STUDY ON THE QUALITY OF HONEY COLLECTED FROM DIFFERENT FLORAL SOURCES OF BANGLADESH

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CERTIFICATE

This is to certify that the thesis entitled ***STUDY ON QUALITY OF HONEY COLLECTED FROM DIFFERENT FLORAL SOURCES OF BANGLADESH*** *submitted to Department of Entomology, the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in ENTOMOLOGY, embodies the result of a piece of bona fide research work carried out by* ***SADIA AFREEN NIPA, Registration No. 13-05265****, under my supervision and guidance. No part of this thesis has been submitted for any other degree or diploma.*

I further certify that any help or sources of information has been availed of during the course of this investigation has duly been acknowledged.

Dated: June, 2020
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DEDICATED TO

MY

BELOVED PARENTS

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STUDY ON THE QUALITY OF HONEY COLLECTED FROM DIFFERENT FLORAL SOURCES OF BANGLADESH

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ABSTRACT

The aim of this study was to investigate some physicochemical properties of various honey samples collected from beekeepers maintainig *Apis mellifera* hives in different regions of Bangladesh during 1st january 2019 to 30 december 2019: The study was conducted in the disease identification and analysis laboratory under department of entomology In Sher-e-Bangla Agricultural University (SAU). Honey samples were analyzed for determining pH, moisture, optical rotation, brix and fructose content. The study revealed that all honey samples had pH, moisture, optical rotation, brix and fructose content were ranging within 1 to 6; 15.47 to 28.13%; -1.84 to -5,deg cm2 g-1; 69.9 to 82.77% and 34.03 to 64.33 % respectively. Considering the pH, the highest pH (6) was recorded in Mustard honey & Foreign honey (sample no.15 & 20) and lowest pH (1) was in Fake honey (sample no. 18). In terms of moisture, the highest moisture content (29 %) was obtained in Olive honey (sample no. 13.) and lowest moisture content (15.47 %) was found in Mustard honey (sample no. 15.). Regarding the optical rotation,the highest specific rotation (- 5, deg cm2 g-1) was recorded in Mustard honey. And the lowest specific rotation (-1.84, deg cm2 g-1) was in Litchi honey (table 4). Considering of brix, the highest amount of brix (82.77) was found in Mustard honey (sample no.15) and lowest (69.9) was in Olive honey (sample no. 13). Regarding fructose, the highest amount of fructose (64.33 %) was recorded in Khoilsa (Alwoan) (sample no. 6.) and the lowest amount of fructose (34.03 %) was in Mustard honey (sample no. 15.) (Figure 3). All samples were significantly difference of each otherat 5% level.

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ABBREVIATIONS AND ACRONYMS

FULL WORD	ABBREVIATION
And others	*et al.*
Total soluble solids	TSS
Edest (means That is)	i.e.
Bee Hive	BH
Water Bath	WB
Moisture Content	MC
Free Acidity	FA
Diastase Number	DN
Total Polyphenols Content	TPC
Water Insoluble Solids	WIS
Brown Pigment Formation	BPF
Glucose Oxidase	GOx
Namely	Viz.
Species (plural number)	spp.
International Honey Commission	IHC
Hydroxymethylfurfural	HMF
Specific Optical Rotation	SOR
Water Activity	Aw
United Arab Emirates	UAE
Total Soluble Solids	TSS
Gallic Acid Equivalents	GAE
Solid-Phase Microextraction	SPM
Gas Chromatography-Mass Spectrometry	GC/MS
Saccharose Syrup Honey	SSH

CHAPTER I

INTRODUCTION

Honey is one of the oldest food linked to the human history and had always attracted the attention of the man, especially because of their sweetening characteristics. Honey is a sweet natural substance that is produced by honeybees from the nectar (secretion) of living parts of plants. Honeybees collect this material, transform and combine it with specific substances of their own, store and leave in the honey comb to ripen and mature (White and Landis, 1980). Honey is characterised by its aroma, sweet taste and potential biomedical activity. Due to continuous expansion of the world honey market, the importance of apiculture as an industry has also grown. Composition and quality criteria of honey states that; honey should not have any ingredients added; no particular constituent can be removed from it; it does not have any objectionable matter, flavour,aroma processing and storage; and it should not be heated or processed to such an extent that its essential composition is changed and/or its quality impaired.

Honey is commonly adulterated through the addition of sweeteners (e.g. sugar syrups and molasses inverted by acids or enzymes from corn, sugar cane, sugar beet and natural syrups such as maple), from different botanical and geographical origins, heat treatment or improper storage conditions or the filtration or addition of colorants (Bogdanov & Martin, 2002; Potraviny na pranyri, 2013).

This product is consumed worldwide because it is considered a natural and energetic sweetener, with predominance of sugars, glucose, fructose, saccharose (70% of carbohydrates) and the water in which the sugars are dissolved. The composition of honey mainly depends on climatic and environmental conditions and the diversity of the plants flowers from which they are harvested. Honey contains at least 200 substances mainly carbohydrates and water. It also contains minerals, proteins, free amino acids, enzymes, vitamins, organic acids, flavonoids, phenolic acids, and other phytochemicals. In addition, honey is valuable for the treatment of cardiovascular diseases, cancer, cataract, and several inflammatory diseases as well as wound healing.The therapeutic actions of honey are due to its antioxidant and antimicrobial properties. The quality of honey is mainly determined by its sensory, physicochemical and microbiological characteristics.Thecriteria for the physicochemical quality of honey are well specified by the European Community Guidelines.Themain criteria of interest aremoisture, electrical conductivity, ash, reducing and nonreducing sugars, free acidity, diastase activity, and hydroxymethylfurfural (HMF) content. Honey is considered as one of the most important natural antioxidant sources among the foods. It is effective against human-threatening health substances such as oxidant agents. In addition, honey consumption can substantially decrease the risk of cardiovascular diseases, cancers, cataract, and inflammatory diseases. The chemical compositionof honey varies from a sample to another and usually contains major and minor elements. To determine honey varieties, some pollen, physicochemical, microbiological, and sensorial analysis must be taken into account. Honey is a strong concentrated aqueous solution of invert sugar, but also contains a very complex mixture of other carbohydrates (mostly glucose and fructose), amino acids proline, minerals, aromatic substances, pigments waxes and pollen grains (Bogdanov *et al*., 1998; Qiu *et al.,* 1999). However, the presence of unstable compounds, such as enzymes, vitamins and other compounds in honey is also reported by different workers (Coco *et al.,* 1996). The variation in physicochemical properties of the honey samples, such as ash contents, the spectrum of saccharides, the activity of enzymes, hydroxymethylfurfural (HMF), electrical conductivity, pH and optical rotation are due to regional and floral differences (Serra and Ventura, 1995; Terrab *et al.,* 2003).

As Honey depends on biotic and abiotic factors around the beehives, therefore, the presence of heavy metals could be related to its geographical and botanical origin. Preliminary studies also confirmed a correlation between the elemental content of honey and the status of the environment (Kump *et al*., 1996; Caroli *et al.,*1999). It has been observed to have therapeutic properties (exhibiting healing power) in the treatment of digestive, respiratory, cardiac and rheumatic disorders (Abdullah and Abdulaziz, 1998). However, the presence of enzymes such as glucose oxidase produces acids like gluconic acid and hydrogen peroxide. While the lowered pH and oxidizing agent serve as antimicrobials to preserve the honey, the low pH tends to impact a sour taste which makes the honey unpalatable if its level is too high (low pH). In addition, it takes advantage of its three antimicrobial components (hyperosmotic, low pH and presence of hydrogen peroxide). Since ancient times, natural unprocessed honey was used to prevent microbial infections and aid wound healing. Honey is a natural dietary antioxidant whose components are responsible for the redox properties are likely to be flavonoids,phenolic acids, enzymes, vitamins and minerals such as copper and iron. Honey can originate from single or multiple plant species, and its biochemical composition is affected by the floral source (Elbanna *et al*., 2014). Due to features such as its geographical position, climatic conditions and three seasons of the year being suited to honey production, Turkey is one of the richest regions of the world in terms of honey production and variety. It is home to a wide variety of nectar and honeydew honey types, both unifloral and multifloral (Can *et al.*, 2015). The melissoplaynological study is an effective method to determine the pollen inside the honey sample. Taxa of the pollen are usually used to indicate the floral nectar sources utilized by bees to produce honey. The relative pollen frequency is usually used to verify a honey sample as to the major and minor nectar sources. A free radical is an atom, molecule or compound that is highly unstable because of its atomic or molecular structure. Free radicals are very reactive as they attempt to pair up with other molecules, atoms, or even individual electrons to create a stable compound (Wu and Cederbaum, 2003). Therefore reactive oxygen species (ROS) occur and free radicals cause molecular transformations and gene mutations in many types of organisms. This is called oxidative stress and is well known to cause many diseases (Kuçuk *et al.*, 2007). Honey has been an important food for humans since the beginning. The relation between bees and humans started as early as the stone age. It has been used in alternative medicine since that time, and its role was to treat burns, gastrointestinal disorders, asthma, infections and some chronic wounds. Honey maintains an important place in terms of nutrients as it is known to be rich in antioxidants, including glucose oxidase, catalase, ascorbic acid, phenolic compounds, carotenoids, organic acids, amino acids and proteins. The botanical origin of honey is one of its main quality parameters, and it has been reported that the composition and antioxidant capacity of honey depend on the floral source used to collect nectar, seasonal and environmental factors, as well as processing (Kıvrak and Kıvrak, 2017). These factors may also have an effect on the honey composition and antioxidant activity. Price depends on the quality and is also related to the floral origin. Fermentation is a problem for honey. The liquid mixture contains water, fructose and acid, so yeast could develop when the water content reaches a certain level. The higher the water content, the greater the occurrence of fermentation and spoilage. The water content in called moisture content (MC) in the food industry. The amount of the free water is really a response to the development of yeast; and not moisture contant (MC). The amount of free water could be described as water activity (Aw). The Aw for honey ranges from 0.5~0.65. The official method for MC measurement is refractometric measurement. It is inexpensive and easy to use. However, it cannot be directly used for crystallized honey. Apart from conventional heating, different moisture reduction systems have been designed across the globe in which either discs, cones or small screen holes have been used for increasing surface area of honey to be dried and vacuum or heated air for removal of moisture from the system. Different trials have also been done by the beekeepers for reduction of moisture directly in a room from honey storage drums. Honey is thermally treated as a part of processing which is required for easy handling, to dissolve large sugar granules, to reduce moisture for increasing shelf life, to destroy spoilage microorganisms i.e. fermentation causing yeast. This also helps in delaying granulation in honey. But honey quality is significantly affected by storage time and heating that may cause deterioration. From the study of different quality parameters as discussed, diastase activity, HMF and colour are more affected by heat treatment and are functions of both time and temperature.

