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QUALITY EVALUATION OF HONEYS IN NORTHWESTERN STATES OF NIGERIA

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ABSTRACT

Natural honey is one of the highly needed products because of its nutritive and medicinal properties. This research is aimed at evaluating the quality of commercial honey and comparing the basic quality parameters (pH, HMF, free acidity, sugars, and EC levels) of each sample with standards. Honey samples obtained from local sellers and beekeepers from seven States in Northwest Nigeria (Kano, Kaduna, Jigawa, Katsina, Sokoto, Zamfara and Kebbi) were analyzed using standard analytical methods. Results indicated that most of the parameters tested, pH (3.62-5.51), Hydroxymethylfurfural, HMF, (43.14-55.91 mg/kg), sucrose (6.18-20.46%) and free acidity (40.08-52.25 meq/kg) were above the permissible range set by the International Honey Commission (IHC). However, lower values were recorded for reducing sugars (44.08-65.62%) and EC (94.6-591.5 µS/cm) in most of the honeys from local sellers except in honeys from Zamfara and Kebbi States which compared favourably with the beekeeper's honeys (control samples). Generally, results from this study indicate that large percentage of the honey products sold locally in the Northwestern Nigeria are suspected to be adulterated while honeys obtained directly from beekeepers were found to closely comply with IHC standards.

Keywords: Adulteration, beekeepers, honey, quality evaluation.

INTRODUCTION

Honey is a natural food produced by honey bees from nectar of flowers and the secretion of plants caused by certain insects [1]. It is one of the major bee products, a semi liquid, sweet and flavoured food stuff produced from nectar of nectarines of flowers, or secretion of plant-sucking insects which the bees collect, transform, deposit, dehydrate, store and leave in honeycomb to ripen and mature [2]. Natural honey is a liquid accepted by many generations, traditions and civilizations, both ancient and modern. It is one of the products most widely sorted for, due to its nutritional and medicinal properties. These properties are attributed to the influence of different groups of substances that honey contains [3]. Honey produced by the honey bee is a natural

super saturated sugar solution which has been seen as a highly nutritive food and is composed of a complex mixture of carbohydrates, minerals, vitamins, aromatic compounds, flavouring and enzymes with the water content of about 17–20% [4]. The major compositions of honey are carbohydrates and water [5]. It is a high-energy carbohydrate food as the honey sugars are easily digestible as those in many fruits [6].

Commercial honeys are sold in separate containers or in bulk. They are available as raw or processed honey. The latter is usually pasteurized, clarified, or filtered and at times fortified. Raw honey is of the highest organic quality and is regarded as 100% pure [7]. Honey sold as such does not have added food ingredients, including food additives, or any other additions. Honey should not have any objectionable matter, flavour, aroma, or taint absorbed from foreign matter during its processing and storage. It should not ferment or effervesce. No pollen or constituent particular to honey may be removed except where this is unavoidable in the removal of foreign inorganic or organic matter. Honey should not be heated or processed to such an extent that its essential composition is changed and/or its quality is impaired. Chemical or biochemical treatments shall not be used to influence honey crystallization [8].

Honey is very popular and has a high economic value. Concerns over making greater profits lead to the production of adulterated honeys [9, 10]. Honey adulteration is a global concern; it has negative effects on the nutrition and health of consumers and has become a common practice because of the high demand and limited availability of the product. Since honey is a mixture, it has become one of the most highly adulterated products [11]. Honey adulteration can be direct or indirect. Direct adulteration means that a substance is added directly to the honey, while indirect adulteration is feeding the honeybee with adulterating substances [12]. Indirect adulteration is peculiar to beekeepers, while direct adulteration is generally done by honey parkers. Direct adulteration with sucrose may give a sucrose content of 30% or more, which is considered unsafe for human consumption [13].

Honey acquires its physicochemical properties through nature and worker bee's conversion of nectar to honey. Colour and flavour of honey depend largely on the kind of plant species foraged by the worker bees. Honey is generally evaluated by a physicochemical analysis of its constituents [14]. Studies of the physicochemical properties have great importance for the determination of honey quality [15]. The constituents such as minerals, moisture content, reducing sugars, electrical conductivity, free acidity, sucrose content and HMF have influence on

nutritional quality, granulation, the storage quality, flavor and texture of the honey [16]. These components are of great importance as they influence the keeping quality, granulation, texture, as well as the nutritional and medicinal efficacy of honey [17]. The medicinal value of honey is also due to these constituents. Therefore, the International Honey Commission has proposed these constituents as quality criteria for honey [18].

