African Journal of Advanced Pure and Applied Sciences (AJAPAS)

Online ISSN: 2957-644X

Volume 2, Issue 2, April-June 2023, Page No: 56-62

Website: *https://aaasjournals.com/index.php/ajapas/index*

||Arab Impact factor 2022: 0.87|| SJIFactor 2022: 4.308 || ISI 2022: 0.557

Effect of Thermal Treatment on the 5-Hydroxymethylforfural for Some Types of Honey (Sader and Zater) In the Markets of The Sabratha City

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Received: March 08, 2023	Accepted: April 04, 2023	Published: April 14, 2023

Abstract

This study was conducted from (6/15/2020) to (1/15/2021) to evaluate the effect of thermal treatment on the content of 5-hydroxymethylfurfural (HMF) for honey (Sader and Zater) in Sabratha, where the total of samples collected was 24 A sample including 12 Sader samples and 12 Zater samples. According to White's method, the two types of honey (Sader and Zater), which totaled 24 samples, were thermally treated through incubation at 40 ° C for 1.5 hours. which is based on UV-Vis Spectroscopy measurement at wave lengths from 284 to 336nm, and the results were compared with Libyan Standard No. 1988. Where a high HMF content was obtained at a high temperature and a longer time when heating all 24 samples by incubating the two types (Sader and Zater) and thus it was concluded that the compound HMF is affected by the action of heat with the passage of time. Where the mean and standard deviation was before thermal treatment for Sader honey (15.64 ± 5.37) and for Zater honey (16.28 ± 5.81), and for after thermal treatment for Sader honey (29.43 ± 14.46) and for Zater honey (24.30 ± 6.90).

Keywords: Honey, Incubation, 5-Hydroxymethyl Furfural, White Method, thermal treatment.

Cite this article as: T. A. Ahabaal, E. E. Gunbaej, S. K. Ogbas, M. O. Emdakem and O. S. Ebrahem, "Effect of Thermal Treatment on the 5-Hydroxymethylforfural for Some Types of Honey (Sader and Zater) In the Markets of The Sabratha City," *African Journal of Advanced Pure and Applied Sciences (AJAPAS)*, Vol. 2, No. 2, pp. 56–62, April-June 2023.

1. Introduction

Honey can be used as a dietary supplement [1]. Honey is a concentrated aqueous solution of sugars with a pH of around 6.5. (3.4 - 6.1). Sugars like glucose and fructose account for a significant portion of its composition. It's a blend of nutrients like proteins, amino acids, organic pigments, and more. Aromatic compounds, waxes, minerals, pollen, and enzymes are only a few examples [2, 3]. Honey has long been used in traditional medicine. Antimicrobial and antifungal activity has been discovered in various studies. Furthermore, these findings backed up the use of honey in the treatment of skin lesions [1]. Honey, which is abundant in phenolic acids, flavonoids, ascorbic acid, and carotenoids, is also a good source of antioxidants [4]. Honey can also be used to help regulate diabetes [5]. Honey has been shown in some studies to have anti-cancer properties, especially in terms of inhibiting