Considering the above facts experiment was designed with the following objectives;

(1) To identify the major honey bee foraging plants in Bangladesh

(2) To investigate some prominent physicochemical properties of various honey samples and

(3) To identify the qualitative characteristics of different honey samples

CHAPTER II

REVIEW OF LITERATURE

The activity of beekeeping provides a beneficial food and income source for rural households in developing countries. Beekeeping has been widely promoted in many countries as a major contributor to rural development. Products such as honey, beeswax, bee pollen, propolis, royal jelly, venom, queen bees and larvae are of socio-economic value. Honey is a sweet and viscous liquid which has sweetness due to the presence of monosaccharides such as fructose and glucose. Its composition and chemical properties make it suitable for long-term storage. However, honey may crystallize after a period of time and this sometimes affect colour and consumer's acceptability. The colour, flavour and aroma are the important quality characteristics of honey from consumer's point of view. Honey is generally evaluated by physico-chemical analysis of its properties. Several of these constituents are of great importance to the honey industry as they influence the storage quality, granulation, texture, flavour and the nutritional and medicinal quality of the honey. The quality of honey is important for both local and international markets to ensure competitive prices and for human health. Honey quality is an aspect generally disregarded by producers and processors especially in developing economies. The International Honey Commission (IHC) has proposed certain constituents as a measure of quality criteria for honey. These include moisture content, electrical conductivity, reducing sugars, amount of fructose and glucose, sucrose content, individual sugars, minerals, free acidity, diastase, HMF, invertase and proline. Proper understanding and standardisation of honey components and attributes that are most vulnerable during processing cannot be ignored.

The major constituents of honey are sugars, water, proteins, enzymes, acids and minerals, while the major causes of quality deterioration include heating at high temperatures, high moisture content, adulteration, poor packaging and poor storage conditions. However, heating not only eases the bottling process by reducing the viscosity of honey, but also reduces the water content in honey to prevent fermentation. It destroys the sugar crystal nuclei to retard granulation by dissolving them during processing. The colour also becomes uniform throughout the sample for the preference of consumers along with destruction of sugar tolerant osmophilic yeasts to prolong the shelf life of honey. The present review aims to discuss the importance of honey processing, different systems developed for moisture reduction of honey and the major quality parameters affected by the heat treatment. Moisture is one of the most important parameter of honey quality. The amount of water present in honey determines its stability against fermentation and granulation. Honey having high water content ferments easily with time. So, it is necessary to process the honey by subjecting it to thermal treatment to prevent fermentation by sugar tolerant yeasts (Fallico *et al.* 2004). Treatment in a closed system minimizes losses of volatile aroma during heating. Processing temperature had significant effect on the moisture content. Honey processed at 60 °C had higher moisture (17.98%) as compared to 17.06 and 16.40% at 70 and 80 °C, respectively while the processing time had non-significant effect on the moisture content of honey packaged in glass jars, plastic jars and poly pack pouches. The storage had significantly decreasing effect on the moisture content of honey packaged in plastic jars and poly pack pouches. The moisture content of honey decreased to 16.41 and 16.63% in plastic jars and poly pack pouches respectively from initial 18.10% after 12 months of storage.

Hossain *et al*. (2015) conducted a study on quality parameters of honey collected from mustard, litchi and sundarban (multiflora) flora of Bangladesh was undertaken to compare with European Union (EU) quality standard viz., moisture content, electric conductivity, specific rotation, hydroxymethylfurfural (HMF) content, invertase content, antibiotic content and sulfonamides content. Sundarban honey (multifloral) (17.7g/100g) and mustard honey (19.5g/100g) were found in standard moisture condition and the litchi honey was found above the limit of EU standard (20g/100g). All the honey samples were found in the EU standard limit in terms of electrical conductivity and specific rotation. The HMF content was found higher (200.5mg/kg) in sundarban sample and no HMF was observed in the litchi honey. Highest invertase activity (99 IU/kg) was found in the litchi honey where as mustard and sundarban honey showed very low invertase activity with 2.0 IU/kg and 0.0 IU/kg, respectively which was far below than EU standard (50 IU/kg). All the samples had shown negative reaction with tetrasensor and sulfasensor. So, it may be concluded that the honey from sundarban was highly overheated. On the other hand, the mustard and litchi honey were overheated and not heated, respectively. Sundarban and mustard honey had failed to reach the quality standard to the EU. But litchi honey was found to met the all the EU quality requirements except the moisture content (Hossain *et al.* 2015).

Singh and Singh (2018) stated that beekeeping has been widely promoted in many countries as a major contributor to rural development. Honey is a sweet and viscous liquid which has sweetness due to the presence of monosaccharides. The major constituents of honey are sugars, water, proteins, enzymes, acids and minerals, while the major causes of quality deterioration include heating at high temperatures, high moisture content, adulteration, poor packaging and poor storage conditions. Heating not only eases the processing of bottling by reducing the viscosity of honey, but also reduces the water content in honey to prevent fermentation and delays the granulation by destroying large sugar molecule. The paper discusses about the different honey moisture reduction systems designed by research workers as well as beekeepers at farm level and the different quality parameters affected by thermal treatment of honey (Singh and Singh, 2018).

Huidobro *et al*. (1995) found that diastase activity of honey and reported values in the range of 11.3–34.5 diastase number. Heating treatment at 55–65 °C did not have much effect on diastase number but was more affected by storage time. However, honey produced in warmer climates has been observed to have lower diastase activity. Any type of honey possesses several kinds of enzyme that play both nutritional and analytical role in the product. One of the most important honey enzymes is diastase that is capable to break down glycosidic linkages in oligo- and polysaccharides i.e; starch into simple sugars. It is very sensitive to heat. Therefore proper heating and storage is of utmost importance to retain the market value of honey. The activity of this enzyme decreases with the time of storage and that of heating. The starch-digesting enzymes of honey are used as indicators of honey quality because of their heat sensitivity.

Hasan (2013) found that heat treatment of honey at 55, 65, 75 °C for 5, 15, 20 and 25 min did not affect diastase and invertase activity but they were more affected by storage time. Diastase can also be inactivated in short time using microwave treatment but it becomes difficult to establish rate constant. The diastase activity is also more sensitive to prolongation of heating time than to increasing the temperature. After 3 h at 50 °C, the enzymatic activity is more drastically reduced (53.71%) than in cases of heating at higher temperature but shorter time ranges such as 48.29% for 0.5 h at 100 °C or 49.55% for 1 h heating at 80 °C. The honey bottled in amber glass recipients was pasteurized to 65 °C for 15 and 21 min and was tyndallized to 80 °C during 5 and 7 min. Honey samples heat treated for pasteurization and tyndallization during 15 and 21 min showed statistically significant differences in diastase activity, in comparison to untreated honey. Diastase activity loss occurred as temperature increases. The decrease in diastase activity was more significant at higher rates of temperature increase. On the contrary, during the isothermal heating (temperature remains constant) the diastase activity decreased for short time treatments (typically for 120 s) but increased when time was increased. The effect was closely related to all temperatures under study i.e. 60–100 °C with the exception of 100 °C treatments in which the diastase activity became null (Hasan 2013).

Sahinler and Gul (2005) observed that Hydroxymethyl furfural (HMF) content increased significantly both with storage time and heating. Hydroxymethyl furfural (HMF) is formed from fructose in the presence of acid. Honey is acid enough to facilitate this change. Its production is very slow in honey at normal temperatures during the handling process. The amount formed, however, increases with increasing heat treatment. Heating honey above 75 °C for a few minutes or storing honey at temperatures above 27 °C for several months increases HMF levels. A maximum content of less than 40 mg/kg is allowed in honey in the international market. Amounts exceeding this maximum limit are considered a main indicator of honey deterioration either through heating or long periods of storage. Higher values of HMF also points towards the possibility of honey adulteration by invert syrup. Moreover, there is always variation in the formation of HMF in different honey types which is mainly due to sugar composition and pH. Heating of honey having low pH results in more HMF formation. Impact of heating temperature and time on formation of HMF in honey when subjected to thermal treatment at 50, 80 and 100 °C for 0.5–5 h time.The way of heating and storage time (at 23 °C) in three types of Iraqi honey heated at 55, 65, 75 °C for 5, 15, 20 and 25 min and concluded that HMF content of honey was significantly affected by storage time and heat treatment. The HMF level increased regularly from 5 to 25 min. for both the honey samples at each temperature and the increase was higher at 70 and 80 °C indicating the temperature dependence on formation of HMF. The HMF content proportionally increase with the increase of heating time up to 60 min at 63 and 90 °C and the kinetic studies showed that the HMF formation in honey followed zero order kinetic model for the first 60 min of heating at 90 °C (Sahinler and Gul,2005).

Chakraborti and Bhattacharya (2014) investigated 21 honey samples collected from different places of India. It was found that 16 samples contain higher values of HMF in respect to international standard limit (80 mg/kg) during storage and concluded that the high HMF content in most of the honey samples was might be due to their exposure in high heat stress during processing and storage. Eucalyptus lanceolatus, Brassica juncea and Trifolium honey types by heating them at 65, 85 and 95 °C for 5, 15 and 30 min. They found significant effect of temperature and time on HMF formation in Trifolium and Eucalyptus lanceolatus honey whereas heating time was more significant in *Brassica juncea* honey. It was concluded that second order polynomials can be used as a tool to study the effect of processing temperature and period on HMF formation in different honey types. In case of microwave heating, the increase in power levels has more significant effect on the formation of HMF in different types of honey as compared to duration of heating. Hydroxymethylfurfural (HMF) in different types of Latakia honey using microwave as well as conventional methods at 50, 80, 100 °C for 1, 2 and 3 h intervals. Heating of honey at 80 °C, during 60 and 30 s in transient and isothermal stages, respectively, destroyed all microorganisms responsible for quality damage without spoiling honey (Chakraborti and Bhattacharya, 2014).

Bogdanov *et al*. (2008) stated that due to the variation of botanical origin honey differs in appearance, sensory perception and composition. The main nutritional and health relevant components are carbohydrates, mainly fructose and glucose but also about 25 different oligosaccharides. Although honey is a high carbohydrate food, its glycemic index varies within a wide range from 32 to 85, depending on the botanical source. It contains small amounts of proteins, enzymes, amino acids, minerals, trace elements, vitamins, aroma compoundsand polyphenols. The review covers the composition, the nutritional contribution of its components, its physiological and nutritional effects. It shows that honey has a variety of positive nutritional and health effects, if consumed at higher doses of 50 to 80 g per intake (Bogdanov *et al*.2008).

Molan (2001) found that the effectiveness of honey as a therapeutic agent has been unequivocally demonstrated in the literature reviewed in Part 1 of this article published in 1999, but the biochemical explanation of these effects is more hypothetical. However, a rational explanation can be seen when one looks at the scientific literature outside that on honey. Some of the components of honey are substances known to have physiological actions that would explain many of its therapeutic effects. In addition, research on honey has shown directly that it has physiological actions that would give therapeutic effects (Molan, 2001).

Gul and Pehlivan (2018) carried out a study for determining the chemical, biochemical properties, and antimicrobial capabilities of some of the monofloral honeys produced in Turkey. In this study, 23 different monofloral honey samples were obtained from diverse geographical regions of Turkey. Floral origin of the honey samples was determined by melissopalinological analyses. Additionally, antioxidant properties were determined. To determine the antioxidant properties of honey samples, four test methods of total phenolic content, DPPH, iron reduction power and b-carotene linoleic acid emulsion method were used. As a result of the antioxidant activity analysis among the honey samples, rhododendron and parsley honey showed most prominent results in terms of the amount of phenolic compounds and antioxidant activity. On the other hand, *acacia* and *citrus* honey samples showed least antioxidant activity. A positive correlation was determined between four methods. Differences between antioxidant activities of honey samples were observed significant ($P < 0.01$) (Gul and Pehlivan, 2018).

