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Studying of Histamine Concentration and Quality Parameters in Some Local Soft cheese and Imported Processed Cheese Types in Basrah Markets

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Abstract

Histamine is an important biogenic amine, formed by decarboxylation of free amino acid histidine. Presence of histamine in food causes allergy to some peoples, so it is important to determine level of this compound in foods like cheese. This study was carried out to extract and estimate histamine in nine local and imported cheese categories in Basrah city markets, by using ELISA. The mean concentrations of histamine in cheese samples ranged from (0.2725 - 23.5025) mg/Kg in Arab cheese and Dhafayer cheese respectively. the results showed that concentrations of histamine didn't exceed permitted level of 100-200 mg/kg. The study also conducted to ensure that the cheese products conform to the Iraqi standard for canned foods.

Introduction

Biogenic amines (Bas) can be defined as the compounds that produced by microorganisms able to remove carboxyl groups from amino acids. It can be present in plants and animals *[1]*. Some species of Gram positive and negative bacteria can produce some biogenic amines such as *Escherichia*, *Bacillus*, *Shigella*, *Lactococcus*, lactic acid bacteria and others *[2]*.

Histamine produced after removing of carboxyl group from the amino acid histidine *[3]*. It is the biogenic amine which has been studied greatly by several researchers according to its toxic effect. Histamine could cause expansion of blood vessels and arteries which cause the headaches, blood hypotension, edemas and digestive diseases *[4]*. The poisoning by histamine happened after minutes or hours of ingestion of food contained high concentration of this compound *[5]* and *[6]*. Histamine is a dangerous component when it presents in high concentration in certain foods such as fermented foods, marine foods and dairy products *[7]*. Fresh milk contains low amounts of histamine (less than 0.3 mg / kg) *[8]*.

Cheese may be contribute to poisoning of histamine, number of poisoning cases via this compound were recorded in some European countries specially when histamine formed in high levels, 100 mg/ 100 g of cheese *[9]*. The first reported case of histamine poisoning from cheese was in 1967 and caused by Gouda cheese. The United states and the EC didn't set a limits for histamine in cheese, but some studies had suggested 900 mg/ 1 kg cheese for the sum of histamine, tyramine, putrescine, and cadaverine *[10]*. Legal limit for histamine, according to the Netherlands Institute for dairy research, has been set between 100-200 mg/kg *[2]*. Ehsani *et al*. (2012) *[11]* recorded the acceptable limit of histamine in cheese, 100 mg / kg. Also, Švarc-Gajic´and Stojanovic´(2011) *[12]* studied the concentration of histamine in ten types of cheese using electro-analytical method and values ranged from 27.91- 244.08 mg/kg. Histamine has been studied by *[11]*

in three categories of Iranian cheese by using Ion-Exchange chromatography, the results of their study stated that the histamine concentration ranged from 4.99 mg/km in Feta cheese to 42.6 mg/kg in Kope cheese. Rohani *et al*. (2013) *[13]* estimated biogenic amines in 85 types of cheese via HPLC, the concentrations were ranged between 2.34 - 1102.24 mg/ kg. Al-khaeun (2016) *[14]* used ELISA technique to determine histamine in some types of ripening cheese such as Cheddar, Gouda, Mozzarella, Edam cheese and smoked Hajdu, the concentrations were 4.267, 2.433, 1.167, 3.667 and 1.833 mg/ 100 g , respectively, the researcher mentioned that the content of histamine in cheese was associated with the increase of *Lactobacillus* bacteria. the conditions of cheese processing represented ideal environment for the production of biogenic amines because of the bacteria and its activity, proteolytic, the process of maturation and duration as the latter lead to produce free histidine from casein which represents the starter material to convert histidine to histamine.

The food label can be described as a substantial and direct means of communicating product information between buyers and vendors. It can be used to help consumers to differentiate between individual foods and brands to make informed purchasing choices *[15]*. When consumers feel there is some health problems such as allergy or intolerance in the household, they use food labeling, specially when they obliged to chose one of two products or if they get anew product for the first time. Generally, buyers read the food label at the time when they buy foodstuffs *[16]*.