The aim of this research work is to evaluate the quality of commercial honeys available in markets from Northwestern Nigeria for the purpose of assessing the state of adulteration in the samples. This is achieved through comparison of physicochemical parameters of the adulterated samples with those of raw natural honeys from credible beekeepers, and also with IHC standards. Sulieman *et al.* evaluated the quality of honey obtained from different sources [19]. Their results indicate that there were no significant differences in most of the chemical and physicochemical characteristics of natural honey and industrial honey. The results also indicated that the various honey types contained sucrose (10.7-3.48%), fructose (14.74-39.01%), glucose (14.09-35. 6%). In another study, Moniruzzaman *et al.* evaluated the physical, biochemical and antioxidant properties of four Malaysian monofloral types of honey. The mean pH, moisture content, EC and TDS of Malaysian honey obtained were 3.90 ± 0.12, 17.01 ± 3.07%, 0.59 ± 0.17 mS/cm and 294.87 ± 81.96 ppm, respectively [20].

Most of the previous studies mainly investigated the qualities of natural and industrial honeys while the present study investigates those of natural and locally managed honeys. Literatures indicated scanty information on the levels of adulteration of locally managed honeys in Northwestern Nigeria. Therefore, this short finding can bridge such information gap.

MATERIALS AND METHODS

The equipment used in this study include digital pH meter (Model No.: 361), conductivity meter (ISE Meter imacimus 10), UV Visible spectrophotometer (Shimadzu UV-1900 spectrophotometer) and analytical balance (Model AB54, Mettler Toledo). All the reagents used (sodium bisulfate, sodium hydroxide and hydrochloric acid) were of analytical grade purchased from Merck, Germany.

Sampling

Exactly one hundred and five (105) honey samples were randomly collected within Northwestern States (Kano, Jigawa, Kaduna, Katsina, Sokoto, Zamfara and Kebbi), Nigeria. Each State was

divided into three senatorial districts: Central, North and South. Five samples were collected from each district including one sample to be used as control making a total of 15 samples from each State. The samples from Central senatorial districts in Kano State were designated KN CI, KN C2, KN C3 and KN C4, while the Control sample was designated KN CC. Also, the samples from Kano North were labeled as KN N1, KN N2, KN N3 and KN N4 with the Control sample as KN NC. Likewise, samples from south districts were labeled as KN SI, KN S2, KN S3 and KN S4, while the Control sample as KN SC. This identification trend was used in all other States. The honey samples were obtained commercially while the pure honey samples used as control were obtained directly from bee keepers in each State. All the samples were collected in sterile containers, labeled and stored in a refrigerator in airtight plastic containers until analysis. Figure 1 shows the map of the sampling sites.

