

## **IMPACT OF BIOGENIC AMINES IN SALTED "MUGIL CEPHALUS" ON PUBLIC HEALTH**

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### **SUMMARY**

The biogenic amines content of various foods has been widely studied because of their potential toxicity. This study aims to present data about biogenic amines content in Egyptian salted "Mugil cephalus" and throughlights on its public health hazard. Twenty random samples of salted "Mugil cephalus" collected form Cairo and Giza governorates were analysed for histamine, tyramine,  $\beta$ -phenylethylamine, tryptamine, cadaverine and putrescine by high pressure liquid chromatography. Results showed that tyramine,  $\beta$ -phenylethylamine, histamine, tryptamine, cadaverine and putrescine were detected in 70%, 50%, 45% , 45%, 40% and 30% of samples respectively. Most of these amines were found in a moderate level that do not represent any hazard unless large quantities are ingested. Cadaverine

and putrescine could potentiate toxic effect of histamine, however medication with monoamine oxidase inhibitor (MAOI) could potentiate the effect of histamine, tyramine and  $\beta$ -phenylethylamine. This result make a great need for controlling biogenic amines production in salted fish to ensure safety and protect consumer's health.

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### **INTRODUCTION**

Biogenic amines are present in a wide range of food products including fish products, meat products and dairy products (Brink et al., 1990; Askar and Treptow, 1986) .Biogenic amines in food are formed by the enzymes of raw material or are generated by microbial decarboxylation of amino acids (Halász et al., 1994 and Brink et al., 1990).

Although biologically active amines occur natu-

rally in many foods and these amines, such as histamine, tyramine and putrescine are needed for many critical functions in man and animals, they seldom present any hazard unless huge quantities are consumed or the normal routes of catabolism are inhibited or are genetically deficient (Koehler and Eitenmiller, 1978; Lovenberg, 1973).

Different biogenic amines (histamine, putrescine, cadaverine, tyramine, spermine and spermidine) have been detected in fish. Histamine is the causative agent of scombroid poisoning as scombroid fish are the type of fish commonly involved, however several species of non scombroid fish have often been implicated in incidents of scombroid poisoning (CDC, 2000; Price and Melvin, 1994). Moreover, tyramine and phenylethylamine cause a rise in blood pressure in contrast to histamine which reduces blood pressure.

Histamine is commonly found in foods that have been aged or fermented especially fermented fish products and fish paste which frequently contain high amounts of histamine that could possibly present a hazard (Stratton et al., 1990; Rice et al., 1975). Meanwhile, Taylor (1985) recorded that Egypt has experienced some problems with fermented "Mugil cephalus".

Little research has been conducted to determine the biogenic amines content of salted fish products. Therefore the objective of the present study is to estimate quantitatively the biogenic amines

in Egyptian salted "Mugil cephalus" concerning its public health hazards.

## **MATERIALS AND METHODS**

Twenty random samples of salted "Mugil cephalus" were collected from local markets in Cairo and Giza governorates. Collected samples were kept frozen until be analysed. Muscle contents of histamine, tyramine,  $\beta$ -phenylethylamine, tryptamine, cadaverine and putrescine were determined by High Pressure Liquid Chromatography at Department of Mycotoxines, National Research Center, Giza, Egypt, following the method described by Mietz (1977).

### **Sample preparation and initial extraction**

Each fish muscle sample was placed in a food processor and was ground to mince. Fifty grams of each ground fish sample were extracted with three 75 ml portions of 5% trichloroacetic acid using homogenizer. Each homogenized mixture was centrifuged and the clear extracts combined.

### **Standards**

Histamine, tyramine,  $\beta$ -phenylethylamine, tryptamine cadaverine and putrescine standards were prepared at concentrations of 1mg in 100ml of 5% trichloroacetic acid solution before use.

### **Amine extraction and derivative formation**

Trichloroacetic acid extract was made alkaline with sodium hydroxide and extracted with 5ml por-

tions of n-butanol-chloroform (1+1). The combined organic phase, after the addition of an equal amount of n-heptane, was extracted with several 1 ml portions of 0.2N HCl and the aqueous extracts taken to dryness. An amount of standard preparation equivalent to 100µg of each amine was similarly extracted. The dansylated derivatives of the amines were formed by adding 0.5ml of saturated sodium bicarbonate solution to the residue, then adding 1ml of dansyl chloride reagent (500mg/100ml) using a vortex mixer while adding the reagent. The dansylamines were extracted by adding water and extracting the mixture with several portions of ethyl ether. The combined ether extracts were evaporated to dryness and the residue volumetrically dissolved in acetonitrile (1.0 ml for samples, 5.0ml for standards).