Draiaia *et al*. (2014) investigated that some properties of various honeybee samples collected from different Algerian regions by using different honey analysis tests as refractive index, ash content, electrical conducting, protein content, acidity, pH and finally HMF content. These determinations indicate the quality of Thirty Two Algerian honey samples from *Apis mellifera* which is needed for international trade. Thirty two Honey samples were collected from beekeepers from the different regions of Algeria between 2012 and 2014 for the investigation of physicochemical composition. Each sample weighed 150 g and clearly identified by date of harvest and floral and geographical origin. Honey samples were analyzed for pH, moisture, total acidity, ash, electrical conductivity, total sugars, sucrose, hydroxymethylfurfural (HMF). All of these analyses were done following the European Honey Commission and International Honey Commission Methods. The study revealed the following results: Moisture content had a value of 13.00% (minimum) and 20.13% (maximum) indicating optimum harvesting and good degree of maturity. All honeys analysed in this work had water contents less than ranged 21%; pH 3.74 (minimum) et 5.55 (maximum); ashes 0.006 % (minimum) and 0.371 % (maximum) ; Electrical conductivities ranging between 0.097 mS / cm and 1.110 mS / cm); hydroxymethylfurfural 0.00 mg kg 1 (minimum) to 1380 mg kg 1(maximum); free acidity 6.87 (minimum) and 58.75 mg g-1 (maximum) and protein 0.09% et 0.81%. To summarize, the results of physic-chemical properties of honey from Algeria indicate good quality of 72% of honeys and that the remaining quality parameters agree in general, with international regulations (Draiaia *et al*.2014).

Qamer *et al*. (2013) found that nepalese honey samples stored for 8 months showed pH in the range of 3.9-4.6, free acidity 48.5-53meq/kg, lactone 15.5-17.1meq/kg, total acidity 61-70meq/kg, electrical conductivity 0.24-0.64mS/cm, Proline content 148-241mg/kg, HMF content 53.4-122mg/kg, Diastase Number 1.02-13.25DN and Invertase Number 0.58-10.5IN. After 16 months of storage the various parameters recorded were: pH 3.7-5.08, free acidity 46.1-57.07meq/kg, lactone 17-19meq/kg, total acidity 64-74meq/kg, electrical conductivity 0.29-0.71mS/cm, Proline content 66.43-120mg/kg. HMF content were beyond international maximum limit even after 8 months storage. Similarly diastase Number (0.22-0.86DDN) and Invertase Number (00-0.71IN) were much below than the minimum standard. Although honey produced by *A. dorsata* from Nepalese forest showed various quality parameters close to International Honey Quality Standards, yet its shelf life was shorter due to high moisture content (Qamer *et al*.2013).

Chua *et al.* (2013) carried out a study on antioxidant activities based on the free radical scavenging, reducing power, and bleaching inhibition were investigated for the three commonly used honeys in Malaysia, namely, tualang, gelam, and acacia honey.The antioxidant capacity of the honey samples was correlated with their biochemical constituents such as total phenol, total flavonoid content, and total water-soluble vitamins (vitamin B1, B2, B3, B9, B12, and vitamin C).The total flavonoid content of honey samples was strongly correlated with the three antioxidative processes (r = 0.9276–0.9910). In contrast, the total water-soluble vitamins was found to be well correlated with the free radical scavenging activity (r = 0.8226). Vitamin B3 was likely to be in the highest concentration, which covered for 69–80% of the total vitamin content. A number of five phenolic acids, three flavonoids, and two organic acids had also been detected from the honey samples using UPLC-MS/MS, without sugar-removal procedure (Chua *et al.* 2013).

Gulfraz *et al*. (2010) conducted a study on forty samples of different honey types (*Acacia, Ziziphus, Brassica* and *Citrus*) were collected from different areas of Pakistan and analyzed for moisture, pH, total acidity, ash, electrical conductivity, hydroxymethylfurfural (HMF), sucrose, total sugars, invert sugar, protein, proline contents as well as macro and micro elements. The variation in composition of honey samples was observed due to different types of flora. Higher pH (6.56 ± 0.05) was observed for *Ziziphus* honey, acidity (45.0 ± 2.35 mg/kg) for *Citrus*, moisture (36.8 ± 1.8%) for *Brasica* and HMF (32.7 ± 0.49 mg/kg) for Acacia. Whereas, higher concentrations of proline (2.1 ± 0.04 mg/kg) and invert sugar (0.38 ± 0.1%) for *Citrus* honey and protein (16.5 ± 1.5 g/100g) for *Acacia* honey were observed. Likewise, a significant level ($P < 0.05$) of ash, electric conductivity, sucrose, total sugar as well as macro and micro elements was also found in these honey types. Different formulations of honey has significantly inhibited growth of pathogenic microorganisms, *Staphylococcus aureus, Escherichia coli,Candida albicans* and *Aspergillus niger* when compared to control group, which is an evidence that honey is a therapeutic agent being used since ancient time throughout the world (Gulfraz *et al*.2010).

Zerrouk *et al*. (2011) carried out a study on the quality of sixteen samples of *Apis mellifera* L. honey, from the center of Algeria and evaluated by determining the physico-chemical characteristics. The studies were carried out to determine water content, total sugar, electrical conductivity, ash, pH, acidity (free, lactone, and total), hydroxylmethylfurfural (HMF) and color. The physicochemical parameters found are within acceptable ranges: water 13.36–17.93%, total sugar 80.17-84.73%, pH 3.58–4.72, total acidity 17.97-49.1 meq/kg, electrical conductivity 2.75×10-4 –7.19×10-4 S/cm, ash 0.075–0.33%, and color 4.1–9.2 Pfund index. The analysis of HMF showed that the majority of samples were exposed to a high temperature during processing or storage (Zerrouk *et al*.2011).

Azonwade *et al.* (2018) found in their study that honey is a very complex biological product. It has great diversity, giving it a multitude of properties, both nutritionally and therapeutically. This study aimed to find the physicochemical and microbiological characteristics of honeys collected during the dry and rainy seasons in the different phytogeographical areas of Benin. The study revealed that all honeys had pH, water content, electrical conductivity, ash content, free acidity, total sugars, and reducing sugars, ranging within 3.65–4.09; 12.07–13.16%; 530.25–698.50 μs/cm; 0.42–0.53%; 35.67–40.52meq/kg; 60–70%; and 58–70% respectively. Moisture content, total sugars, and reducing sugars varied significantly ($p< 0.05$ to $p< 0.001$) from one area to another and from one season to another. However, only the production season has a significant influence ($p< 0.05$) on the pH of the honey. With regard to the ash content, free acid, and electrical conduction, no significant difference ($p> 0.05$) between the zones or between the seasons was observed. The results of the microbiological characterization showed that there is heterogeneity in the microbial load. These results have shown that these honeys meet international standards and their characterization will make it possible to obtain Beninese quality labels (Azonwade *et al*.2018).

Aloisi (2010) evaluated that the quality chemical parameters of 62 *Apis mellifera L.* honey samples, from the Province of Chubut, Argentina. Samples were obtained from the three melliferous areas of this province: Andean region, lower Chubut River valley and plains of Senguerr River. The average values obtained for electrical conductivity (0.33 mS cm-1) and pH (4.17) indicate that the analyzed honeys came mainly from nectar. Electrical conductivity was higher in honeys from the Andean region than in honeys from the other two regions. Moisture water content) was low, with an average value of 14.67%. The hydroxymethylfurfural (HMF) content was very low and ranged between 0.0 and 14.70 mg kg-1. Moisture, HMF and free acidity values show good maturity and absence of undesirable fermentation in all the samples. Diastase activity had a mean value of 13.50 units on the Gothe scale. Color parameter presented variations between honeys from different areas. The analytical values for the samples from the Andean region, the lower Chubut River valley and the plains of the Senguerr River were: 67.73, 40.33 and 23.26 mm Pfund, respectively. Results obtained in this study, indicate that honeys produced in Chubut present excellent quality properties according to international standards.

Anthony and Balasuriya (2016) detected that adulteration of honey is a considerable challenge in the Sri Lankan context. The usual practice is to independently check the different parameters in order to determine the quality of a given honey sample. However, measuring and employing a single parameter for the classification reduces the accuracy of the classification. Thus, in this paper a multi-parameter based honey quality classification is proposed to ensure a better accuracy. The design of a parameter detector and a classifier which can automatically complete the classification of a given sample. This classifier operating on support vector machines is first trained using an array of honey samples obtained in Sri Lanka. The resultant classifier shows a high level of accuracy of 97.5% for the randomly selected test sample set. The proposed system is a handy tool for accurate, quick, low cost and simple honey quality checking.

Pires *et al*. (2015) evaluated that the quality of honey from *Apis mellifera* L. obtained in Piauí, Brazil. The completely randomized design (CRD) was used in the experiments. Two treatments of honey were prepared: one from beekeepers that use Extraction Units for Bee Products (EUBP) with Best practices for beekeeping (T1), and another one from those which use EUBP without the best practices (T2). Parameters analyzed were: moisture, water activity (aw), pH, acidity, color, detection of *Salmonella spp.*,N MPN.g-1 of coliforms at 35°C and at 45°C, counting of coagulase-positive Staphylococcus, standard counting of mesophilic heterotrophic bacteria and detection of yeast and filamentous fungi. The counting of mesophilic heterotrophic bacteria and yeast and filamentous fungi showed abnormalities ($p<0.05$) in the counting performed in log10.g-1 with samples of T1 and T2, respectively. There were presence of fungi of various genus and species, especially *Aspergillus spp*.and *Penicillium spp*. The quality of honey from

Apis mellifera bees from Piauí, Brazil, was satisfactory regarding parameters of moisture, AW, pH and HMF. *Neither Salmonella spp*.nor coliforms, nor coagulase-positive *Staphylococcus* were found. The presence of filamentous fungi in the samples reinforces the need for quality control of honey from Piauí, Brazil.

Chen (2019) stated that water activity (Aw) and moisture content (MC) data of floral honey at five temperatures were determined using the Aw method and it was found that temperature significantly affected the Aw/MC data. The linear equation could be used to express the relationship between Aw and MC of honeys. The empirical regression equations between parameters and temperature were established. To evaluate the factors affecting the Aw/MC data, we used categorical tests of regression analysis to assess the effect of the correlation between Awand MC of honey and examined the factors affecting the regression parameters. Six datasets from five countries were selected from the literature. The significance of the levels of qualitative categories was tested by t-test. The slope of the relationship between Aw and MC was affected by the state of honey (liquid and crystallized). The intercepts were significantly affected by honey type (flower or honeydew), harvesting year, geographical collection site, botanical source and other factors. The outliers in the datasets significantly affected the results. With modern regression analysis, useful information on the correlation between Aw and MC could be found. The results indicated that no universal linear equation for Aw and MC could be used. The Aw value could be used as a criterion for the honey industry; then, the MC of honey could be calculated by the specific linear equation between Aw and MC.

Pridal and Vorlova (2002) showed the close relations exist between the results of microscopic analysis, optical rotation and electrical conductivity in 55 honey samples from the Czech Republic.the obtained results were to be interpreted in relation to the classification of honey samples according to their origin (honey groups: blossom, honeydew and compound honey). The relations were positive, very close ($r > 0.80$) and very highly significant ($P < 0.001$). Several analysed samples would have been inserted into honey groups wrongly if only conductivity had been measured. The fact and high correlation coefficients revealed that exact classification of honey must be carried out not only by measuring the conductivity but also on the basis of optical rotation and microscopic analysis – namely in transition intervals of conductivity between the particular honey groups.

