in carp meat, and the amounts of 15.34 crude protein, 6.98 lipid, 75.89 moisture and 1.09 crude ash were discovered in minced meat as %. In our research, moisture, crude ash, crude fat and crude protein amounts of fish fingers prepared for experimental purposes were determined. The significance of the difference between the groups in terms of the determined nutrient composition values was not found statistical ($p > 0.05$).

4.2. Changes in Chemical Properties

It was reported that pH value increased from 6.25 to 6.48 after 6 days of storage in Sardine meatballs (Kılınç et al., 2008). Similarly, the pH value of anchovy meatballs increased from 6.33 to 6.56 after 10 days of storage. In another study, it was reported that the pH value of anchovy meatballs increased throughout the storage (Turhan et al., 2001). In general, the pH value of fresh fish is stated as 6.0-6.5, consumability limit value as 6.8-7 (Glyavuz and Ünlüsayın, 1999). While the pH value of meatballs obtained from sablefish was 6.67, it was determined as 6.72 in the control group and 6.70 in the group with vacuum packing at the end of the storage period of 6 months (Ersoy and Yılmaz, 2003). Öksüztepe et al. (2010) expressed in researches that the value was found between 6.12 and 6.49 in meatballs of rainbow trout stored at 4 ± 1 °C when the effect of sodium lactate addition on the meatballs of Rainbow trout (*Oncorhynchus mykiss* W.) was investigated. In a study conducted by Varlık et al. (2000) in order to determine the shelf life of the marinated fish meatball, the initial pH value of the meatball marinades stored in cold was 4.19, that is, it firstly increased. Then, it decreased depending on the storage duration. In the study on making meatballs from remaining fillets of sander and tench, the pH values of sander and tench meatballs at $+4\pm1$ °C were initially determined as 6.16 ± 0.02 and 6.93 ± 0.42 , respectively. During the study, the values first decreased and then increased; they were then discovered as 9.12 ± 0.88 and 6.78 ± 0.09 on the 14th day of the study (Ünlüsayın et al. 2002). At the end of the storage, the pH values in all the meatball groups were observed between 4.92 ± 0.00 - 5.56 ± 0.01 . In the study investigating the effect of sodium lactate and thymol on meatballs made from mirrored carp, the pH value which was 6.69 on the 0th day dropped to 6.58 on the 6th day. Again in the other groups with sodium lactate and thymol addition, pH which was initially in the range of 6.58-6.70 decreased in general during the storage (Erol, 2013). In the study on the determination of the shelf life of marinated fish meatballs, the pH of the marinated meatballs was determined as 4.19 on average, while this value was found as 3.40 at the end of the storage. The pH values obtained in our study show similarities and differences with the findings in the aforementioned studies. The reasons behind the differences can be attributed to the applied solution proportions, fish species, storage temperature and the extracts used.

The TVB-N value is used as an indicator of spoilage in aquaculture products. This value increases as a result of the activities of endogenous enzymes and bacteria causing spoilage (Kyrana et al., 1997). In other words, Huss (1995) reports the amount of TVB-N contained in the newly caught fresh fish as 5–20 mg/100g and the acceptable freshness limit value as 30–40 mg/100g. Schormüller (1968), makes the quality classification according to the TVB-N values contained as “very good” up to 25 mg/100g, “good” up to 30 mg/100g, “marketable” up to 35 mg/100g, “spoiled” if more than 35 mg/100g. The same researchers state the consumability limit value of TVB-N in freshwater fish as 32–36 mg/100g. The TVB-N values of the meatballs obtained from carp fish varied between 10.52-13.78 mg/100g during the storage period of 6 months (Yanar and Fenercioglu, 1999). In the meatballs made from sablefish, these values ranged from 14 to 18.2 in the control group, and from 14 to 16.8 in the group with vacuum packing (Ersoy and Yilmaz, 2003). In a study, the TVB-N value, which was found as 11.0 in the 6th month of the storage of the sander meatballs at -18 ° C, was obtained as 12.0 in fish meatballs with rice (Yanar and Fenercioglu, 1999). In the study of Damarlı et al. (1992), it was revealed that the edibility limit was not exceeded in fish meatball samples stored by freezing during 12 months of storage, the initial level of 10 mg/100g reached 21 mg/100g at the end of 12 months. In the study conducted on investigating some quality parameters of meatballs made from raw and boiled fish meat, the TVB-N value of meatball samples made from raw anchovy increased depending on the storage duration. 39.33 mg/100 g was obtained on the 18th day of the storage in the 1st experiment, and 36.03 mg/100 g was discovered on the 15th day of the storage in the 2nd experiment, which means both values exceeded the limit (Akkuş et al., 2004). In the studies executed on making meatballs from remaining Sander and tench fillets, Ünlüsayın et al., (2002) initially found the TVB-N values in sander and tench meatballs as 11.4±1.10 mg/100 g and 11.2±1.14 mg/100 g at +4±1°C, respectively. On the 14th day of the study, 39.6±1.90 mg/100 g and 36.2±1.57 mg/100 g were obtained, respectively. In the study of Varlık et al. (2000), the TVB-N value was initially determined as 10.70 mg/100g in marinated meatballs, which is a sign of fish spoilage, and it was 10.45 mg/100g at the end of the storage. In Erol's (2013) study, the TVB-N value was discovered as 16.27 on the 0th day and 22.87 at the end of the storage in the control group. Again in the meatball groups with different contents, this value was determined as 15.83-17.83 on the first day and 20.28-23.08 on the last day of the storage. In the study of Öksüztepe et al. (2010), the TVB-N value increased from 8.16 mg/100g to 22.56 mg/100g in the control group. In the other meatball groups, it changed as 7.13-23.65