tumor cell growth [6]. However, honey may contain some harmful and toxic substances, such as HMF, which may have an effect when ingested by humans. Moreover, multiple studies have shown that the compound has harmful effects on mice, including inducing mutations, being toxic genetically, and being carcinogenic [7]. It also has harmful effects on blood cells, and it has been discovered that it causes tumors and colon cancer [8]. Honey quality is greatly affected by storage time and heating, and HMF is important for determining the conformity of honey due to possible toxic effects [9]. Honey thermal treatment is important for maintaining honey integrity during processing. As a result, pasteurization of honey can be used to safely remove fermentation [10]. Honey should not be heated or treated thermally in any way that affects its basic formulation or consistency, and chemical or biochemical treatments should not be used on crystallized honey [11]. It has been found to enhance the appearance and texture of honey while also preventing crystallization [12]. Furthermore, honey nutrients such as sugars, proteins, and others are vulnerable to thermal decomposition, resulting in a substantial loss of nutritional value. HMF is also a thermal byproduct and a significant predictor of honey consistency [13]. HMF is produced by the caramelization of carbohydrates, especially hexa-polysaccharides, in an acidic medium [3, 14], as well as the degradation of Maillard reaction (MR) products [15]. Ascorbic acid decomposition can result in the formation of (HMF) compound [16]. According to Libyan standard requirements No. 1988, the maximum amount of HMF in honey should be 40 mg/kg for most varieties and 15 mg/kg for citrus honey. The growing concentration of the (HMF) compound on the limit refers to the honey getting older [17] or being subjected to thermal processing during getting and/or filling it [18] or during heating to prevent crystallization [10]. HMF content that exceeds the defined limits suggests that honey has been adulterated, either by marketing old honey as fresh, replacing one honey class with another that is cheaper, or by adding cheap materials to honey, such as corn starch glucose rich syrup, starch syrup, and fructose-rich syrup. Feeding bees sucrose syrup [19] is another form of honey falsification. Physical, chemical, and biological classes can be used to identify honey consistency parameters. Sensory, color, taste, and smell, as well as moisture and viscosity, can all be measured using physical methods. such as [20] and [19], as well as water operation. For the study, quality management has many chemical choices. Acidity, smoke, and sugar recognition as glucose, fructose, and sucrose are some of the chemical parameters of honey. In addition, the Diastase enzyme's efficacy was tested. Detecting and measuring (HMF) may be used in this sense [21]. As a result, and using White's process, we hope to investigate the existence and content of hydroxymethylfurfural HMF levels in some honey samples purchased from a local market in Latakia on the one hand, and to demonstrate the effect of thermal treatment and storage on hydroxymethylfurfural levels in honey on the other. Thermal processing induces changes in the quality of hydroxymethylfurfural. Honey consistency is calculated by the HMF.

2. Materials and Methods

2.1 Collection of Samples

A variety of honey samples are available; 24 samples were collected from various local markets in Sabratha. A variety of honey samples from different botanical sources were analyzed in the study (Sader and Zater). Each sample was labeled with a card identifying the source, location, and date of collection, they were then transported to the University of Sabratha Public Health Colology - Eljmail biochemistry laboratory and the Tajoura Center for Industrial Science, where they were prepared for study. The concentration of 5-hydroxymethylforfural (mg/kg) was calculated.

2.2 Chemical and Reagents

- Carrez solution (I): Dissolve 15 g of Potassium hexacyanoferrate (II) trihydrate (K_4 Fe $(CN)_6.3H_2O$) in Distilled water and makeup to 100 ml.
- Carrez solution (II): Dissolve 30 g of zinc acetate dehydrate (Zn $(CH_3COO)_2.2H_2O$) in Distilled water and makeup to 100 ml.

- Sodium metabisulfite solution $Na_2S_2O_3$ 1 g\500ml.

2.3 Apparatus and Instruments

- Sensitive balance.

- Spectrophotometer (Safas.V-320 UV).

- Incubator.

- Filter paper (general purpose).

- Curettes' (10 mm path length).

- Vortex mixer.

- Distilled water.

- Beaker (200ml).

- volumetric flasks (50ml, 500ml).

- pipits (10ml).

2.4 Effect of temperature on HMF

Thermal treatment of two honey type for 24 samples from Sader and Zater was performed in an incubator at temperatures of (40°C) for time intervals (an hour and half hours). The effect of heat treatment temperature and length on the formation of HMF was investigated, and the HMF concentration was calculated using White's method before and after heating. Each sample test was done three times and the results were given as a mean value and standard deviation.