Bogdanov *et al*. (1999) exclaimed that International honey standards are specified in a European Honey Directive and in the Codex Alimentarius Standard for Honey, both of which are presently under revision. In this article, present knowledge on the different quality criteria is reviewed. The standard drafts include standards and methods for the determination of the following quality factors: moisture, ash, acidity, HMF, apparent reducing sugars, apparent sucrose, diastase activity and water-insoluble matter. International honey standards for fructose/glucose content, the sucrose content and electrical conductivity are proposed. Also the use of other quality factors, such as invertase activity, proline and specific rotation, used in many countries, is also discussed.

Achouri *et al.* (2015) find out a physicochemical analyzes of honey such as pH, acidity, electrical conductivity, and humidity of 46 samples of honey of *Ziziphus sp* and 24 type of *Acacia sp* consumed in the United Arab Emirates (UAE). Results: The physicochemical analysis confirmed the similar pH values (*Ziziphus sp* = 4.18 ± 1.58, 4.68 ± *Acacia sp* = 1.21), same thing for the conductivity (*Ziziphus sp* = 1.09 ± 0.3 mS / cm *Acacia sp* = 1.09 ± 0.16 mS / cm), and humidity (*Ziziphus sp* = 17.35% (17.35 g / 100 g) ± 1.5, *Acacia sp* = 15.92% ± 1.14). In addition, honey of *Ziziphus sp* is less acidic with a low tendency to fermenting (8.6 ± 3.5 meq / Kg) than *Acacia*honey (36.7±18.9 meq / kg). Conclusions: The physicochemical parameters of honey of *Ziziphus sp* (Sider), and *Acacia sp* (Samar) are complies with the national standards (UAE.S GSO 147: 2008) in the UAE and international standards (Codex Alimentarius 2001). Honey of *Ziziphus sp* is relatively better than *Acacia sp.*

Tucak *et al*. (2004) find out a result on agricultural producers apply numerous technological procedures, and enlarging efforts to produce the high-quality products. This initiative is present in the beekeeping, too. The quality of the honey produced by the honey bee colonies depends of various factors, but prevailing are the ecological conditions and the floristic composition of the honeyfull plants. The aim of our research was to discover the influence of the beehive type on the quality of honey, which is produced at apiaries under the similar environmental conditions. The whole studied honey bee colonies belong to the European race, *Apis mellifera carnica,* and they used the same honeyfull plants pastures. The results indicate that different beehive type used at apiaries influenced on the quality of honey.

Obiegbuna *et al.* (2017) conducted an experiment on honey sold in Awka market is supplied from within and outside Anambra State, Nigeria. The quality from identified sellers in the market was evaluated and compared with honey from two floral regions in the state and some reported international standards. Analyses were carried out on the proximate composition, some mineral elements, physical properties, microbial counts and inhibition activities, and organoleptic qualities using standard methods. The parameter values of the market samples and samples from apiarists in the floral regions were found to be similar. The moisture content of the samples ranged between 8.42 and 10.52 g/ 100 g; protein, 0.70 and 1.27 g/ 100 g; ash, 0.40 and 0.60 g/100 g; fat, 0.14 and 0.20 g/ 100 g; and carbohydrate 87.80 and 89.19 g/ 100 g. In descending order, elemental mineral values of K, Ca, Na, Mg and Fe ranged from 47.77 to 54.86 mg/ 100 g, 4.21 – 6.04 mg/ 100 g, 3.82 – 4.28 mg/ 100 g, 2.11 – 3.40 mg/ 100 g and 0.54 – 1.09 mg/ 100 g, respectively. Hydroxymethyl furfural (HMF) values of 13.62 and 10.28 g/ 100g were observed for floral regions of Adazi-Enu and Ikenga, respectively, but values of 23.26, 24.35 and 45.48 g/ 100 g for market samples 1, 2 and pharmshop, respectively. Market honey samples inhibition activity against P. *aeroginosa* was 4 cm as against 1 cm for floral region samples. The honey samples had inhibition activity against E. coli except one market sample with activity of 2.6 cm. Adazi-Enu floral region sample exhibited slightly above double the inhibition activity of 7.6 cm against *S. aureus* than the market samples (3.4 – 3.8 cm). Organoleptic qualities of the floral region samples were comparable to the market samples except the Pharmshop sample that was less acceptable. Except for HMF of pharmshop sample that exceeded international standard, parameters in all honey samples are within the standard and comparable indicating non adulteration of the samples.

Dele (2017) stated that honey samples were obtained from wild and domesticated sources and analyzed for some physicochemical properties such as color, pH, moisture content, ash content, refractive index, specific gravity, total solid, viscosity, glucose and fructose content following Standard Association of Official Analytical Chemistry. The following range of values for pH (3.55-4.20), moisture content (18.50-25.60%), soluble solids (74.10-81.20%), ash content (0.08-0.14%), specific gravity (1.38-1.47), refractive index (81.3-83.4%), fructose content (40.5-63.04%) and glucose content (19.35-32.34%). The mineral composition analyzed revealed potassium to be the dominant mineral in the honey samples followed by Calcium. However, Cadmium and lead where not detected in the honey samples. The results indicated that parameters such as pH, moisture content, ash content, specific gravity, sugar (majorly fructose and glucose content), fructose/glucose ratio, glucose/water ratio conform within the limit of the international standard for honey. However, moisture contents of the wild honey samples (22.05% and 25.60%) were a little higher than the Codex Standards of $\leq$ 21%. In conclusion, the honey samples investigated have the needed quality criteria and are good for human consumption. The results also revealed excellent organoleptic acceptability of the honey samples, hence are suitable for human uses.

Pavlova (2019) exclaimed that honey is a sweet natural product, which is produced by bees generally from the nectar of flowers and sweet deposits from plants. It is a complex mixture that contains nutrients and bioactive compounds such as carbohydrates (primarily fructose and glucose), enzymes, proteins, amino acids, organic acids, minerals, vitamins, aromatic substances, polyphenols, pigments, beeswax, and pollen that contribute to its color, smell and flavor. The composition and quality of honey is variable and it depends mainly on the botanical source of nectar from which it is obtained, but also depend on the geographic location, seasonal and climatic conditions, processing type and storage. Due to its special composition, honey is a functional food, which is consumed for its effects on human health, with antibacterial, antioxidant, anti-inflammatory and antimicrobial properties, as well as wound and sunburn healing effects. Honey is used in pure form after little or minimal processing as liquid, crystals or other types. The uses of honey as food include flavourant and sweetener in honey cookies, dairy products and fruit juices, as well as industrial production of beverages by mixing with alcohol. In this review, the physical properties and nutritive chemical composition thoroughly reviewed to underscore the quality of honey.

Rahman *et al.* (2017) found that natural honey is one of the most valued items in the market due to its unique properties and diversified usages. To ascertain the biochemical properties of commercially available honey products in Bangladesh, an experiment was conducted at Entomology and Horticultural laboratory in Bangabandhu Sheikh Mujibur Rahman Agricultural University (BSMRAU). Honey samples of eight different commercial brands were collected and subjected to analyze analyzed on different bio-chemical parameters. Significant differences in nutritional qualities were observed. Highest TSS (total soluble solids) and total reducing sugar were observed in the product Premium honey brand of Australia, however moisture content, protein and ash percentage were found the lowest. Other foreign honey brands provided better results compare to some local originated honey products based on particular parameters especially for moisture and electrical conductivity. The results of this study suggested that although locally originated commercial honey products such as BSCIC honey, Litchi and Mustard honey from BSMRAU have meet some international quality standard level however other parameters like moisture contents and EC needs to be improved.

Ghorbani *et al.* (2017) conducted an experiment on evaluate some chemical quality characteristics of honey produced in Iran. Totally, natural (n=80) and commercial (n=20) honey samples were randomly collected from North-West and South-West regions of Iran. Hydroxymethylfurfural (HMF) levels, phenolic contents, antiradical activity, and antioxidative potency of the samples were analyzed. Data were statistically analyzed using SPSS version 16. Results: The HMF level in 4 out of 80 (5%) natural honey samples and 14 out of 20 (70%) of commercial honey samples was higher than the recommended safety limit set by the Iran national standard. The average phenolic content was significantly ($p<0.05$) lower for commercial honeys (20.51 mg of Gallic Acid Equivalents (GAE)/100 g of honey) than for natural honeys (55.37 mg of GAE/100 g of honey). The mean IC50 of natural honey samples was 40.63 mg/ml with a range of 14.2-84.1 mg/ml; however, the mean IC50 of commercial honey samples was 341.96 mg/ml, with a range of 115-997.4 mg/ml. The difference between mean values of β-carotene bleaching inhibition of natural honey (54.54 mg/ml) and commercial ones (25.37 mg/ml) was found to be statistically significant ($p<0.05$). Also, there was a significant ($p<0.05$) difference between chemical quality characteristics of honeys between two geographical regions. Conclusion : The HMF levels, phenolic contents, antiradical activity, and antioxidative potency of Iranian natural honeys were remarkably more acceptable and suitable than commercial honeys produced in the country.

Kruzik *et al.* (2017) carried out an experiment on honey quality is a topical and significant problem of the food industry, bee keepers and consumers. In this work, 22 samples of commercially available honey aromas (with methyl and ethyl esters of phenylacetic acid predominated), 13 samples of authentic honey collected directly from bee keepers (characterised by high content of benzaldehyde, 2-phenylethanol, hotrienol and 2-phenylacetaldehyde) and 63 honeys purchased from an outdoor market were evaluated based on volatiles profiles determined through solid-phase microextraction coupled with gas chromatography-mass spectrometry (SPME-GC/MS) and then suspicious samples were identified. The results were statistically processed and compared with results of a sensory analysis. Six honeys, which differed significantly in volatiles profiles (outliers detected by Factor Analysis), selected volatile substance representation (furan-2-carbaldehyde, 1,4-dimethylpyrazole, benzaldehyde, 2-phenylacetaldehyde) and honey aroma intensity and pleasantness were subjected to targeted analyses (i.e. determination of 5-(hydroxymethyl)-2-furaldehyde, diastase activity, unauthorized additive presence). Four of these suspicious samples were found to have high content of 5-(hydroxymethyl)-2-furaldehyde (more than 40 mg/kg), three honeys had low values for diastase activity (less than 8) and three samples positive for triacetin addition. The fact that all these samples revealed a breach of least one of the selected quality parameters defined by the Codex Alimentarius standard proved the proposed methodology to be a useful tool for fast quality evaluation of honey.

Kugonza and Nabakabya (2008) find out an experiment that affected honey quality in Uganda were surveyed in 120 beekeeping households. Honey was sampled from supermarkets, hawkers and stall markets along four transects across Kampala, the capital. Honey quality parameters assessed were diastase number (DN), free acid (FA), moisture content (MC), hydroxymethylfurfural (HMF), and water insoluble solids (WIS). Honey was mostly harvested from basket and grass hives. Pressing, boiling and straining were popular honey processing methods. Honey quality was mainly compromised by harvesting immature honey, bad extraction methods and contamination by extraneous materials. Constraints to beekeeping were lack of appropriate equipment (52%), inadequate farmer skills, bad weather and vermin. Honey brands differed ($P < 0.05$) in DN, most failed the Uganda and Codex Alimentarius standards, and 20% met European Union HMF and DN standards. Correlation was observed between HMF vs. DN ($r = 0.94$); MC vs. FA ($r = 0.56$). Supermarket honey (4.65) was more superior ($P < 0.05$) in DN than stall markets (1.93), and hawkers (2.3). Similarly, WIS levels differed ($P < 0.05$) between honeys from supermarkets (0.08), stall markets (3.0) and hawkers (3.15). All honeys met MC standards, while DN and WIS were major shortcomings. Farmer training and extension in proper honey harvesting, handling and processing should be strengthened. Quality monitoring at all levels should be emphasized.