mg/100g. When the TVB-N values are examined during the storage in our study, there is an increase in all the groups depending on time, and the TVB-N amounts can be said to increase later in meatball groups with extract addition compared to group A. The increases in the TVB-N value was determined to be significant as the storage time passed ($p<0.05$), and the change in the TVB-N values between the groups was found statistically significant ($p<0.01$). When all the groups were compared in terms of TVB-N values during storage days, significant differences were discovered ($p<0.05$). When compared to different studies, these values obtained depending on the storage are similar. Some similarities and differences in our findings can be attributed to the applied solutions and their proportions, fish species, storage temperature and the extracts used.

The amount of TBA is an indicator of lipid oxidation and it is another method used to determine the freshness of fish and fish products. The amount of thiobarbituric acid (TBA) may vary depending on fish species, hunting places and methods, storage conditions. The TBA value, which emerges as a result of fat oxidation in aquaculture products and is the index of bitterness, is considered as "good quality" between 1–3 mg MDA/kg, "medium quality" between 3–5 mg MDA/kg, "low quality" between 5–8 mg MDA/kg, and the products exceeding 8 mg MDA/kg is classified as "inconsumable" (Sinnhuber and Yu, 1958; Varlık et al., 2007). In the study conducted on the making fish meatballs from carp meat, the TBA values of the product ranged from 0.8 to 2.2 (mg MDA / kg) at -18 ± 1 °C during the storage (Yanar and Fenercioğlu, 1999). In the study conducted on making meatballs from remaining fillets of sander and tench fillets, the TBA values of sander and tench meatballs were initially determined at 4 ± 1 °C as 1.71 ± 0.87 mg MDA/kg and 1.41 ± 0.6 mg MDA/kg, respectively. On the 14th day of the study, 9.12 ± 0.88 mg MDA/kg and 8.56 ± 1.23 mg MDA/kg were obtained respectively (Ünlüsayın et al. 2002). In another study, the value was 0.08 during the storage in the control group, but it varied between 0.06- 0.10 in other carp meatball groups (Erol, 2013). In another study, Can (2012) found the value as 0.79 mg MDA/1000g on the first day and 4.8mg/100g on the last day in the control group within the storage days, but it was determined as 0.74- 0.79mg/kg on the first day and 1.74 mg MDA/kg - 1.79 mg MDA/kg on the last day of the storage in other meatball samples. In our study, increases in TBA values were determined in all the groups as the storage time passed ($p<0.05$) and significant differences were also observed in all the groups when they were compared within the days of storage ($p<0.05$). In our study, the TBA counts determined during the storage were assessed and found similar to the aforementioned studies. In

addition, the TBA counts determined in our study during preservation were lower in goji berry extract groups than in other groups. Its reason can be expressed as the antioxidant effect caused by the components in the extract used. The reason behind the similarities and differences in our findings can be attributed to the applied solutions and their proportions, fish species, storage temperature and the extracts used.