3. Sample preparation

Approximately 5 g of honey weighed into a 50 ml beaker. Samples have dissolved in approximately 25 ml of distilled water and then transferred quantitatively into a 50 ml volumetric flask (including washing the residue from the beaker with a small amount of distilled water). 0.5 ml of Carrez solution **(I)** were added and mixed, followed by adding 0.5 ml of Carrez solution **(II)**, the mixture mixed and makeup to the mark with distilled water (a drop of ethanol may be added to suppress surface foam). Then the mixture has filtered through filter paper and the first 10 ml of the filtrate have rejected. 5 ml of the mixture has pipetted in each of the two test tubes. 5 ml of distilled water has pipetted to one of the test tubes and mixed well (sample solution). While 5 ml of sodium metabisulfite solution (0.1 %) has added to the second test tube and mixed well (Standard solution).

3.1 Spectrophotometric Analysis

UV-Vis absorbance of the sample solution against the reference at 284nm and 336nm has measured in 10 mm quartz cells within one hour, at the biochemistry laboratory ecology of public health- eljmail used photometric spectrophotometer systems (Safas. V-320 UV). If the absorbance at 284 nm exceeds a value of about 0.5, dilute the sample solution with distilled water and the reference solution with sodium metabisulfite solution to the same extent to obtain a sample absorbance for accuracy.

3.2 Dilution

Dilution of sample and reference solutions have carried out as follows: -

- Additions to test tube sample preparation reference solution
- Initial honey solution 5 ml in 5ml Water.
- 0.2 % sodium metabisulfite solution solute 5 ml.
- Dilution D = final volume of sample solution \10.

3.3 Calculation

HMF (mg/100g of honey) = (A284-A336) * Factor / weight of the sample

A284, A336 = Absorbance reading

Factor= 126 *100 *1000 *100 / 16830 *1000 = 74.87

M.W. of HMF = 126

16830 = Molar Absorptive of the HMF.

4. Results and Discussions

4.1 Statistical Analysis

Statistical significance (P-value sig.) , P is an abbreviation of the Latin word (Probare)، meaning probability, meaning that P-value is a number indicating and indicating probability, meaning that it is a purely statistical term used by the SPSS program for the purpose of interpretation and interpretation of statistical numerical values, and the probability value is used to make a decision about imposing nothing in terms of Accept it or reject it. The value of P-value depends on the level of significance or the level of probability (a), which is the probability of error. And in most applied sciences two levels of significance are chosen, namely (a = 5%) or (a = 1%). The level of 5% is form general, except in some laboratory experiments in which the conditions of the experiment are fine-tuned where the level is 1%. P-value (Sig.) < (a) There are significant differences between the parameters at the probability level (a), P-value (Sig.) > (a) There are no significant differences between the coefficients at probability level (a). The results of the study were interpreted statistically using the T-test. Differences between

honey styles were measured using a T-test (P-value ≤ 0.05). Statistical data analysis was conducted using (SPSS v.22) and we obtained the following results:

First: The effects of temperature degree and duration of heating on HMF level.

After making sure that the data follow a normal distribution, use the Paired T-test for test the effect of temperature degree and duration of heating on HMF level. The obtained results are shown in the following table:

Table (1): shows the mean, standard deviation and T-test for honey type

Types	Before	After	Diff	T-test	P-value
	Mean±St.d	Mean± St.d	Mean± St.d		
Sader	15.64± 5.37	29.43±14.46	13.79±13.34	3.850	0.004**
Zater	16.28± 5.81	24.30 ± 6.90	8.03 ± 6.97	3.986	0.002**

Source: From SPSS analysis based on the sample data. ** Significant at 1%.

The average difference between (before - after) of heating of the Sader form is (13.79), with a standard deviation of (13.34), which is the highest average, as shown in Table (1) above. It is also apparent that the mean difference for Zater form is (8.03), with a standard deviation of (6.97), and that these variations are considered important at the levels of P-value (Sig.) (a), where (a) = (1 to 5%) P-value of sader (0.004) and P-value of zater (0.002), respectively, any P-value range (0.01 to 0.05). We conclude from this that temperature and duration heating has a significant effect on the Hydroxymethyfurfural level at the level of 1%.