Adekanmbi *et al*. (2019) conducted an experiment on Pollen analytical study of two locally produced honey samples collected from Akwa Ibom and Cross River State, Southern Nigeria were carried out in order to ascertain the preferentially foraged plants by honey bees, as well as the concentration of heavy metals. Samples were subjected to standard laboratory procedures using an acetolysis mixture (pollen analysis) and atomic absorption spectrometry (heavy metals). The results from the analysis revealed 32 taxa belonging to 17 botanical families. The number of pollen grains in the honey samples varied significantly (between 4,887 - 12,755 pollen grains), indicating their richness in pollen grains. 12 elements - Ni, Cu, Zn, Se, Br, K, Ca, Ti, Cr, Mn, Rb, and Fe were detected. Potassium had the highest concentration, followed by iron, calcium, titanium, zinc, copper, chromium, nickel, among others. What is more, in this study, some important honey plants: Spondias mombin, Daniella oliveri, Manihot esculenta, Treculia africana, Syzigium guineensis, Diospyros mespiliformis, Parkia biglobosa, Terminalia superba, Senna hirsuta, among others were found to be predominant and have been identified to be characteristic of the vegetation typical of Southern Nigeria. With regard to honey, this study gives an indication of the geographical and botanical origins, as well as types, source, and degree of contamination, and also an overall measure of honey purity. Such information, when displayed, can help consumers make informed decision when purchasing honey and will also help beekeepers to avoid possible contamination. Moreover, it will assist regulatory agencies in taking proper measures for environmental and consumer protection, since the concentration of heavy metals in honey is influenced by environmental pollution.

Turkmen *et al*. (2006) carried out a study on the kinetics of changes in total antioxidant activity as assessed by DPPH radical and brown pigment formation (BPF) in honey heated at different temperatures (50, 60 and 70 °C) for up to 12 days were studied. Antioxidant activity and BPF increased with treatment temperature and time. BPF increased following zero-order kinetics with the activation energy value of 122 kJ/mol−1 at 50–70 °C. However, antioxidant activity variation showed different trends according to heating temperatures following second-order, first-order and zero-order kinetics at 50, 60 and 70 °C, respectively. Heating of honey at 70 °C was found to be more effective than 50 and 60 °C for both two parameters. The results demonstrated that antioxidant activity was correlated with increased browning of the samples.

Escuredo *et al.* (2013) experimented on one hundred eighty-seven honey samples from an Atlantic European area were studied to determine their nutritional compositions and antioxidant capacities, as well as the relationships between them. The results showed that heather, polyfloral, blackberry, and eucalyptus honeys had the highest carbohydrate contents, whereas honeydew and chestnut honeys had the lowest. There were some important differences among the honey types, which were related to the presence of minor components. The protein contents were significantly higher in honeydew and chestnut honeys, and the same results were obtained for mineral contents. Related to the presence of several antioxidant compounds, heather honey had the highest phenolic content, whereas honeydew and chestnut honeys had the highest flavonoid contents. Multivariate analysis showed that some variables, such as the amounts of flavonoids, minerals, proteins, and phenols, were significantly correlated with antioxidant activity. The regression analysis produced a significant model (R2 = 0.716; F = 154.680; P < 0.001) that related the antioxidant activity and the flavonoids, K, and P contents.

Cimpoiu *et al.* (2013) found that the physical and biochemical properties of some Romanian honeys in order to discriminate between their floral origins. The evaluated properties were total phenolic content, total protein content, total free amino acids content, color intensity (ABS450), pH, ash content, antioxidant activity. Twenty-six commercial honeys from six types of flowers (*acacia*, sunflower, forest, polyfloral, lime and Sea Buckthorn) were investigated. All samples showed considerable variations with reference to their properties. The properties values were in the range of approved limits (according to EU legislation). The total phenolic, total protein and total free amino acids contents and color intensity varied considerably. Similarly, forest honey had the highest antioxidant activity while the lowest was found in *acacia* honey. Correlation between the floral origin of honeys and the physical and biochemical properties, respectively, was observed. Moreover, this study demonstrates remarkable variation in DPPH scavenging activity and content of total phenols in honey, depending on its botanic source.

Ribeiro *et al.* (2014) stated that the effect of honey adulteration by high fructose corn syrup in different concentrations from 0% (pure honey) to 100% (pure high fructose corn syrup) was investigated using Low Field Nuclear Magnetic Resonance spectroscopy (LF 1H NMR) and physicochemical analytical methods. The LF 1H NMR data were analyzed by bi-exponential fitting and compared with physicochemical data. The physicochemical parameters demonstrated that water content, water activity, pH and color differed significantly in honey samples adulterated with different concentrations of high fructose corn syrup. These differences were also observed by transverse relaxation (T2). Bi-exponential fitting of T2 resulted in the observation of two water populations in all samples, T21 and T22, with relaxation times in the range of 1.26–1.60 ms and 3.33–7.38 ms, respectively. Relaxation times increased with higher percentages of high fructose syrup in adulterated honey. Linear correlations were observed between the T2, T21 and T22 parameters and physicochemical data, suggesting that LF 1H NMR can be used to discriminate pure blossom honey from honey adulterated with high fructose corn syrup.

Cervantes *et al.* (2000) found that bee honey is one of the main export produces of the *Yucatan Peninsula*. Nevertheless, its international trade becomes more and more difficult, because of the competition of other countries. Heating of honey could accelerate certain chemical reactions that lessen its quality during storage. The purpose of this study consists in assessing the effect of temporary heating the honey on the variation of its main quality characteristics during storage. For the study, Tahonal honey was used (*Viguiera dentatavar.helianthoides),* as well as Dzidzilché honey (*Gymnopodium* *antigonoides*, Blake). Samples of each honey were heated at 55°C during 3, 6, 9, and 15 minutes, after which they were left to cool at the room temperature (26±2°C); they were stored at that temperature and, during three months and a half, there were taken samples in order to be assessed. The changes in the diastase activity were observed, as well as in color, total acidity, and the HMF. The temporary thermic treatment to which the samples were submitted exerted no significant influence on the examined quality characteristics, except the diastase activity in the Dzidzilché honey, which has diminished. During the storage, the average monthly growth of HMF was higher in the *Tahonal* honey than the Dzidzilché honey, without the initial heating to influence the variation. The temporary heating during 9 and 15 minutes, respectively, has significantly influenced only the growing of the acidity during the storage of the *Tahonal* honey. Also, the *Tahonal* honey grew darker in color more rapidly than the Dzidzilché honey, without heating to influence this difference. Generally, there was initial thermic treatment has influenced changing certain quality parameters, and the behavior of both sorts of honey during storage was different, probably owing to their different composition.

Bang *et al.* (2004) found that Honey is an effective antiseptic wound dressing, mainly the result of the antibacterial activity of hydrogen peroxide that is produced in honey by the enzyme glucose oxidase. Because the rate of production of hydrogen peroxide is known to vary disproportionately when honey is diluted, and dilution of honey dressings will vary according to the amount of wound exudate, it is important to know more about the production of hydrogen peroxide at different concentrations of honey.The rates of hydrogen peroxide production by honey with respect to honey dilution were measured in eight different samples of honey from six different floral sources. Honey Research Unit, Waikato University, Hamilton, New Zealand. The maximum levels of accumulated hydrogen peroxide occurred in honey solutions diluted to concentrations between 30% and 50% (v/v) with at least 50% of the maximum levels occurring at 15-67% (v/v). This is equivalent to a 10 cm × 10 cm dressing containing 20 mL of honey becoming diluted with 10 to 113 mL of wound exudate. Maximum levels of hydrogen peroxide reached in the diluted honeys were in the range of 1-2 mmol/L. Significant antibacterial activity can be maintained easily when using honey as a wound dressing, even on a heavily exuding wound. Concentrations of hydrogen peroxide generated are very low in comparison to those typically applied to a wound, thus, cytotoxic damage by hydrogen peroxide is very low.

Brudzynski (2020) found that hydrogen peroxide plays a key role in honey antibacterial activity. The production of H_2O_2honey requires glucose oxidase (GOx) that oxidizes glucose to gluconolactone and reduces molecular oxygen to hydrogen peroxide. The content of GOx of honeybee origin was believed to be the main predictor of H_2O_2concentration in honey. The observed variations in H_2O_2 levels among honeys questioned however the direct GOx-H_2O_2relationship and left its absence opened for exploration. Here, we evaluated principal causes underlying the imbalance in the quantitative enzyme-product relationship with respect to: (a) enzyme and the product inactivation or destruction by honey compounds; (b) non-enzymatic pathway of H_2O_2 formation, and (c) a potential contribution of enzymes with GOx activity originating from nectars and microorganisms inhabiting honey. We also bring new facts on the relationship between honey colloidal structure and H_2O_2 production that change our traditional understanding of honey function as antimicrobial agent.

Bang *et al.* (2003) carried out a study that honey is an effective antiseptic wound dressing, mainly the result of the antibacterial activity of hydrogen peroxide that is produced in honey by the enzyme glucose oxidase. Because the rate of production of hydrogen peroxide is known to vary disproportionately when honey is diluted, and dilution of honey dressings will vary according to the amount of wound exudate, it is important to know more about the production of hydrogen peroxide at different concentrations of honey. The rates of hydrogen peroxide production by honey with respect to honey dilution were measured in eight different samples of honey from six different floral sources at Honey Research Unit, Waikato University, Hamilton, New Zealand. The maximum levels of accumulated hydrogen peroxide occurred in honey solutions diluted to concentrations between 30% and 50% (v/v) with at least 50% of the maximum levels occurring at 15-67% (v/v). This is equivalent to a 10 cm x 10 cm dressing containing 20 mL of honey becoming diluted with 10 to 113 mL of wound exudate. Maximum levels of hydrogen peroxide reached in the diluted honeys were in the range of 1-2 mmol/L. Significant antibacterial activity can be maintained easily when using honey as a wound dressing, even on a heavily exuding wound. Concentrations of hydrogen peroxide generated are very low in comparison to those typically applied to a wound, thus, cytotoxic damage by hydrogen peroxide is very low.

Willix *et al*. (1992) conducted an experiment on both honey and sugar were used with good effect as dressings for wounds and ulcers. The good control of infection is attributed to the high osmolarity, but honey can have additional antibacterial activity because of its content of hydrogen peroxide and unidentified substances from certain floral sources. Manuka honey is known to have a high level of the latter. Seven major wound-infecting species of bacteria were studied to compare their sensitivity to the non-peroxide antibacterial activity of manuka honey and to a honey in which the antibacterial activity was primarily due to hydrogen peroxide. Honeys with activity in the middle of the normal range were used. A comparison of the median response of the various species of bacteria showed no significant difference between the two types of activity overall, but marked differences between the two types of activity in the rank order of sensitivity of the seven bacterial species. The non-peroxide antibacterial activity of manuka honey at a honey concentration of 1.8% (v/v) completely inhibited the growth of *Staphylococcus aureus* during incubation for 8 h. The growth of all seven species was completely inhibited by both types of honey at concentrations below 11% (v/v).