4.3. Microbiological Changes

In general, the muscle of a newly caught healthy fish is sterile. Microorganisms are normally found in the skin, gills and intestines of the fish. Depending on the operations, temperature and duration applied after the fish are caught, microorganisms can move to muscle from gills, skin and intestines. As a result, the quality of the product deteriorates depending on the type of the microorganism, so consumers may be exposed to infection or toxicity. Therefore, the number and type of microorganisms in the muscle of the fish are important for health and storage (Gram and Huss, 1996; Gram and Huss, 2000).

When the data determined during the storage were evaluated statistically, the increase in the number of the microorganisms over time was found significant ($p < 0.01$). In addition, the effect of the storage duration on all the groups was discovered to be statistically significant ($p < 0.05$).

The acceptable limit value of the total count of aerobic mesophilic microorganisms in fresh fish was reported by ICMSF (1986) as 7 log cfu/g. The total count of aerobic mesophilic bacteria determined in our study exceeded the acceptable limit on the 12th day in group A and on the 15th day in group B and C, which were stored at 4 °C. In the study conducted on making meatballs from remaining sander and tench fillets, total mesophilic aerobic bacteria values of sander and tench meatballs were initially determined as 4.0×10^4 cfu/g and 4.2×10^4 cfu/g at $+4 \pm 1$ °C, respectively; afterwards, they increased and appeared respectively as 1.2×10^7 cfu/g and 1.4×10^7 cfu/g on the 14th day of the study (Ünlüsayın et al., 2002). Erol (2013) discovered in a study on meatballs that the count of aerobic mesophile bacteria generally increased during the storage. In the study conducted on the shelf life of anchovy meatballs stored in cold, total mesophilic aerobic bacteria value increased by the storage duration and was determined as 4 log cfu/g (1.0×10^4 cfu/g) at the beginning of storage, reaching 7.76 log cfu/g (5.8×10^7 cfu/g) on the 10th day of the storage (Turhan et al, 2001). In the study investigating some quality parameters of the meatballs made from raw

and boiled fish meat, total mesophilic aerobic bacterial value of the meatballs made from raw anchovy increased depending on the storage duration and it was found as 4.8 ± 0.007 log cfu/g in the 1st experiment and 4.6 ± 0.021 log cfu/g in the 2nd experiment at the beginning of the storage, exceeding the limit value by reaching 7.4 ± 0.014 log cfu/g in the 1st experiment and 7.3 ± 0.002 log cfu/g in the 2nd experiment on the 12th day of the storage (Akkuş et al. 2004). In the study where Öksüztepe et al. (2010) investigated the effect on the fresh rainbow trout (*Oncorhynchus mykiss* W.) meatballs prepared with sodium lactate addition, the following levels were reached: 8.83 log cfu/g on the 8th day of the storage in the control group, 8.79 log cfu/g on the 10th day in group A, 8.90 log cfu/g on the 12th day in group B and 8.80 log cfu/g on the 16th day in group C. It can be expressed that these findings are in parallel with our findings. In addition, when we look at the total aerobic mesophile counts determined in our study, it can be seen that herbal extracts have antibacterial effect. When the findings are compared, we can attribute the reason behind some similarities and differences to the differences in the applied solutions and their proportions, fish species, storage temperature and the extracts used.

Psychrophilic bacteria are the most important group of microorganisms responsible for aerobic spoilage of fresh fish stored at low temperatures (Sallam, 2007). In our study, the counts of psychrophilic bacteria increased in all the groups during the storage depending on time. In a study conducted on the suitability of tench (*Tinca tinca* L., 1758) for making meatballs and the changes in the nutritional properties after processing, the total psychrophilic bacteria load values ranged between 3.99 ± 0.11 log cfu/g and 6.91 ± 0.014 cfu/g at $+4 \pm 1$ °C. The total psychrophilic bacteria values increased time-dependently. All the samples remained below the limit value (Çapkın, 2008). In the study investigating some quality parameters of the meatballs made from raw and boiled fish meat, the total aerobic psychrophilic bacteria load value of the meatballs made from raw anchovy increased depending on the storage duration; it was found as 4.0 ± 0.001 log cfu/g in the 1st experiment and 4.5 ± 0.004 log cfu/g in the 2nd experiment at the beginning of the storage, exceeding the limit value by reaching 7.7 ± 0.002 log cfu/g in the 1st experiment and 7.8 ± 0.014 log cfu/g in the 2nd experiment on the 12th day of the storage (Akkuş et al., 2004). Erol (2013) discovered in a study on meatballs that the count of aerobe psychrophilic bacteria generally increased during the storage. On the 6th day of the storage, the total aerobic mesophile counts increased above 7 log cfu/g in the control group and groups containing 0.1% thymol and 1% sodium lactate + 0.1% thymol. In the group containing 1% sodium lactate + 0.25% thymol,

almost 7 log cfu/g was reached on the 8th day, 7 log cfu/g was exceeded on the 10th day of the storage in the group containing 1% sodium lactate. Similar results were also obtained in this study. It can be stated that the antibacterial effect of goji berry extract used in our study influenced the results.