The dansylamines were then quantitatively and qualitatively determined by high pressure liquid chromatography using a reverse phase micro particular type column (Waters Associates µ-Bondapak C-18). A waters associates ALC-202 Liquid chromatograph with Mode 660 solvent programmer, a U6K injector and differential U.V. (254nm) detector were used.

A linear solvent program (gradient elution) was selected from 60% solvent B in A to 100% B in 30 minutes, at a constant flow rate of 1.0 ml/min where solvent A was 10% acetonitrile in 0.02 N acetic acid and B was 10% 0.02N acetic acid in

acetonitrile + methanol (1+1). Instrument conditions were adjusted to give 1/2 scale response (0.32 AUFS) for each standard for a 10µl injection (200 ng each standard). Each of the six compounds was calculated for each sample and expressed as mg biogenic amine/100gm sample.

## RESULTS AND DISCUSSION

Table (1) declared that about 70% of salted "Mugil cephalus" samples containing detectable level of tyramine, however β-phenylethyamine was detected in 50% samples. Meanwhile histamine and tryptamine could be detected in 45% samples, cadaverine in 40% and finally putrescine in 30% of the examined samples.

Data illustrated in table (2) revealed that variable quantities of biogenic amines could be detected in examined salted "Mugil cephalus" samples. The highest content was for histamine followed by tyramine then β-phenylethylamine. While tryptamine, cadaverine and putrescine were present in relatively lower levels.

Histamine level (7.8-48mg/100gm) were lower than that reported by Taylor (1985) in sugar, salted herring and Lebedzinska et al. (1990) in salted herring and siberian salmon. This is can be attributed to the difference in type of fish, production conditions availability of oxygen and sodium chloride concentration. Silla Santos (1996) recorded that histamine formation was inhibited by

salting during storage of mackerel muscle at 5°C, however at 25°C the inhibition effect was proportional to the increase in brine concentration, moreover the author concluded that oxygen supply also appears to have significant effect on the biosynthesis of biogenic amines.

Scombroid poisoning is usually a mild illness with a variety of symptoms although death may ensue at some circumstances, such as taking monamine oxidase inhibitor (MAOI) medications or the presence of other biogenic amines, such as cadaverine or putrescine, may potentiate its toxicity (Stratton and Taylor, 1991 and Taylor, 1986).

It is worthy to mention that ingestion of 70-1000mg histamine will usually cause clinical symptoms of intoxication (Henry, 1960), however Blackwell et al. (1969) reported that less than 225mg histamine taken orally usually does not produce symptoms, although susceptible subjects might be adversely affected by smaller quantities. Askar and Treptow (1993) have suggested histamine at a concentration of 500mg/kg to be hazardous for human health, meanwhile 10mg/100gm histamine in all kinds of salted fish is the permissible limit indicated by the Egyptian Organization for Standardization and quality control (EOS) (1989). Symptoms of scombroid poisoning include nausea, vomiting, facial flushing, intense headache, epigastric pain, burning sensation in the throat, dysphagia, thirst, swelling of the lips, and urticaria (CDC, 1973).

Concerning tyramine level (2.94-23 mg/100gm) reported herein, it was lower than that reported by Nuessle et al., (1965) in pickled herring, Sen (1969) in salted dried fish however Mower and Bhagavan (1989) reported a lower level in salted ziganid fish.

$\beta$ -phenylethylamine, tryptamine, cadaverine and putrescine levels (2.8-21.8 mg/100gm, 1.45-11 mg/100gm, 0.5-14.5 mg/100gm and 1.84-8.0mg/100gm respectively) were higher than those recorded by Tsai et al. (2006) in fermented fish products, meanwhile Mah et al. (2002) reported also a lower levels for cadaverine but nearly similar level for putrescine in salted and fermented fish products.

Tyramine and  $\beta$ -phenylethylamine have been proposed as the initiators of hypertensive crisis in patients treated with MAOI and of dietary-induced migraine (Stratton et al., 1990; Koehler and Eitenmiller, 1978), moreover tyramine has also been identified as the major mutagenic precursor in animal (Ochiai et al., 1984). Shalaby (1993) reported that 6mg total tyramine intake is a dangerous dose for patients receiving monoamine oxidase inhibitors (MAOI), while 3mg of phenylethylamine causes migraine headaches in susceptible individuals (Sandler et al., 1974).