Resolution: the difference between the mean sample and the difference is statistically significant.

The following diagram shows the average HMF for the honey types:

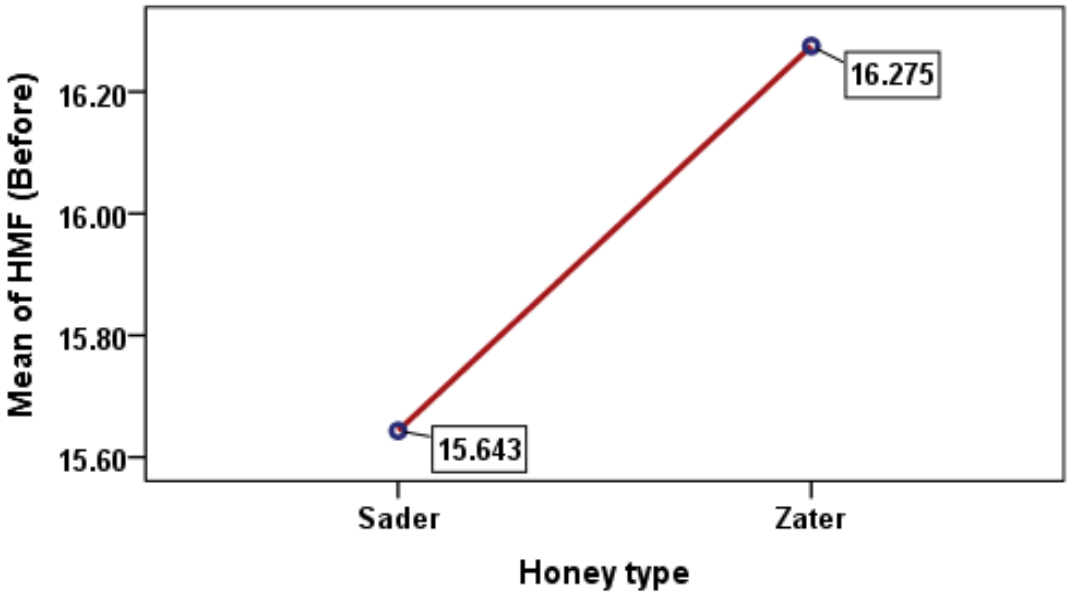

Fig 1: HMF Content of honey before of heating

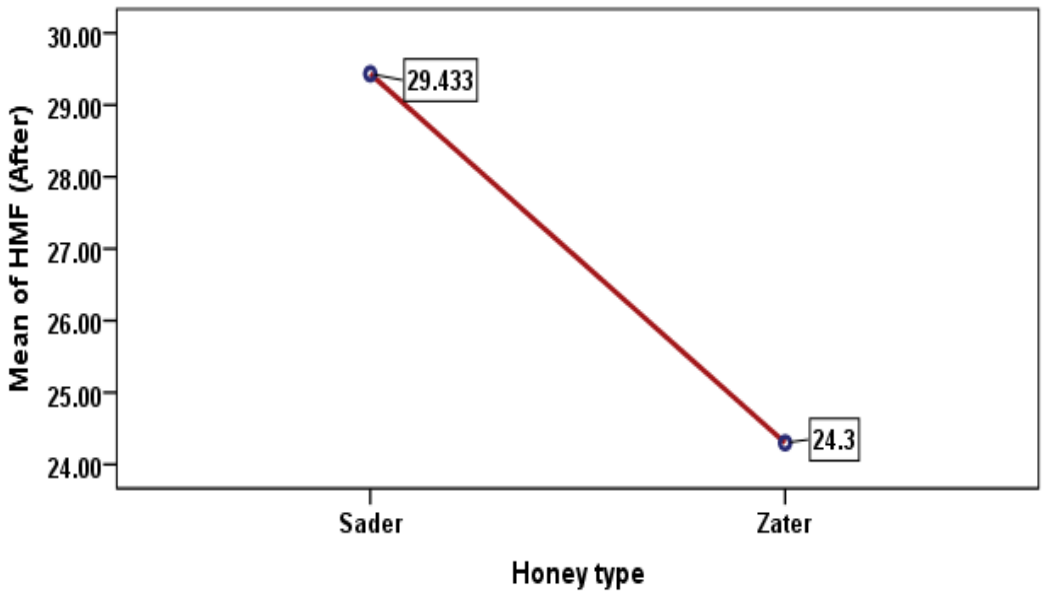

Fig 2: HMF Content of honey after of heating

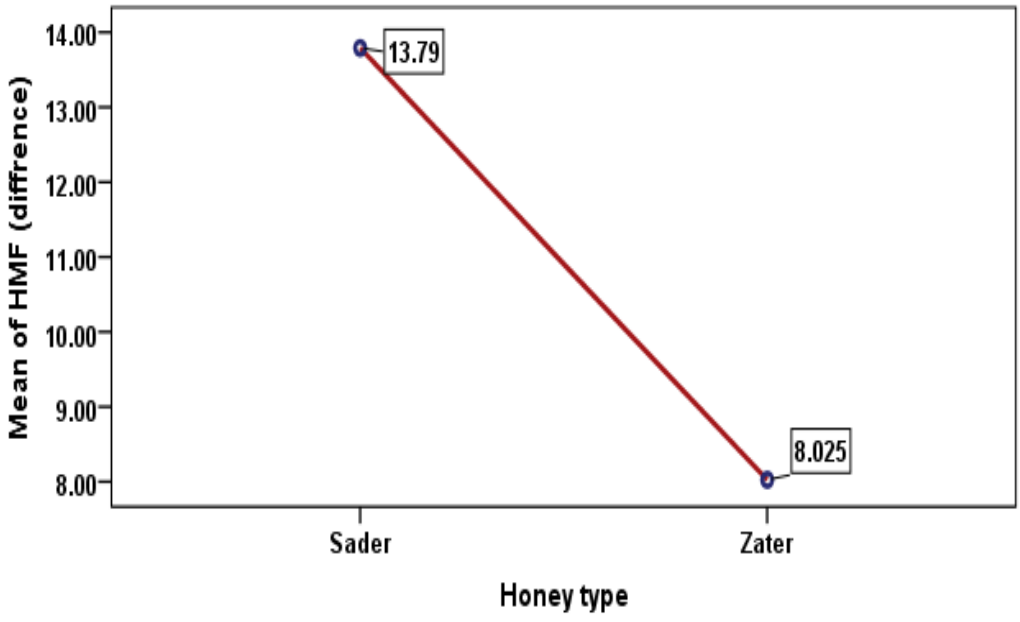

Fig 3: HMF Content of honey for average difference

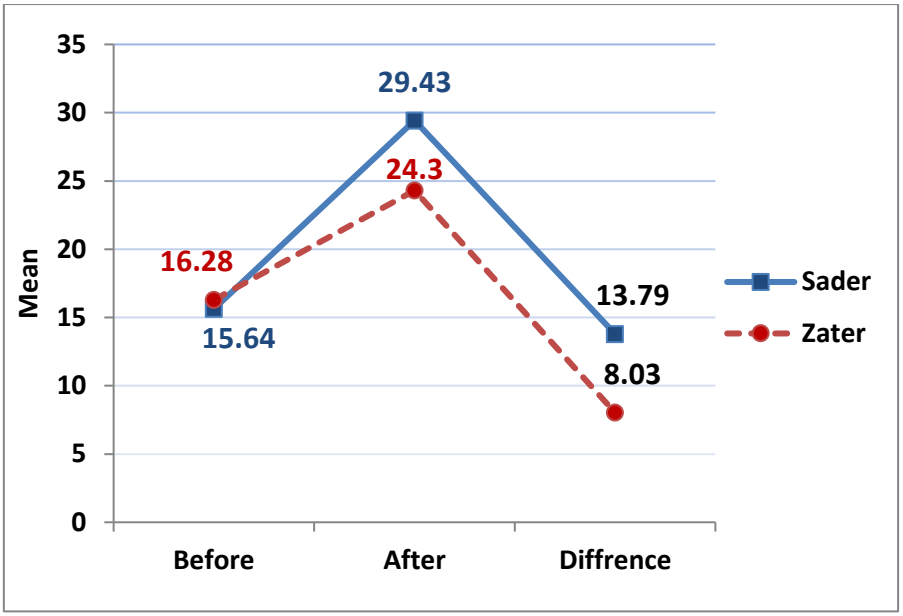

Fig 4: HMF average content of honey for (Before –After – difference).

Second: Spiked HMF

Content of honey in function of temperature degree and time of heating.

The aim of the analysis is to make is to study the effect of temperature and duration of heating on Hydroxymethyfurfural level. The obtained results are shown in the following table.

Table (2): HMF level present in the thermally treated honey samples under duration of heating, and percent recoveries of spiked samples.

Honey type	(HMF) concentration before Heating (mg/Kg)	Thermal treatment time (hr.)	Thermal treatment by inceptor 40 °C Mean ±St.d	(HMF) Recovery %
Sader	15.64	1.5	29.43 ± 14.46	88
Zater	16.28	1.5	24.30 ± 6.90	49

Source: From SPSS analysis based on the sample data.