Yeasts and molds are not found in normal flora in fish. They are generally of earth-origin and transmitted from water as soon as they are caught or tools and materials used after catching the fish. In our study, the yeast-mold counts at the end of the storage were determined in samples stored at 4 °C as 6.47 ± 0.37 log cfu/g in group A, 4.96 ± 0.69 log cfu/g in group B and 4.58 ± 0.59 log cfu/g in group C. In the samples, difference was found between the groups in terms of the yeast-mold counts ($p < 0.05$). Erol (2013) discovered in a study on meatballs prepared from mirrored carp that the yeast-mold counts had increases during the storage. Only in the group containing 1% sodium lactate + 0.25% thymol, the yeast-mold count on the 4th day of the storage was slightly lower than the 0th day. In another study, the yeast and mold count of fish meatballs obtained from Rainbow trout was determined as 1.56 log cfu/g in the fillet. This number increased in all the groups, including the control, due to the progress of the storage and it was observed to be at the same levels in the other groups except the group C with 2% sodium lactate addition (Öksüztepe et al., 2010). In terms of the yeast-mold count, the effect of storage duration on all groups was found to be statistically significant ($p < 0.05$). In our study, when the change in yeast and molds during the storage was examined considering the antimicrobial properties of goji berry, the difference in group B and C with extract addition is seen compared to the control group.

4.4. Sensory qualities

The sensory analysis used when purchasing any food product is the first method we apply to determine the freshness of the product. It is widely used in determining the quality of foods because it gives fast results. Chemical, microbiological and physical analyses are associated with sensory analyses in determining the shelf life.

Although physical, chemical and microbiological analyses used to determine the freshness level of the fish can significantly evaluate the shelf life, sensory analyses have a much more important place in consumption, because people make use of sensory analyses instead of experiments organized under laboratory conditions when deciding on consuming a food.

As a result of the sensory analyses on the products prepared in the research, a major part of the panelists liked the fish finger samples very much. In terms of the examined fish finger samples, group B and C received higher scores due to some specific physical properties of the goji berry extract.

In their study conducted for the purpose of developing new products and determining the duration of lasting by applying freezing technologies on these products, Damarlı et al. (1992) stated that fish meatball with rice, fish stick and anchovy with vegetables were "good" in 9 months and "marketable" in 9-12 months at -18 °C.

Regarding the sensory analysis results obtained from Gökoğlu's (1994) study conducted to assess Mackerel (*Scomber scombrus*) as meatballs, the samples were evaluated for tissue, smell, taste and appearance, and it was observed that the tissue softened, smell got heavier and the taste disappeared in further stages of the storage although these four qualities were fine initially. It was discovered that the samples were in good condition until the 6th day, marketable on the 8th day and spoiled at the end of the storage, on the 10th day.

In Avcı's (1996) study conducted to determine the shelf life of trout salad and meatball stored at +2 °C±1, it was stated that the trout salad was "very good" on the first days of the cold storage, "good" until the 16th day and inconsumable after the 16th day. It was reported that the trout meatball was "very good" until the 2nd day, "good" until the 8th day, "marketable" until the 10th day and "inconsumable" after the 10th day.

The products, which were obtained in the study of Baygar et al. (2002) conducted to increase the Fish consumption, present different products to consumers and to determine the shelf life of the stuffed trout that was stored in cold, were liked very much and a different taste was put forward. As a result of the sensory analysis of the products obtained, most of the panelists liked the fish meatballs in terms of taste, smell, flavor, appearance and general appreciation. It was reported that they felt the smell of meatballs rather than fish in terms of smell and taste, and a pleasant taste was obtained with the effect of spices. The obtained meatballs received 8.00 to 9.50 out of 10 after the first sensory analysis. At the end of the storage, meatballs of group C became the group which was liked mostly. The sensory values of the cold stored fish meatballs decreased depending on the storage duration.

As a result of the sensory analysis on the products obtained in a study conducted by Ünlüsayın et al. (2002) in respect of making meatballs from the remaining sander and tench fillets, sander and tench meatballs were liked to a large extent, sander meatballs were liked

more than tench meatballs. Damarlı et al., (1992) reported that the product prepared in their study related to sander meatballs was liked by 90%.

