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Research Article

Investigation of Biogenic Amine Levels in Fish and Fish Products Sold in Şanliurfa

Esra OĞUZ EKİNCİ[1] and Muhammed Yaşar DÖRTBUDAK[2]*

[1]Harran University Institute of Science, Department of Animal Science, Şanliurfa, Türkiye

[2]Harran University Faculty of Veterinary Medicine, Department of Fisheries and Diseases, Şanliurfa, Türkiye

***Corresponding Author:** Muhammed Yaşar DÖRTBUDAK, Harran University Faculty of Veterinary Medicine, Department of Fisheries and Diseases, Şanliurfa, Türkiye.

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Abstract

Fish is an important source of protein, minerals, vitamins and fatty acids and offers a unique and balanced nutritional source for all age groups. However, since fish is a very sensitive food product, if it is not stored properly, it starts to deteriorate immediately after leaving the water and biogenic amines may occur. The aim of this study was to determine the biogenic amine levels in four fish species Sardine {*Sardina pilchardus* (Walbaum, 1792)}, Haddock {*Merlangius merlangus* (Linnaeus, 1758)}, Mackerel {*Scomber scombrus* (Linnaeus, 1758)} and Tuna {*Thunnus thynnus* (Linnaeus, 1758)} sold in the markets in the central districts of Şanlıurfa province. For this purpose, 20 fish of four different species were collected from the markets in the central districts of Şanlıurfa. The samples were analyzed by gas chromatography (GC-FID) for heptilamine, putrescin, cadaverine,Histamine , tyramine and spermidine biogenic amines. Although the highest levels of Heptylamin, tyramine, cadaverine were detected in sardine species, there was no statistically significant difference between fish groups ($p > 0.05$). Heptylamin (48,254 mg/100 g), Tyramine (106,053 mg/100 g) and Cadaverine (121,756 mg/100 g) were highest in sardine fish samples. As a result, it was concluded that low levels of biogenic amines were detected in the fish sold in Şanlıurfa, the fish were stored at appropriate temperatures and conditions and the fish sold in the markets in Şanlıurfa province did not have any harmful effects on human health in terms of biogenic amines.

Keywords: Aquaculture; Biogenic Amines; Canned Fish; Şanliurfa

Introduction

Fish and fish products are an excellent source of nutrients for human nutrition due to the proteins, vitamins, salt minerals and polyunsaturated fatty acids they contain [1]. However, when hygiene and storage conditions are not suitable for these products, they can cause significant health problems because they deteriorate rapidly [2]. Even if seafood products are stored under very cold conditions, they are foodstuffs that can deteriorate and decrease in quality very quickly compared to other foods through autolysis, microbial and oxidation pathways. Therefore, it is important to consume these products fresh. Fish and fish products are extremely sensitive to biogenic amine formation due to the metabolism of spoilage microorganisms, which causes quality loss and deterioration [3]. Therefore, the analysis of spoilage metabolites (biogenic amines) in fish and fish products is accepted as a quality index and provides information about the quality and freshness of fish. This is because biogenic amines are normally found in very low concentrations in fresh fish. Their amount can

increase rapidly due to contamination and improper conditions during processing and storage [4].

Biogenic amines are naturally occurring low molecular weight compounds with aliphatic, aromatic and heterocyclic chemical structures. Biogenic amines are mainly formed in foods through microbial decarboxylation of amino acids and transamination of aldehydes and ketones [5]. Biogenic amines can occur in all foods that contain protein and can perform biochemical and microbial reactions. The amine level in foods can vary significantly depending on the quality of the food and the microorganism. As a result of bacterial activities, amino acids in foods can be separated from proteins [6]. Biogenic amines are formed due to the decarboxylation of free amino acids by enzymes [7,8]. Although biogenic amines play very important roles in the structure of organisms (cell growth and development, regulation of gastric pH, regulation of blood pressure), they can cause food poisoning and allergic reactions in humans and animals in excess [9-11].