Mandal and Mandal (2011) stated that medicinal importance of honey has been documented in the world's oldest medical literatures, and since the ancient times, it has been known to possess antimicrobial property as well as wound-healing activity. The healing property of honey is due to the fact that it offers antibacterial activity, maintains a moist wound condition, and its high viscosity helps to provide a protective barrier to prevent infection. Its immunomodulatory property is relevant to wound repair too. The antimicrobial activity in most honeys is due to the enzymatic production of hydrogen peroxide. However, another kind of honey, called non-peroxide honey (viz. manuka honey), displays significant antibacterial effects even when the hydrogen peroxide activity is blocked. Its mechanism may be related to the low pH level of honey and its high sugar content (high osmolarity) that is enough to hinder the growth of microbes. The medical grade honeys have potent in vitro bactericidal activity against antibiotic-resistant bacteria causing several life-threatening infections to humans. But, there is a large variation in the antimicrobial activity of some natural honeys, which is due to spatial and temporal variation in sources of nectar. Thus, identification and characterization of the active principle(s) may provide valuable information on the quality and possible therapeutic potential of honeys (against several health disorders of humans), and hence it discussed the medicinal property of honeys with emphasis on their antibacterial activities.

Shambaugh *et al.* (1990) found that dietary carbohydrate components influence the prevalence and severity of common degenerative diseases such as dental problems, diabetes, heart disease and obesity. Fructose and sucrose have been evaluated and compared to glucose using glucose tolerance tests, but few such comparisons have been performed for a "natural" sugar source such as honey. In this study, 33 upper trimester chiropractic students volunteered for oral glucose tolerance testing comparing sucrose, fructose and honey during successive weeks. A 75-gm carbohydrate load in 250 ml of water was ingested and blood sugar readings were taken at 0, 30, 60, 90, 120 and 240 minutes. Fructose showed minimal changes in blood sugar levels, consistent with other studies. Sucrose gave higher blood sugar readings than honey at every measurement, producing significantly (p less than .05) greater glucose intolerance. Honey provided the fewest subjective symptoms of discomfort. Given that honey has a gentler effect on blood sugar levels on a per gram basis, and tastes sweeter than sucrose so that fewer grams would be consumed, it would seem prudent to recommend honey over sucrose.

Kowalski (2013) found that the microwave irradiation and conventional heating of honey. These two kinds of thermal treatment result in the formation of 5-hydroxymethyl-2-furfural (HMF), and changes in the antioxidant potential of honeys, which were studied as well. Four types of honey (honeydew, lime, acacia, buckwheat) were analyzed. Honey samples were subjected to conventional heating in a water bath (WB) at 90 °C up to 60 min or to the action of a microwave field (MW) with constant power of 1.26 W/g of the sample up to 6 min. Changes in the antioxidant capacity of honeys were measured as a percentage of free radical (ABTS+and DPPH) scavenging ability. Changes in the total polyphenols content (TPC) (equivalents of gallic acid mg/100 g of honey) were also determined. Formation of HMF in honey treated with a microwave field was faster in comparison with the conventional process. Changes in the antioxidant properties of honey subjected to thermal or microwave processing might have been botanical origin dependent.

Molar (2012) stated that there has been a renaissance in recent times in the use of honey, an ancient and traditional wound dressing, for the treatment of wounds, burns, and skin ulcers. In the past decade there have been many reports of case studies, experiments using animal models, and randomized controlled clinical trials that provide a large body of very convincing evidence for its effectiveness, and biomedical research that explains how honey produces such good results. As a dressing on wounds, honey provides a moist healing environment, rapidly clears infection, deodorizes, and reduces inflammation, edema, and exudation. Also, it increases the rate of healing by stimulation of angiogenesis, granulation, and epithelialization, making skin grafting unnecessary and giving excellent cosmetic results.

Ozcan (2006) found that three groups of honey [natural honey; honey produced by the supplementary feeding of bees with saccharose syrup honey (SSH) and heat and acid (88 °C, 2 h; 0.1% HCl) treated saccharose syrup honey (ISSH)] were produced and physicochemical (water content, pH, free acidity, ash, HMF, diastase activity, sucrose, protein and viscosity), microbiological and sensory properties of these honeys were determined. Also, mineral contents of the honeys were measured. Moisture and ash contents of SSH were higher, acidity level was lower than those of other honeys. The mineral content of natural honey was higher than that of the others, except for Pb and Zn. Diastase activity of ISSH was below the standard limit and HMF content of this honey was high, but not exceeding the limit. Supplementary feeding of honey bees with inverted (acid and heat treatment) *saccharose* yielded a honey which had a higher HMF content and a lower diastase activity, moisture content and free acid than natural honey or SSH.

Almasaudi *et al.* (2017) carried out a study on honey exhibits antimicrobial activities against a wide range of bacteria in different milieu. This study aims to compare the effects of five types of honey (both imported and local Saudi honey) against *Staphylococcus aureus.* The five types of honey (Manuka Honey UMF +20, Manuka Honey UMF +16, Active +10 Manuka Honey, Sidr honey and *Nigella sativa* honey) were evaluated for their bactericidal/bacteriostatic activities against both methicillin resistant and sensitive *S. aureus*. The inhibitory effect of honey on bacterial growth was evident at concentrations of 20% and 10% (v/v). Manuka Honey showed the best results. Manuka Honey UMF +20 had a bactericidal effect on both methicillin resistant and sensitive S. aureus. However, Sidr and *N. sativa* honey exerted only a bacteriostatic effect. The efficacy of different types of honey against *S. aureus* was dependent on the type of honey and the concentration at which it was administered. Manuka Honey had the best bactericidal activity. Future experiments should be conducted to evaluate the effects of honey on bacterial resistance.

Kamal *et al.* (2002) stated that a total number of 40 honey samples were collected from *Apis melifera* colonies forged on the five flora i.e., *Ziziphus spp.Acacia modesta,Trifolium spp.,Citrus spp*. and*Eucalyptus spp.* These samples were analyzed for fifteen standard physico-chemical parameters of honey quality control i.e., free acid, lactone, total acidity, refractive index, specific gravity, reducing sugars, sucrose, total sugars, hydroxymethylfurfural (HMF) content, diastase value, ash contents, water insoluble solids and total soluble solids. The biochemical variation in the composition of honey due to floral type shows *Ziziphus* honey with high pH, ash and diastase value along with low acids and sucrose contents whereas *Trifolium* honey contained high moisture content, acids and sucrose along with low quantity of reducing sugars. Highest HMF was detected in *Acacia* honey along with lowest diastase and ash contents.

Sarker *et al*. (2015) determined that the heavy metal levels and the physicochemical parameters (pH), electrical conductivity (EC), and ash, moisture, and total sugar content of honeys from Bangladesh. Three different floral honeys were investigated, namely, khalsi (*Aegiceras corniculatum*), mustard (*Brassica juncea*), and litchi (*Litchi chinensis*) honeys. The heavy metals in the honeys were determined by using a High Temperature Dry Oxidation method followed by Atomic Absorption Spectroscopy. The mean pH, EC, and ash, moisture, and total sugar contents of the investigated honeys were 3.6, 0.51mS/cm, 0.18%, 18.83%, and 68.30%, respectively. Iron was the most abundant among all the investigated heavy metals, ranging from 13.51 to 15.44mg/kg.The mean concentrations of Mn and Zn in the investigated honeys were 0.28mg/kg and 2.99mg/kg, respectively. Cd was below the detection limit, and lead was found in some honey samples, but their contents were below the recommended MaximumAcceptable Level. Cr was also found in all of the samples, but its concentration was within the limit.The physiochemical analysis of the honey samples yielded levels within the limits set by the international honey legislation, indicating that the honey samples were of good quality and had acceptable values for maturity, purity, and freshness.

Erejuwa *et al.* (2012) stated that honey is a natural substance with many medicinal properties, including antibacterial, hepatoprotective, hypoglycemic, antioxidant and antihypertensive effects. It reduces hyperglycemia in diabetic rats and humans. However, the mechanism(s) of its hypoglycemic effect remain(s) unknown. Honey comprises many constituents, making it difficult to ascertain which component(s) contribute(s) to its hypoglycemic effect. Nevertheless, available evidence indicates that honey consists of predominantly fructose and glucose. The objective of this review is to summarize findings which indicate that fructose exerts a hypoglycemic effect. The data show that glucose and fructose exert a synergistic effect in the gastrointestinal tract and pancreas. This synergistic effect might enhance intestinal fructose absorption and/or stimulate insulin secretion. The results indicate that fructose enhances hepatic glucose uptake and glycogen synthesis and storage via activation of hepatic glucokinase and glycogen synthase, respectively. The data also demonstrate the beneficial effects of fructose on glycemic control, glucose- and appetite-regulating hormones, body weight, food intake, oxidation of carbohydrate and energy expenditure. In view of the similarities of these effects of fructose with those of honey, the evidence may support the role of fructose in honey in mediating the hypoglycemic effect of honey.

Aljohar *et al.* (2018) conducted an experiment that honey is becoming accepted as a reputable and effective therapeutic agent by practitioners of conventional medicine and by the general public. It has many biological activities and has been effectively used in the treatment of many diseases, e.g. gastrointestinal diseases, skin diseases, cancer, heart diseases, and neurological degeneration. Honey is an excellent source of energy containing mainly carbohydrates and water, as well as, small amounts of organic acids, vitamins, minerals, flavonoids, and enzymes. As a natural product with a relatively high price, honey has been for a long time a target for adulteration. The authenticity of honey is of great importance from commercial and health aspects. The study of the physical and chemical properties of honey has been increasingly applied as a certification process for the purpose of qualification of honey samples. The current work focusses on studying the authenticity of various types of honey sold in Riyadh market (24 samples). For this purpose, physical properties (pH, hydroxylmethylfurfural HMF, and pollen test) were measured. Besides, sugar composition was evaluated using Fehling test and an HPLC method. Elemental analysis was carried out using inductively coupled plasma (ICP). In addition, the presence of drug additives was assessed by means of GC–MS. The obtained results were compared with the Saudi Arabian standards, Codex Alimentarius Commission (2001), and harmonized methods of the international honey commission.

Krishnasree and Ukkuru (2017) found that each honey is unique on the basis of chemistry, quantity and combination of the various components that attributes towardsits quality. The control and characterization of quality are of great importance and interest in apiculture. Henceforth the present study analyzed the quality parameters of honeys from different bee origin and compared the results with the quality criteria laid by European Union Directive for honeys. Processing was noted to have detrimental effect on several quality parameters of honeys; in spite of which it was remarkably within the critical limits. Moisture and electrical conductivity were within the limits as specified by EU, while slight elevation was noticed in honeys viz., *Apis mellifera*and *Apis floreain* their ash content,sucrose content and acidity. Hydroxymethyl furfural content and diastase activity of the honeys were within the levels of EU directive. The microbial analysis indicated that none of the honeys analyzed were infected with pathogenic coliforms.