According to available information, there are three categories of food labels that used to transport information to consumer about health and nutrition, the nutrition labels, ingredient lists and claims *[15]*. In this study, nine types of cheeses, including seven imported and two domestic, were used to write down some information about the trademarks to ascertain their compliance with the Iraqi standard regulation. The other important side is the estimation of histamine in all samples to ensure the validity of the cheese categories for human consumption.

Materials and methods

A. Sampling

9 cheese types have been used in current study. Arab cheese, Dhafayer cheese, Almarai, Lebna, Sabah, Abo-al walad, and Luna cream cheese were manufactured from fresh cheese. Craft and Goody cheese were made from ripened cheese. All samples were collected from local and super markets in Basrah city, then taken to laboratory to complete preparation and analyses.

B. The labeling

For the cheeses types used in this study, the information on their packaging were recorded in order to compare them with the Iraqi standard regulation that deal with foodstuff labeling *[17]* and *[18]*.

C. Extraction and determination of histamine

Samples were prepared to determine histamine according to procedure mentioned in DEE3100 ELISA kit. 10 gm of cheese were homogenized in 90 ml of distilled water for 1-2 minutes by using blender. Then 1 ml of the suspension was pipetted to an Eppendorf-tube and centrifuged for 5 minutes at maximum speed. After removing lipid layer by suction, 20 µl of supernatant were taken and diluted with 10 ml of distilled water. The remaining steps of histamine estimation were done as mentioned in ELISA kit.

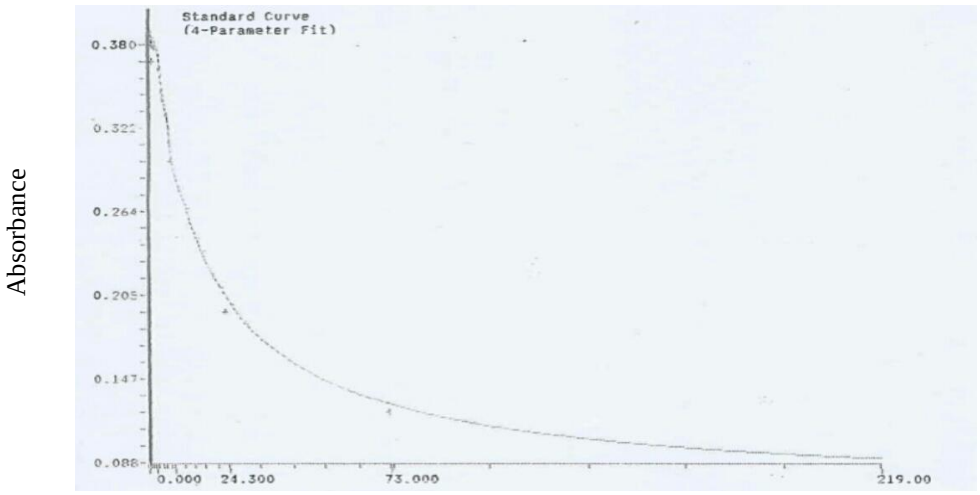

Figure- 1: Histamine standard curve of ELISA

D. Statistical analysis

Data in current study were subject to one-way analysis of variance (ANOVA) using XL Stat program for Windows. Differences Between the means were tested by Duncan's multiple range tests. The level of significance was chosen at $P<0.05$ and the results are presented as mean *[19]*.

Results and discussion

One of the most important conditions for the marketing of food products is labeling. As it is the way that connects between sellers and buyers, it makes the latter fully aware of all what he buys of foodstuffs, especially canned foods. Results in (*Table*: 1) showed the labeling information for the cheese samples used in the study. Two of the samples were locally made and are not subject to Iraqi standard regulation that deal with foodstuff labeling *[17]* and *[18]*. For other samples, the information on the package was written in a manner that conformed to the Iraqi standard regulation, and all the information about the product inside it were fixed, but there is a problem, the font used in writing was very small in some samples, as it is difficult for a large number of consumers to read written information, especially when the consumer read information during the purchase of food, for example the manufacturing and expiry date.