Biogenic amines that are frequently formed in consumed foods include Histamine, cadaverine, spermine, spermidine, putrescin and tyramine. Among the different biogenic amines,Histamine is considered the most important toxin [12]. Biogenic amines cannot be degraded by commonly used food preservation methods such as freezing, cooking, sun drying and smoking [13]. The presence of biogenic amines in foods and their levels in food content depend on many factors such as the presence of free amino acids, water activity, pH, salt level, bacterial density and temperature [14].

The aim of this study was to determine the biogenic amine levels in Sardine, Haddock, Mackerel and Tuna sold in markets in Şanlıurfa province.

Materıals and Methods

Since there is no application for live animals in our study, it is not subject to Local Ethics Committee for Animal Experiments permission in accordance with Article 8 (k) of the "Regulation on the Working Procedures and Principles of Animal Experimentation Ethics Committees". Ethics committee certificate is not required.

Supply of fish samples

2 frozen Sardine {*Sardina pilchardus* (Walbaum, 1792)}, 2 frozen Haddock {*Merlangius merlangus* (Linnaeus, 1758)}, 2 frozen Mackerel {*Scomber scombrus* (Linnaeus, 1758)} and 7 canned Tuna {*Thunnus thynnus* (Linnaeus, 1758)} were purchased from markets in the central provinces of Şanlıurfa (Eyyubiye, Haliliye and Karaköprü). Fish samples were collected whole and the internal organs were cleaned**.** The purchased fish and fish products were brought to Harran University, Faculty of Agriculture, Department of Animal Husbandry laboratory under sterile conditions and cold chain. They were kept at -18ºC until the analyzes were performed.

Preparation of fish samples

Each fish was homogenised thoroughly in a homogeniser together with the skin. A random sample was taken from this mixture for analysis. In each sample, 5 mg/kg of each fish species was taken under sterile conditions and homogenized by beating with ethyl alcohol (30 ml) for 5 min. The samples were kept in a 45°C bath for 45 minutes. After the bath, the samples were brought to 30°C in a water bath. After this process, the samples were filtered, distilled and centrifuged (2000 rpm for 5 min). After centrifugation, 3 ml of each sample was taken and analyzed by Gas Chromatography (GC-FID / Thermo, Trace GC ultra, Italy).

Gas chromatography (GC-FID) analysis

Samples were analyzed for biogenic amines heptilamine, Histamine, putrescin, tyramine and cadaverine. A temperature program was applied to the column for the separation of biogenic amines. The oven temperature was set at 110 °C for 2 minutes and increased to 190 °C with an increase of 5 °C per minute. Derivatised biogenic amines were analysed using a GC, where a HP-5 phenyl methyl siloxane (30 m x 0.25 mm x 0.25 µm) silica capillary column was installed in a Hewlett Packard 6890 and equipped with a FID. Carrier gas was hydrogen. Confirmation of peaks and retention times were obtained using GC-FID, derivatised biogenic amines were identified with GC-MS where it was performed on a capillary BPX-5 column (30 meter x 0.25 mm x 0.25 µm). Helium was used as carrier gas. The injection volume was set to 1 µl [15].

Statistical analysis

IBM SPSS Version 16.00 (SPSS Inc., Chicago, IL, USA) software was used for statistical analyses. One-way analysis of variance (One-way ANOVA) was applied to compare the biogenic amine levels detected in each fish species. After the analysis of variance, Duncan's multiple comparison test was applied to determine the differences between the means of the groups. Results were presented as mean ± standard error (IBM SPSS, 2017).

Results

Biogenic amine levels detected in fish samples are given in table 1. Although the highest levels of heptylamin, tyramine, cadaverine were detected in sardine species, there was no statistically significant difference between fish groups ($p > 0.05$).