From Table (2) above, it is clear that there are the effects of heat treatment on the HMF of the honey types at to 40 °C temperature for on hour and half, and HMF content also increased for honey types. Where the honey types after the heating the concentration HMF to Sader (88) (mg/Kg) and Zater (49) (mg/Kg).

We conclude that The HMF content of honey types affected significantly from time of heating and temperature, because According to the previous descriptive measures (averages), it is clear that two are differences in the mean concentration of the compound 5-hydroxyl methyl furfural according to the type of honey since the normal range allowed for this compound in honey is 40 mg / Kg.

4. Conclusions

A spectrophotometric (white) method for determining HMF content in honey samples was developed in this study. For HMF quantification, high resolution spectrometry proved to be a useful method, allowing standard deviations to be held to a minimum. HMF concentrations that are too high are a symptom of honey that has been overheated in the past. Although heating had a major impact on HMF content in honey samples, HMF content turned out to be a reliable predictor of Incubator heating, It's worth noting that not all of the energy supplied by Incubator irradiation was used to counteract the mechanism of honey heating, which couldn't consume all of the Incubator propagated to the cavity and thus lost some energy to the cavity's other sections and surroundings. As a result, the constituents of honey can be kept healthy. HMF content increased in all honey samples, but not nearly as much as it did in the unheated sample. As a result of the damage to enzyme activity and the honey composition, heavy heating causes an increase in HMF content. It is currently believed that when large quantities of honey are stored correctly, it is realistic to stick to the stricter internal requirement for honey, which specifies that it should be kept cool and dark and and should be consumed when fresh.

5. References

[1] Kumar KB, Bhowmic D, Chiranjip Biswajit, Chandria MR. Medicinal uses and health benefits of Honey: An Overview. Journal of chemical and pharmaceutical research 2010; 2(1):385-395.

[2] Yassine M. Food chemistry a textbook for college of pharmacy undergraduate students level -super senior, an edition in Arabic. Tishreen university publications, Latakia, Syria, 2009-2010.

[3] Belitz HD, Grosch W, Schieberle P. Food chemistry, 4th revised and extended edition 2009; 19:906-930.