Rice et al. (1975) reported that although tryptamine has a pharmacological action similar to tyramine, there are no report of tryptamine intoxi-

cation or of hypertensive crises due to tryptamine. However some researchers suggested that the schizoid symptoms of auditory or visual hallucinations could be due to an inborn deficit in the MAO enzyme, allowing small amounts of tryptamine from foods to build up in the body (Food Intolerance Network Factsheet, 2006).

Putrescine and cadaverine have been identified as potentiators that increase the toxicity of histamine to humans by depressing histamine oxidation (Lehane and Olley, 2000; Ibe et al., 1991; Bjeldanes et al., 1978 and Arnold & Brown, 1978).

Moreover, putrescine and cadaverine have also

reported to be the potentially carcinogenic by converting to nitrosamines (Warthesen et al., 1975 and Bills et al., 1973).

In conclusion biogenic amines in Egyptian salted "Mugil cephalus" may constitute a public health hazard. Some of these amines when found in moderate levels can not produce a direct toxicity, unless large quantities are ingested, but these levels may be potentiated either by certain pharmaceutical agents taken by consumers or by presence of other amines in the same fish or other food item.

**Table (1):** Positive samples for biogenic amines in examined salted "Mugil cephalus" (n=20).

	Histamine	Tyramine	$\beta$ - Phenylethylamine	Tryptamine	Cadaverine	Putrescine
No. of positive samples	9	14	10	9	8	6
%	45	70	50	45	40	30

**Table (2):** Biogenic amines content among positive examined salted "Mugil cephalus" (mg/100 gm).

	Histamine	Tyramine	$\beta$ - Phenylethylamine	Tryptamine	Cadaverine	Putrescine
Minimum	7.8	2.94	2.8	1.45	0.5	1.84
Maximum	48.0	23.0	21.8	11.0	14.5	8.0
Mean	23.71	13.88	9.93	6.31	5.67	4.18
*SD	14.58	7.53	6.24	2.75	4.83	2.44

\* SD: Standard deviation.

Generally biogenic amines in food can be controlled by strict use of good hygiene in both raw material and manufacturing environments with corresponding inhibition of spoilage microorganisms and finally proper storage temperature. In addition, it is of great importance to establish regulatory limits for all biogenic amines which may be found in salted fish to safeguard public health.

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## تأثير الأميونات الحيوية في سمك البورى المملح (الفسيوخ) على الصحة العامة

اسامة على عطاالله - هيام عبدالعال منصور

قسم الرقابة الصحية على الأغذية - كلية الطب البيطرى - جامعة القاهرة - مصر.

توجد الأميونات الحيوية فى العديد من المنتجات الغذائية والتي تشمل منتجات الأسماك ، اللحوم والألبان ونظرا للسمية الشديدة لهذه الأميونات الحيوية كانت هذه الأميونات ومدى تواجدها فى شتى المنتجات الغذائية محل بحث مستمر . ويهدف هذا البحث إلى تقديم معلومات عن مدى تواجد هذه الأميونات الحيوية فى سمك البورى المملح (الفسيوخ)، وفى هذا الصدد تم تجميع عشرين عينة عشوائية من سمك البورى المملح (الفسيوخ) المتواجد فى الأسواق وتم تحليل هذه العينات من حيث مدى تواجد وكمية الهستامين ، التيرامين ، البيتافينيل ايثيل أمين ، التريبتامين، الكدافرين، والبيوترسين وذلك باستخدام جهاز الكروماتوجراف السائل ذو الضغط العالى . ولقد أظهرت النتائج أن كلا من التيرامين ، البيتافينيل ايثيل أمين ، الهستامين التريبتامين، الكدافرين والبيوترسين كانوا متواجدين بنسبة ٧٠٪ ، ٥٠٪ ، ٤٥٪ ، ٤٠٪ ، ٣٠٪ من العينات التى تم فحصها على التوالى . والجدير بالذكر أن بعض هذه الأميونات كان متواجدا بمستويات معتدلة والتي قد لا تؤدى إلى التسمم أو الضرر المباشر إلا اذا تم تناول هذه الأسماك بكميات كبيرة . أو فى حالة وجود بعض الأميونات الحيوية الأخرى مثل الكدافرين والبيوترسين والتي من شأنها أن تزيد من التأثير السام للهستامين وأيضا عند تناول بعض الأدوية المثبطة لأنزيم المونو أمين أوكسيديز والتي تزيد من التأثير الضار للهستامين والتيرامين والبيتافينيل ايثيل أمين . ومن هنا فهذه النتائج أوضحت مدى أهمية التحكم فى تكوين هذه الأميونات الحيوية فى الأسماك المملحة للتأكد من سلامتها وحماية صحة المستهلك .