In their study, Yanar and Fenercioğlu (1999) produced fish meatball from Carp (*Cyprinus carpio*) meat and researched the sensory qualities and the shelf life of the product. Fish meatballs were prepared in five different contents as garlic, garlic + sunflower oil, onion, onion + sunflower oil and control group. As a result of sensory evaluation, the products obtained scores between 7.4 and 9.07 out of 10 in terms of appearance, tissue, juiciness, smell, taste and flavor and general appreciation, and they were liked very much. The effects of onion and garlic use on appreciation were found similar ($p < 0.05$). Addition of fat to meatball had no significant effect on appreciation ($p > 0.05$).

In a study conducted by Varlık et al. (2000) in order to determine the shelf life of Marinated fish meatball, sensory values of meatball marinades stored in cold decreased depending on the storage duration. The 120th day of the storage was evaluated as spoiled quality. Meatball marinades received 9.00 out of 10 at the beginning of the storage, 5.57 on the 120th day and 3.50 on the 150th day of the storage.

According to the sensory analysis findings in carp sticks; color, smell, taste and general appreciation properties were evaluated as 8.750 (Tokur et al., 2006). In a different study, the general appreciation in the carp sticks was 8.471 on average.

The general appreciation in whiting, sardine and sander sticks were considered “good” (Çaklı et al., 2005). The value of sensory analysis obtained in trout sticks was found to be very good as reported by different researchers in fish stick studies (Berik et al., 2011).

According to the results of sensory analysis in the study conducted on the determination of the shelf life of the fish meatball made from mackerel at +4 °C, the samples were revealed to be 'good' until the 6th day, 'marketable' on the 8th day and spoiled on the 10th day (Akkuş et al., 2004).

As a result of the sensory analysis of the products obtained in Çapkın's (2008) study conducted on the suitability of tench (*Tinca tinca* L., 1758) for making fish meatballs and the changes in post-processing nutritional properties, it was revealed that a great part of the panelists liked the fish meatballs in terms of taste, smell, texture and appearance. It was reported that they felt the smell of meatballs rather than fish in terms of smell and taste and received a pleasant taste with the effect of spices. The obtained meatballs received 8.26 to 9.05 out of 10 after the first sensory analysis.

5. CONCLUSION

The results obtained from this study showed that goji berry extract prolongs the shelf life of food. The use of goji berry extract in the food industry will contribute to the shelf life of the products in a positive way. The results show that fish fingers prepared with goji berry addition can contribute to the national economy.

Now, the positive effect of fish consumption on human health is known through scientific studies conducted in various fields. Increasing fish consumption in our country is an important matter for both economy and health. Countries that are aware of balanced nutrition are looking for new products which will satisfy the consumer in sensory terms in order to further enrich the protein sources and can be prepared easily, and they make investments accordingly. Although the consumption of fish is low in our country, the consumption form of fresh fish is limited to a few cooking methods. In this study, fish finger was prepared with mirror carp as a ready-to-use product. As it is understood from the sensory data, the product prepared in the study was highly appreciated by the panelists and the appreciation continued during the storage duration.

With this study, it is possible to express that the shelf life can be extended through natural extracts without the use of chemical additives, which are used to prolong the duration of lasting that can also be defined as the shelf life of the food, and if used unconsciously, threaten the consumer health.

According to the findings obtained in this study, it was concluded that fish fingers with goji berry addition had a positive effect on chemical, microbiological and sensory qualities and this study could also establish a basis for many scientific studies with the application of herbal extracts used in experimental samples on various fish products in more different concentration

6. SUGGESTIONS

As a result of the data obtained in the study; it is thought that

1. Consumption of the fish finger form will increase depending on its low consumption in our country although the nutritional value of the mirrored carp fish used in the study is high,
2. Goji berry extract has a positive effect on the chemical, microbiological and sensory qualities of the product and therefore the shelf life of fish fingers is prolonged,
- 3 Chemical, microbiological and sensory properties of the fish fingers which are made ready will be preserved for a longer period.
4. In today's economic and social conditions, factors such as increasing number of people working outside home, changing dietary habits and having less time to prepare meals lead people to the consumption of foods which are ready for consumption or more practical to prepare. It is thought that this study will contribute to ready food consumption.
5. It has been concluded that the findings obtained in this study will be a good source for such researches.

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