Fish species (n)	Heptylamin	Putrescin	Histiamine	Tyramine	Cadaverine
Tuna (2)	17,87 ± 3,39[a]	Not detected	Not detected	44,41 ± 2,20[a]	38,79 ± 8,27[a]
Mackerel (3)	22,50 ± 7,71[a]	Not detected	Not detected	54,302 ± 28,00[a]	46,502 ± 22,81[a]
Haddock (2)	22,25 ± 2,35[a]	Not detected	Not detected	54,37 ± 8,72[a]	73,401 ± 15,80a[b]
Sardine (2)	48,254 ± 40,95[b]	Not detected	Not detected	106,053 ± 36,88[b]	121,756 ± 67,05[b]

Table 1: Biogenic amine levels (Mean±standard deviation) detected in fish samples (mg/100g).
n: number of samples

Discussion

The maximum histamine level in seafood in Turkey is 20 mg/100g [16]. Histamine value was found in fish. Therefore, it was determined that there was no risk in terms of Histamine biogenic amine in the samples. In support of our results; reported that Histamine levels of different canned fish sold in Van were not at a level that would adversely affect human health [17,18].

Found that the amount ofHistamine in mackerel, bonito and anchovy fish was below the toxic level even after three days of storage [19].

Reported that biogenic amine levels were low or moderate in most fish products and that not all biogenic amines were detected in all fish product samples [20].

Reported that the amount ofHistamine was below the detection limit in the frozen gutted samples examined during storage in frozen whole fish samples, while it could be detected only on the 30th and 60th days of storage in frozen whole fish samples [21].

Although the highest biogenic amine levels were detected in sardine species, there was no statistically significant difference between the fish groups ($p > 0.05$). Low levels of biogenic amines were detected in fish sold in Şanlıurfa. In support of our study, reported in their study that canned fish samples in Ghana generally contained low levels of biogenic amines and that the order of these amines was tyramine, putresin, agmatine, Histamine and cadaverine. Histamine and tyramine concentrations were found in mackerel and sardine, respectively. They reported that none of the analyzed samples exceeded theHistamine level set by the European Union [4].

Determined Histamine, tyramine, putrescin and spermidine levels of canned tuna fish in Iran. They detected Histamine in 57.5% and tyramine in 55% of the samples and stated that these values were below the permissible levels in foods [22].

Found that Histamine levels in thirty-five different types of fish from 12 different countries, including Turkey, exceeded the levels approved by the FDA in eleven samples and were above the levels approved by both the EU and Turkey in four fish [23].

Found high levels of Histamine, phenylethylamine, putrescin, tyramine, tyryptamine, spermine and spermidine in fish samples collected in the Czech Republic [24].

Reported that almost 10% of the fish samples from Turkey and Europe exceeded the biogenic amine limits approved by the FDA and the European Union, the most abundant amine was tyramine and the highest Histamine was found in anchovies [25].

Reported that a large amount of Histamine was encountered in the samples used [26].

The results of these studies are not in accordance with the results of the present study. It is thought that the reason for this may be due to the low quality of the fish or the storage in unhealthy environments before processing and the faulty processing, as well as the high storage temperatures.

The detection of low levels of biogenic amines in fish sold in Şanlıurfa shows that the fish are kept at appropriate temperatures and conditions. Because it is reported that the most important way

to prevent the formation of biogenic amines in fish is to keep the fish in a cold environment of 0 ºC or colder as soon as it is caught [27]. Additionally, it has been reported that the storage time and temperature of fish have an effect on the formation of biogenic amines [28].

Conclusion

In this study, 4 fish species (Sardine, Haddock, Mackerel and Tuna) were used and a total of 20 samples were examined. As a result of the study, it was determined that the levels of biogenic amines in the fish samples were low and there were no biogenic amines at a level that would cause toxic effects in any species. This shows that the fish sold in markets in Şanlıurfa province do not have any harmful effects on human health in terms of biogenic amines. Since biogenic amines cause toxicity, constant controls must be ensured and necessary precautions must be taken. In particular, inspections should be carried out for hygiene and cold chain monitoring. In this way, biogenic amine formation will be prevented and human health will be secured.

Conflict of Interest

The authors declared that they have no actual, potential or perceived conflict of interest for this article.

Acknowledgment

The summary of this study was presented as an oral presentation at "2. INTERNATIONAL HASANKEYF SCIENTIFIC STUDIES AND INNOVATION CONGRESS 25-26/06/2022"BATMAN / TÜRKİYE"

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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