CHAPTER III

MATERIALS AND METHODS

The experiment was conducted at the Research laboratory of Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar, Dhaka, 23°41′N latitude and 90°22′E longitude with an elevation of 8.6 meter above sea level. The experiments were carried out during 2017-2019. The material used and methodology adopted for these experiments are described as follow:

Sampling Area

All of the honey Samples were collected from different areas of Bangladesh, i.e. Mustard from Sirajgonj, Litchi from Gazipur, Coriander from Shariotpur, Black kumin from Shariotpur, Jujube from Mymensingh, Olive from Gazipur, Khulshi from Sundarban, Shatkhira district and rest of the samples were collected randomly. (Plate:1)

Sampling

Honey samples were collected at the maturity phase of production from beekeepers. Thus, different honey samples were collected aseptically for this study. Samples were kept in sterile vials, hermetically sealed, labeled, dated, and stored at room temperature (25–30∘C) until analysis. The sample collection periods varied according to the zones and availability of respective flower. The quality analysis of honey was done in disease identification and analysis laboratory, under department of entomology in Sher-e-Bangla Agricultural University. (Plate: 2)

Optical Rotation: It indicates either honey is pure or adulterated. If honey shows negative optical rotation it indicates honey is real/pure and positive optical rotation honey is adulterate. But, honeydew honey and syrup rotates positive angle. In respect of, honeydew honey positive angle is real honey. Optical rotation was measured by using RePo-4. (Plate: 3,4)

Moisture: Moisture was determined by refractometry, using an atago (japan) model.All measurements were performed at 25°c. Besides honey refractometer, RePo-4 also was used to measure the moisture content of honey. (Plate: 3, 4)

Brix: Sugar content was determined with a special refractometer with direct reading display, and the results were expressed as °Brix. Percentage of sugar. It also means rest of the percentage is moisture. It was measured by using RePo-4.

Fructose: It indicates quality of honey. It was also measured by using RePo-4.

pH: It was measured by using pH strips. (Plate: 5)

Experimental Duration: The experiment was conducted during 1st January 2019 to 30 December 2019.

Materials required

i)Refractometer ii) pH Strip iii) RePo-4 iv) Handgloves v) Apron. vi)Bottle/jar vii) Marker

Collection of honey: By using honey extractor.

Data Collection

- Amount of honey/sample (g)
- Assessment of honey moisture, pH, optical rotation, brix, fructose.

Plan of Work (Table 1).

Table 1. Time frame

Area selection and visit	2 month
Sample collection	6 months
Sample processing and analyzing	2 months
Report writing	2 month
Total	12 months

Data analysis

Data were statistically analyzed using SPSS version 25.

Data was analyzed using MSTAT-C computer program. Wherever necessary, data was transformed following appropriate methods before ANOVA. Standard error calculated by MSTAT-C and plotting bars and boxes in R i386 3.4.0 (R Gui-32 bit).

LIST OF PLATES

Plate 1. Collected honey samples

Plate 2.pH strips for pH measurement

Plate 3. Data collection in laboratory

Plate 4.Honey sample analysis in laboratory

Plate 5. Working in laboratory

Plate 6. Working in laboratory

CHAPTER IV

RESULTS AND DISCUSSION

Major nectar producing plants and their blooming period

Some plants constitute the source of nectar for production of honey. Honeybees collect nectar and pollen as their food. For this reason they always visit plants around their hive. Some plants are selected only for pollen as food and flowers of these plants may not contain nectar. Some plants are selected both for nectar and pollen, and some only for their nectar. The honey plant of an area can be identified by analysis of the pollen present in honey or by direct observation of the foraging patterns of bees. Honey bees collect nectar from various plants available around 4 km of their hive. Here enlisted some flowering plants those are supplying maximum amount of honey (Table 2.)

Table 2. Major nectar producing plants and their blooming period

Name of the plants	Scientific Name	Blooming Period
Litchi	*Litchi chinensis*	March - April
Sundari, Khoilsa(Alwoan)	*Heritiera littoralis*	April - May
Mango	*Mangifera indica*	February - March
Jamrul	*Syzygium samarangense*	March - April
Lemon	*Citrus limon*	March - June
Rose Apple	*Syzygium jambo*	March - April
Jujube	*Ziziphus jujuba*	September - November
Olive	*Elaeocarpus tectorius*	July - August
Mustard	*Brassica nigra*	December - January
Black cumin	*Nigella sativa*	Mid-February - the end of February
Coriander	*Coriandrumsativum*	February
Sunflower	*Helianthus annuus*	February - March
Sesame	*Sesamum indicum*	June
Sweet Gouard	*Cucurbita pepo*	November - December
Drumstick tree	*Moringa oleifera*	October - February
(antana(KutuskataL	*Lantana camara*	March - September
Cosmos	*Cosmos atrosanguineus*	January
Rose	*Rosa species*	Year Round
Marigold	*Tagetes erecta*	November - December
Jackfruit	*Artocurpus heterophyllus*	November - December

Brix

The total soluble solids (TSS) of the honey analyzed had ranged from 69.9 to 82.77 brix. Among the honey samples studied the highest amount of brix (82.77) was found in mustard honey (sample no.15) and lowest amount of brix (69.9) in olive honey (sample no. 13) (Figure 1). Higher brix means, presence of less amount of moisture, Honey was harvested in ripe condition, better quality, honey can be stored for long time and less chance for fermentation. The higher water content consequently lowers sugar content.

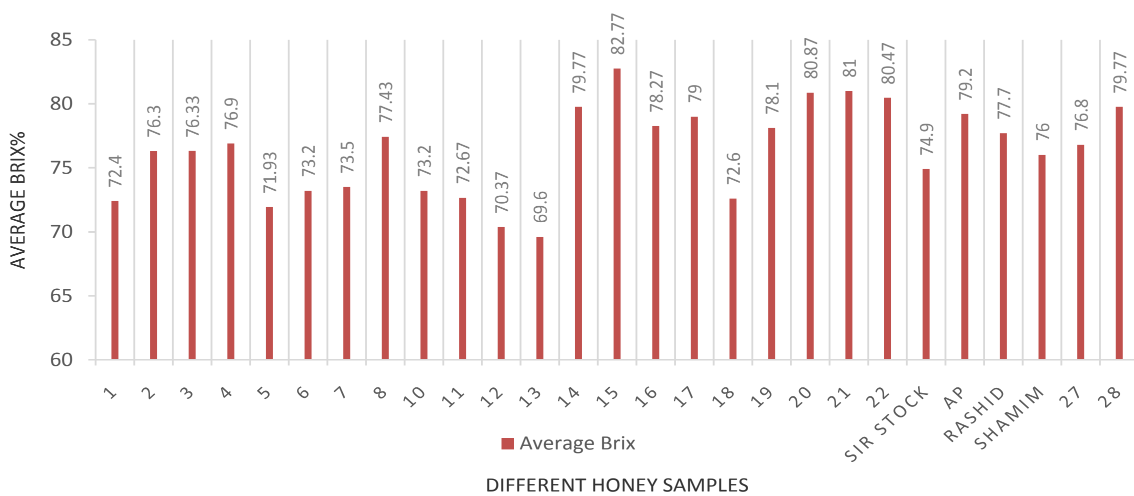

Figure 1. Average brix percentage in different honey samples of Bangladesh

1.Khoilsa 2.Unknown source (1) 3. Unknown source (2) 4. Kalojira (Black Cumin) 5. Sundarban 6. Khoilsa (Alwoan) 7. Jujube 8. Dhonia-kalojira 10. Litchi 11. Goran 12. Bakki 13. Olive 14. Balparmark 15. Mustard 16. Mustard(Old) 17. SR (Creamed Honey) 18. Fake Honey 19. Mustard (New) 20. Foreign Honey 21. Multifloral (Abroad) 22. Forest Honey 27. Pran Honey 28. Dabur

A one-way ANOVA is a type of statistical test that compares the variance in the group means within a sample while considering only one independent variable or factor. A one-way ANOVA compares three or more than three categorical groups to establish whether there is a difference between them. Brix significantly differed among all the other group with different sample means at 5% level of significance (Table 3).

Maximum honey samples were analyzed in this study had soluble sugars lesser than 80%. The findings in this study on soluble sugars are in agreement with their total soluble solids content. As there were lesser soluble sugars in the honeys analyzed their TSS also tends to be lower.

Table 3. One way ANOVA between groups with mean difference and standard error (SE)

LSD			
Groups	Groups	Mean Difference	SE
Brix	Moisture	54.27259*	3.84924
	Fructose	35.15370*	3.84924
	pH	71.96481*	3.84924

*. The mean difference is significant at the 0.05 level.

Specific Rotation

The highest specific rotation (- 5, deg cm2 g-1) was recorded in mustard honey. And the lowest specific rotation (-1.84, deg cm2 g-1) was in litchi honey (Table 4). Some honey samples showed zero (0) specific rotation, means those samples were over heated during processing or stored for long period of time under high temperature.

Table 4. Average specific rotation (deg cm2 g-1) in different honey samples of Bangladesh

Type of sample	Average specific rotation (deg cm2 g-1)
Khoilsa	-4.17
Unknown source (1)	0
Unknown source (2)	-3.18
Kalojira (Black Cumin)	0
Sundarban	0
Khoilsa(Alwoan)	-4.84
Jujube	-2.11
Dhonia-kalojira	-2.79
Litchi	-1.84
Goran	-2.88
Bakki	0
Olive	0
Balparmark	-2.34
Mustard	-5
Mustard(old)	0
SR(Creamed Honey)	0
Fake Honey	-2.32
Mustard(New)	-0.16
Foreign Honey	-3.9
Multifloral(Abroad)	-4.27
Forest Honey	-4.2
Sir Stock(old)	0
AP	-4.61
Rashid	-2.92
Shamim	-3.96
Pran Honey	-3.55
Dabur	-4.38

EU standard (deg cm2 g-1)
(+) indicate extra floral honey (-) indicate nectar honey

The specific optical rotation (SOR), is a physical property of honey and can be defined as the angle of rotation of polarized light at the wavelength of the sodium D line at 20ºC. The SOR analytical determination proposed by the International Honey Commission (Bogdanov, Martin, & Lullmann, 2002) might be affected by several factors. The optimization of this method on those affecting factors might improve its precision and accuracy and thus, its capability of discriminating between honeys. The aim of specific optical rotation was differentiating between blossom and honeydew honeys and their mixtures.

Maximum honey samples showed negative specific rotation. So, these honey samples were obtained from floral origin (nectar honey). If honey samples show positive specific rotation it will indicate extra floral honey (except blossom). Honey can have two different botanical origins, being classified as blossom honey or honeydew honey. Blossom or floral honey is produced by bees from nectar of flowers of blossoming plants. Honeydew honey is obtained from secretions produced by certain trees and other plants (genera pinus, abies, castanea, and quercus, among others) or excretions of plant-sucking insects, mainly from the family aphididae, on the living parts of plants (pita-calvo & vazquez, 2017).

Moisture Content

The highest moisture content (29 %) was obtained in olive honey (Sample no. 13.) and lowest moisture content (15.47 %) was found in mustard honey (Sample no. 15.) (Figure 2). So, moisture percentage among analyzed honeys ranged from 15.47 to 28.13. The samples contained more than 20% water, those are not allowed by international legislations. Moisture content significantly differed among all the other parameter with different sample at 5% level of significance (Table 5).