[4] Khalil MI, Sulaiman SA, Boukraa L. Antioxidant Properties of Honey and Its Role in Preventing Health Disorder. The Open Nutraceuticals Journal 2010; 3:6-16.

[5] Nasrolahi O, Heidari R, Rahmani F, Farokhi F. Effect of natural honey from Ilam and metformin for improving glycemic control in streptozotocin-induced diabetic rats.Avicenna Jou storage of jams and fruit-based infant foods. Food chemistry Spain, Vol. 85, 2004, 605–609. Of phytomedicine 2012; 2:212-221.

[6] Oršolic N. Bee honey and cancer. Journal of apiproduct and apimedical Science 2009; 4:93-103.

[7] Teixidó E, Santos FJ, Puignou L, Galceran MT. Analysis of 5 hydroxymethylfurfural in foods by gas chromatography–mass spectrometry. Journal of chromatography A 2006; 1135:85-90.

[8] Lakka NS, Gosewami N. A novel isocratic RP-HPLC method development and validation for estimation of 5-HMF in Levofloxacin Hemihydrate intravenous infusion. Int. J. Res. Pharm. Sci 2011; 2:45-51.

[9] Chua LS, Adnan NA, Abful-Rahman NL, Sarmidi MR. ~ 334 ~ Journal of Entomology and Zoology Studies Effect of thermal treatment on the biochemical composition of tropical honey samples. International food research journal 2014; 21:773-778.

[10] Kretavhičius J, Kurtinaitiene B, Racys J, Ceksteryit V. Inactivation of glucose oxidase during heat-treatment decrystallization of honey. Žemdirbyste Agriculture 2010; 97:115-122.

[11] Codex alimentarius. Codex standard for honey, CODEX STAN 1981, 2001; 12:1-8.

[12] Thrasyvoulou A, Manikis J, Tselios D. Liquefying crystallized honey with ultrasonic waves. Apidologie 1994; 25:297-302.

[13] Spano N, Casula L, Panzanelli A, Pilo MI, Piu PC, Scanu R *et al.* An RP-HPLC determination of 5-hydroxymethylfurfural in honey The case of strawberry tree honey Talanta, Italy 2006; 68:1390-1395.

[14] Gökmen V, Senyuva H. Rapid determination of hydroxymethylfurfural in foods using liquid chromatography-mass spectrometry, (Food industry) Ankara, Improved method for the determination of hydroxymethylfurfural in baby foods using liquid chromatography-mass spectrometry. Journal of agricultural and food chemistry, Vol. 54, 2006, 2845- 2849.

[15] Rada-Mendoza M, Sanz ML, Olano A, Villamiel M. Formation of hydroxymethylfurfural and furosine during the storage of jams and fruitbased infant foods, Volume 85, Issue 4, May 2004, Pages 605-609.

[16] Koca N, Burdurlu HS, Karadeniz F. Kinetics of nonenzymatic browning reaction in citrus juice concentrates during storage. Turk j agric for, 2003; 27:353-360.

[17] Tosi EA, Ré E, Lucero H, Bulacio L. Effect of honey high-temperature short-time heating on parameters related to quality, crystallization phenomena and fungal inhibition. Lebensm.-Wiss. u.-Technol 2004; 37:669-678.

[18] Grujić S, Komić J. Classification of honeys from three geographical regions based on their quality control data. Quality of life 2012; 3:13-26.

[19] Cordella C, Faucon JP, Cabrol-Bass D, Sbirrazzouli N. Application of DSC as a tool for honey floral species characterization and adulteration detection. Journal of thermal analysis and calorimetry 2003; 71:275-286.

[20] Bogdanov S. Harmonized method of the International Honey Commission. International honey commission, Switzerland, 2002, 28-30.

[21] Ayansola AA, Bango AD. Physico-chemical evaluation of the authenticity of honey marketed in Southwestern Nigeria. Journal of basic and applied scientific research, 2011; 12:3339-3344.