Table- 1: The labeling form of the nine cheese types

Information / Samples	Trade mark	Origin	Production date	Expire date	Net weight	Batch number	Using language
Arab cheese/ Local made	-	Hand made (Basrah)	-	-	-	-	-
Dhafayer cheese/ Local made	-	Hand made (Basrah)	-	-	-	-	-
Almarai cheese/ Imported	+	Saudi Arabia	+	+	+	+	Arabic, English
Lebna/ imported	+	Turkey	+	+	+	+	Turkish, Arabic, English
Sabah cheese/ Imported	+	Iran	+	+	+	+	Persian, Arabic, English
Abo-al walad/ Imported	+	Egypt	+	+	+	+	Arabic, English
Craft cheese/ Imported	+	Saudi Arabia	+	+	+	+	Arabic, English
Luna cream cheese/ Imported	+	Saudi arabia	+	+	+	+	Arabic, English
Goody cheese/ Imported		Saudi arabia	+	+	+	+	Arabic, English

Results in (*Table*: 2) showed mean concentrations of histamine in cheese types of local and imported samples. It appears that histamine levels were different in most samples. Results of statistical analysis showed no significant differences ($P<0.05$) in histamine concentration in most of the studied cheeses, but these differences were found among the Arab cheese (local made) and the other cheese categories.

Biogenic amines concentrations may vary among varieties of cheese, as well as within the varieties. Many factors may be cause the difference of biogenic amines concentration in cheese such as manufacturing processes, decarboxylating bacteria, heat treatments, storage temperature, packaging and starter culture that used in manufacturing processes of cheese *[20]* and *[21]*. Continuous outage of power causes changes in temperature of cold stores, so this will associate in increasing of contaminated bacterial load.

Table- 2: Mean concentrations of histamine in cheese types (mg/kg)

Cheese type	Concentration of histamine (mg/kg)	Mean (mg/kg)	Treatment
Arab cheese (Local made)	20.745-26.260	23.5025^{c}	No heat treatment, no packaging
Dhafayer cheese (Local made)	0.00 - 0.545	0.2725^{a}	heat treatment, no packaging
Almarai cheese (Imported)	0.425 - 3.425	1.9250^{a}	Processed cheese, packaging
Lebna (imported)	0.00 - 5.155	2.5775^{a}	
Sabah cheese (Imported)	0.00 -1.375	0.6875^{a}	
Abo-al walad (régal picon) (Imported)	3.105-6.415	4.7600^{ab}	
Craft cheese (Imported)	5.155 - 8.500	6.8275^{ab}	
Luna cream cheese (Imported)	2.370-6.450	4.4100^{ab}	
Goody cheese (Imported)	13.990-7.075	10.5325^{b}	

Means with different letters in the same column are significantly different ($P<0.05$) for each sample.

All what had mentioned above, in addition to another factors, will contribute in increasing of histamine level in local made cheese, such as exposure of cheese to contamination via bad trading of vendors, absence of hygiene, high temperature of environment which increases microbial counts. So, histamine concentration will be variable.

Conclusions and recommendations

1. The main factor that control the formation and accumulation of histamine is the presence of bacteria that can decarboxylate histidine.
2. All results were less than permissible limits.
3. Histamine accumulates during ripening and storage.
4. The current research recommends to study histamine content in cheese types at different storage conditions.
5. Separation and identification of another biogenic amines such as, tyramine, putrescine, cadaverine, phenylethylamine, ... etc, in another types of cheese in Iraqi markets.
6. Because of the absence of histamine permitted level in cheese within the Iraqi standard regulations, it is good if the results of this research adopted to include the proportion of histamine within the compounds that must has maximum limits in milk and dairy products.

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