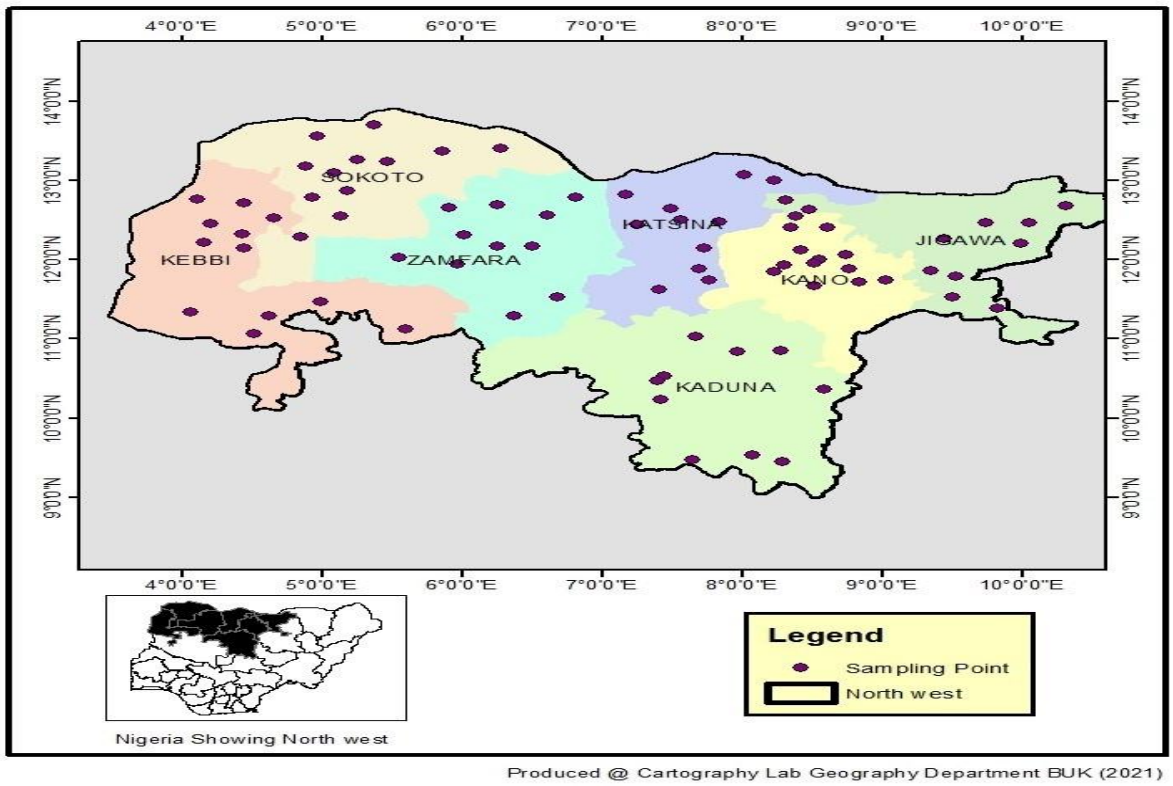

Fig. 1: Map of the sampling sites

Determination of pH

Exactly ten grams (10 g) of honey was weighed accurately and diluted in 75 cm^3 deionized water. The pH meter electrode was immersed in the honey solution after calibration and the pH value was recorded [2].

Determination of Electrical Conductivity

A 20% (w/v) solution of honey was prepared and using a conductivity meter, the electrical conductivity (EC) of each sample was measured [3].

Determination of Hydroxymethylfurfural (HMF)

Exactly five grams (5 g) of honey was weighed into a 50 cm^3 beaker. The sample was dissolved in approximately 25 cm^3 of deionized water and transferred into a 50 cm^3 volumetric flask. Carrez solution I (0.5 cm^3) and 0.5 cm^3Carrez solution II were added and mixed. The solution was made to the mark with deionized water with a drop of ethanol to suppress foam. It was then filtered using a filter paper. The filtrate (5 cm^3) was pipetted into two test tubes, 5 cm^3 of water was added to one of the test tubes and mixed well. Again 5 cm^3 of sodium bisulfate solution (0.2 %) was added to the second test tube and mixed well (the reference solution). The absorbance values of the sample solution against the reference at 284 and 336 nm in a 10 mm quartz cells were determined within one hour [18].

Determination of Free Acidity

Free acidity as tartaric acid was determined according to the method of Ndife *et a*l.[21]. Exactly ten grams (10 g) of each sample was diluted with deionized water in a 100 cm^3 volumetric flask. It was treated with 0.1M NaOH solution using titration kit, where 3 to 5 drops of phenolphthalein indicator were used. The acidity was determined using the formula.

Free Acidity (meq/kg) = Vol. of NaOH used x Weight of Honey.

Determination of Reducing Sugar

Honey sample (1 g) was diluted in a 100 cm^3 volumetric flask with deionized water. A 50 cm^3 burette was filled with the honey solution. Mixed Fehling's solution (10 cm^3) was pipetted into a 300 cm^3 conical flask, 7 cm^3 of deionized water and 15 cm^3 of the sample solution was added from the burette into the same conical flask. The liquid was boiled on asbestos covered gauze, and further quantities of the sample solution were added to the boiling liquid until the blue colouration was nearly discharged. Then, 3 drops of aqueous methylene blue solution (1%) were added and the titration continued until the indicator was completely decolourized [22].

Determination of Sucrose

Sucrose was determined by inverting a portion of the test solution with acid followed by neutralization with an alkali and titration by the Lane and Eynon method. The inversion was achieved by pipetting 50 cm^3 of the test solution into a 100 cm^3 volumetric flask; 5 cm^3 of concentrated hydrochloric acid was added and left to stand overnight. It was then neutralized with 1 M sodium hydroxide using methyl red indicator (pH 4.2 – 6.3), cooled and made up to the mark [22].

Statistical Analysis

All the tests were done in triplicate and the data were expressed as mean ± standard deviation (SD). Statistical significance was determined using a one-way Analysis of Variance (ANOVA) and the Duncan multiple range test with significant level at 95% ($P<0.05$) were considered significant.

RESULTS AND DISCUSSION

Physicochemical Analysis

The addition of foreign substance(s) to a food modifies certain components or creates an irregularity in its composition [23].

Table 1 presents the mean concentrations of the physicochemical parameters determined in the honey samples.

Table 1: Mean Concentrations of Physicochemical Parameters in Honey Samples Collected from the Study Areas.

States	pH	EC (µS/cm)	HMF (mg/kg)	Free Acidity (meq/kg)	Reducing Sugar (%)	Sucrose (%)
Kano	5.33±0.81	126.9±38.02	52.75±19.32	47.58±6.59	51.35±7.0	17.92±5.22
Jigawa	5.13±0.74	110.5±46.61	49.41±9.80	48.66±11.63	47.60±53.98	18.24±6.23
Katsina	5.37±1.03	107.6±38.84	55.91±12.10	47.66±8.03	53.98±8.38	12.24±7.24
Kaduna	5.51±0.57	140.3±53.22	55.40±5.52	52.25±13.91	44.08±2.44	20.46±3.00
Kebbi	3.71±0.56	172.2±57.60	45.01±15.08	45.66±8.69	55.22±11.09	16.79±6.65
Sokoto	3.62±0.40	94.6±15.31	46.90±6.29	50.33±6.41	53.22±7.41	16.86±4.34
Zamfara	3.87±0.26	591.5±169.2	43.14±14.46	40.08±9.99	65.62±12.79	6.18±1.78
Control	3.81±0.09	533.2±143.83	31.05±8.68	31.95±7.41	73.81±3.35	4.69±0.64
IHC	3.7 – 4.5	≤ 800	≤ 40	≤ 50	≥ 65	≤ 5
NAFDAC		≤800	≤50	≤50	≥60	≤5

From Table 1, the pH values obtained in all the honey samples ranged from 3.62±0.40 to 5.51±0.57. The mean pH values obtained for samples from Kebbi, Zamfara, and the control samples were found to be within the permissible limit of 3.7-4.5 set by the IHC [18]. However, the mean pH value (3.62) from Sokoto samples was slightly below the minimum value of 3.7 set by the IHC. Aljohar *et al.* reported that a relatively more acidic pH in honeys indicates impure product possibly due to improper storage [22]. Additionally, the samples from Kano, Jigawa, Katsina and Kaduna were found to be above the IHC standard. Generally, honey is mildly acidic with an average pH of 3.9 [22]. The mean pH value 3.81 in honey sample from source (control) was not significantly different ($p>0.05$) with samples from Kebbi and Zamfara but was significantly different ($p<0.05$) with samples from Kano, Jigawa, Katsina, Kaduna and Sokoto. The acidic nature of honey is due to the minor acid content, mainly amino and organic acids that are responsible for the characteristic taste of honey. It is an important parameter during extraction and the conservation of honey. It increases the quality constancy and shelf life of honey [24]. Honey samples with a pH above 5 are considered to be of low quality [25]. The low PH in honey is compatible with natural acidic foods, which also contribute to the product's

flavor profile. pH plays an important role, especially during honey extraction and for long term storage [26]. The honey samples, which are blended with high fructose corn syrup, have higher pH value compared to pure honey [27]. The analysis of pH in the honey is considered as one of the quality factors used in the international honey rate [28].

The EC can be considered a good criterion in evaluating the purity of honey [28]. The mean electrical conductivity in honey samples obtained in this study ranged from the minimum of 94.6±15.31 µS/cm to the maximum of 591.5±169.2 µS/cm. Electrical conductivity in honey samples from source 533.2±143.83 µS/cm and Zamfara 591.5±169.2 µS/cm were significantly higher ($p<0.05$) compared with EC values obtained in honey samples from all the study areas. Electrical conductivity is a key parameter for the authentication of honeys. It is an excellent parameter for identifying the purity of honey [3]. All the EC values measured fall below the permissible limits of 800 µS/cm set by both NAPDAC and IHC [8, 18].

HMF in all the analyzed honey samples ranged from the lowest of value of 31.05 mg/kg to maximum of 55.91 mg/kg. HMF values in all the analyzed honey samples obtained had exceeded the permissible limit of 40 mg/kg set by IHC [18] with the exception of samples from source (control). However, samples from Kano, Katsina and Kaduna have exceeded the 50 mg/kg set by NAFDAC [8]. The HMF value of 31.05 mg/kg in honey samples obtained from the source (control) was significantly lower ($p<0.05$) compared with the HMF values in honey samples obtained from all the study areas. HMF concentration is widely recognized as a parameter affecting honey freshness because it is typically absent (or is present in only very small amounts in fresh, well processed honeys), while its concentration tends to rise during processing and/or because of aging or during temperature conditioning to dissolve crystallized fragments, in storage and upon adulteration [29]. Also, high HMF content in suspected samples adulterated with sucrose could be attributed to decomposition of sucrose to glucose and fructose by enzyme invertase, thereby releasing HMF as a by-product of further dehydration of glucose and fructose in the presence of good acidic medium and high temperature. Some Nigerian harvesters use smoke to drive the bees away or to render the insects less active in order to facilitate honey harvesting which may lead to elevated concentrations of HMF in honey [30].

Free acidity of honey is mainly due to presence of organic acids, particularly the gluconic acid and inorganic ions such as phosphate and chloride [31]. The free acid values in all the analyzed honey samples varied from 31.95 meq/kg to 52.25 meq/kg. The results revealed that

free acidity values obtained in all the honey samples in this study were within the recommended ≤50 meq/kg set by both NAFDAC and IHC [8, 18] except in honey samples obtained from Sokoto and Kaduna, that were slightly higher than the maximum permissible level. The free acid value of 31.95 meq/kg obtained from the source (control) was significantly lower ($p<0.05$) compared with free acid values obtained in samples from all the study areas. The low free acid value obtained in samples from the source (control) demonstrates a low level of undesirable fermentation due to high moisture content. The rate of conversion from fructose and glucose to ethyl alcohol and carbon dioxide by osmotolerant yeast and the further formation of acetic acid and water in the presence of oxygen is relatively low, resulting in a small free acidity [26]. Increased free acidity in honey is an indicator of fermentation and transformation of alcohol into organic acid [32].

The reducing sugar in the samples obtained in this study varied from the minimum of 44.08% to maximum of 73.81%. The reducing sugar values obtained in the analyzed samples from all the study areas were lower than the acceptable limits of ≥65% and 60% set by IHC [18] and NAFDAC [8] respectively except in control and Zamfara State honey samples. The maximum value of reducing sugar 73.81% in honey samples from source (control) was significantly higher ($p<0.05$) compared with values obtained from all the study areas. These results confirm that sugars are major constituents of honey. Samat *et al.* [33] confirmed that total reducing sugar of adulterated honey is low compared to pure honey. Addition of water into the honey decreased the reducing sugar content and also the sweetness [23].

The distribution of sucrose levels in the honey samples studied was broad, ranging from the lowest of 4.69 % to maximum of 20.46 %. The sucrose contents in all the analyzed honey samples were far above the acceptable limits of ≤5 % set by both NAFDAC and IHC [8, 18] except in samples from the source and Zamfara. The sucrose value of 4.69 % obtained in honey samples from the source (control) and Zamfara were significantly lower ($p<0.05$) compared with values in honey samples obtained from all the study areas. Although, this sugar has minor importance, its presence can provide information about adulteration of the honey. The sucrose content in honey is analyzed with the purpose of identifying any improper manipulation of honey, and high levels may indicate a variety of adulteration, such as adding cheap sweeteners like other sugars, cane sugar or refined beet sugar, early harvest, indicating that the sucrose has not completely transformed into glucose and fructose; or prolonged artificial feeding of

honeybees with sucrose syrups, resulting in high commercial profits [34, 35]. Feeding with high concentration of sugar diverts the bees from collecting nectar and causes them to collect pollen instead [36]. Also, over heating of the honey sample might denature invertase, stopping the enzyme activity that breaks down the sucrose into glucose and fructose. Thus, sucrose level remains high in the adulterated honey [23]. Hence, it can be concluded that high sucrose levels in a honey sample might be due to the adulteration of honey [37].

CONCLUSION

The results of this study show that adulteration of honey has taken place to some degree and might be by direct addition of adulterants, indirect bee-feeding or by combining with other cheap honey. The results obtained in honey samples from Zamfara and Kebbi States were almost in agreement with standard values or limits and therefore are assumed to be free of adulteration. However, samples obtained from Katsina, Sokoto, Kano, Jigawa and Kaduna were suspected to have undergone some form of adulteration when compared with samples obtained directly from beekeepers and standard acceptable limits set by International Honey Commission and NAFDAC. The findings in this research indicated that most of the honeys from open markets are adulterated.

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