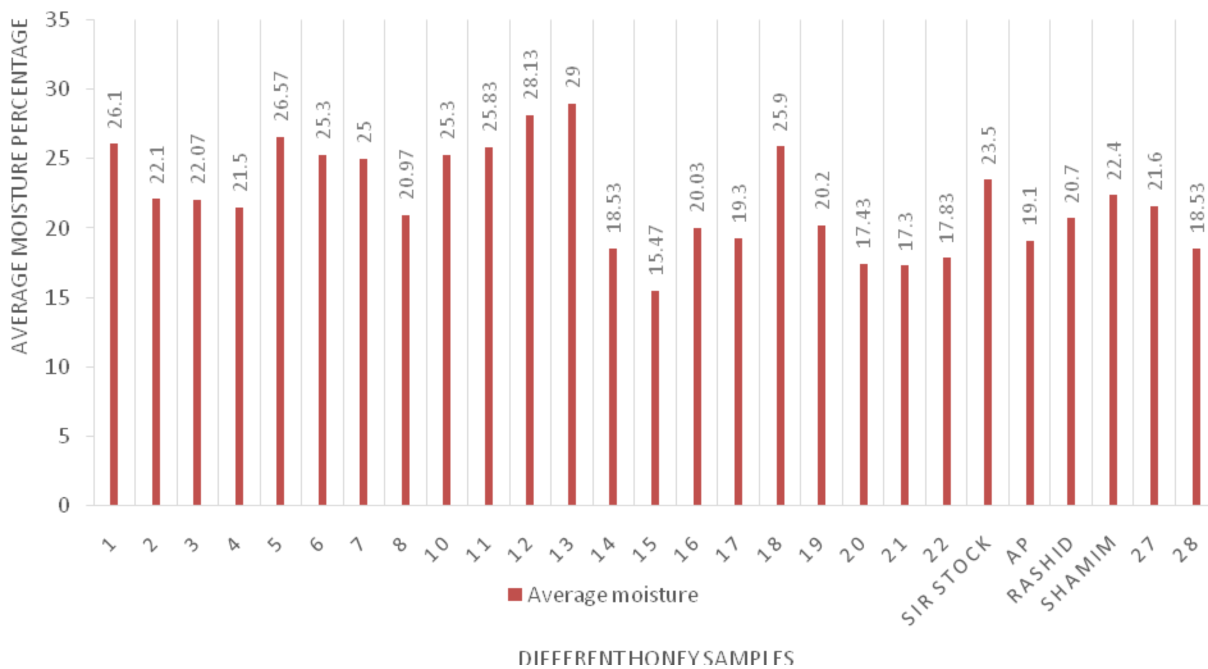

Figure 2. Average moisture percentage in different honey samples of Bangladesh

1. Khoilsa 2. Unknown source (1) 3. Unknown source (2) 4. Kalojira (Black Cumin) 5. Sundarban 6. Khoilsa (Alwoan) 7. Jujube 8. Dhonia-kalojira 10. Litchi 11. Goran 12. Bakki 13. Olive 14. Balparmark 15. Mustard 16. Mustard (Old) 17. SR (Creamed Honey) 18. Fake Honey 19. Mustard (New) 20. Foreign Honey 21. Multifloral (Abroad) 22. Forest Honey 27. Pran Honey 28. Dabur

EU standard (g/100 g)
Not more than 20

The water content of honey depends on various factors, like the harvesting season, the degree of maturity reached in the hive and climatic factors. The maximum amount of water contained in honey is regulated for safety against fermentation. In general, high moisture content occurred if honey is harvested in unripe condition and in that case moisture content is crossed over international legislations. To maintain the acceptable limit honey is processed either in processing plant or manually through steaming method. As some honey samples were steamed manually and processed in processing plant their moisture content was found low in moisture content remained below 20%. Some honey samples were collected as fresh condition and no processing were done (only filtered), and therefore, their moisture content was high. These higher moisture content samples would impare storage for long time, become fermented and also change its color, smell as well as the quality.

Table 5. One way ANOVA between groups with mean difference and standard error (SE)

LSD			
Groups	Groups	Mean Difference	SE
Moisture	Brix	-54.27259*	3.84924
	Fructose	-19.11889*	3.84924
	pH	17.69222*	3.84924
*. The mean difference is significant at the 0.05 level.			

Fructose

The highest amount of fructose (64.33 %) was recorded inKhoilsa (Alwoan) (Sample no. 6.) and the lowest amount of fructose (34.03 %) was in Mustardhoney (Sample no. 15.) (Figure 3). Higher the fructose indicates better quality of honey as well as those samples have less chance to form crystallization. On the other hand, lower the fructose indicates those samples have more chance to form crystallization. Some honey samples showed zero (0) means those samples were over heated during processing or stored for long period of time under high temperature. Fructose significantly differed among all the other parameter with different sample at 5% level of significance (Table 6).

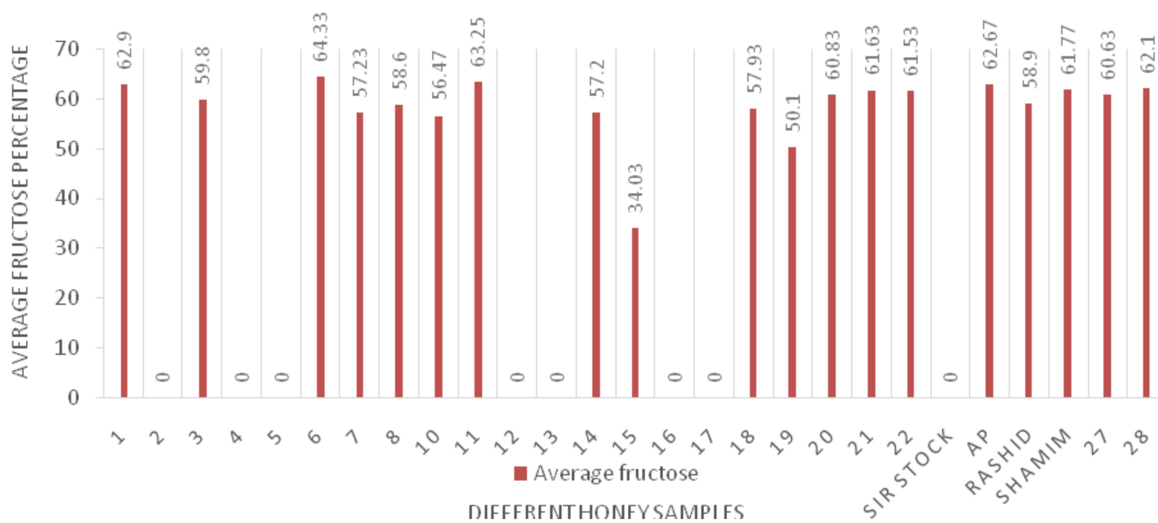

Figure 3. Average fructose percentage in different honey samples of Bangladesh

1. Khoilsa 2. Unknown source (1) 3. Unknown source (2) 4. Kalojira (Black Cumin) 5. Sundarban 6. Khoilsa (Alwoan) 7. Jujube 8. Dhonia-kalojira 10. Litchi 11. Goran 12. Bakki 13. Olive 14. Balparmark 15. Mustard 16. Mustard (Old) 17. SR (Creamed Honey) 18. Fake Honey 19. Mustard (New) 20. Foreign Honey 21. Multifloral (Abroad) 22. Forest Honey 27. Pran Honey 28. Dabur

Table 6. One way ANOVA between groups with mean difference and standard error (SE)

LSD			
Groups	Groups	Mean Difference (I-J)	SE
Fructose	Brix	-35.15370*	3.84924
	Moisture	19.11889*	3.84924
	pH	36.81111*	3.84924

*. The mean difference is significant at the 0.05 level.

Fructose is the sugar responsible for the sweetness of honey while glucose content depends upon the source of nectar.The problem of honey adulteration with syrups (e.g. maple syrup, sugar cane syrup) should be detected as recommended by the reference organization (Bogdanov, 2009). Determination of each of the glucose content, fructose content, sucrose content, and fructose to glucose ratio, are among the most important assessment used to distinguish syrup from honey. Another important problem in honey production is the crystallization of sugars in the comb cells. The crystallization problem could be detected by measuring glucose to fructose ratio since crystallization is accompanied with lower content of fructose and higher content of glucose (Rybak-Chmielewska, 2007).

pH Measurement

Honey is characteristically acidic. All honey samples were acidic with pH values ranging between 1 to 6 (Figure 4).The highest pH (6) was recorded in Mustard honey & Foreign honey (sample no.15 & 20) and lowest pH (1) in Fake honey (Sample no. 18). All tested samples had pH values within the accepted range (3.24–6.1) with the exception of Fake Honey &Forest Honey (Samples no.18 & 22) compared to range of international honey stated by The Codex Alimentarius Standard [(CAC, 2001)]. It has been reported that low pH inhibits the presence and growth of microorganisms, so all honey samples may have the potential to be used as good antibacterial agents.

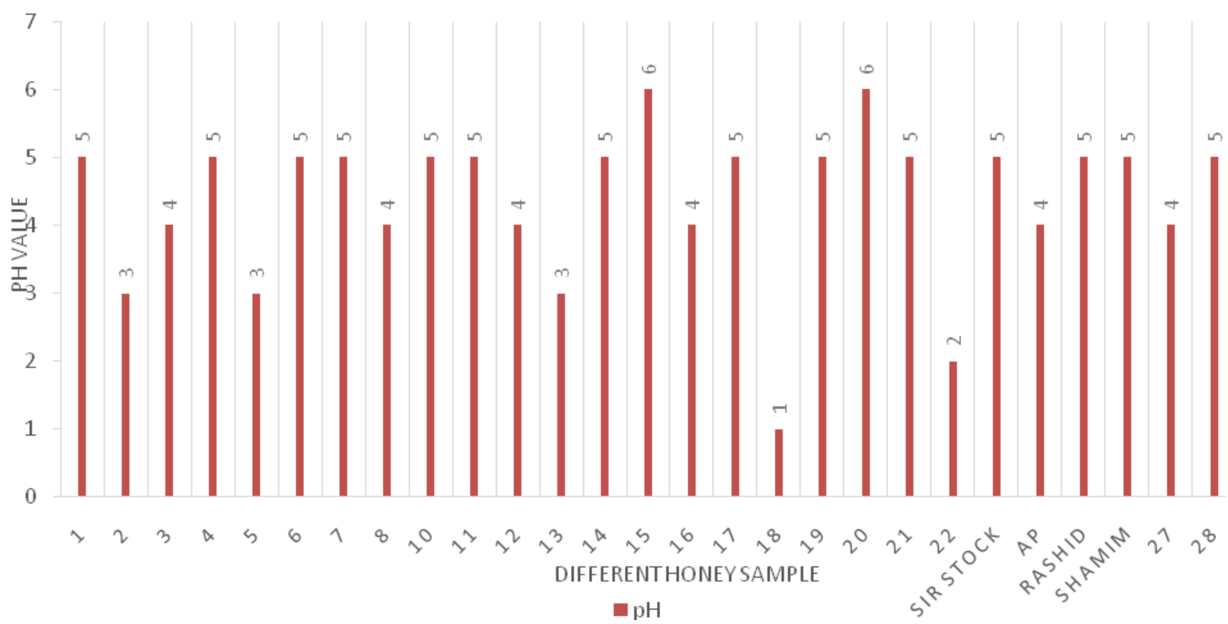

Figure 4. pH values in different honey samples of Bangladesh

1. Khoilsa 2. Unknown source (1) 3. Unknown source (2) 4. Kalojira (Black Cumin) 5. Sundarban 6. Khoilsa (Alwoan) 7. Jujube 8. Dhonia-kalojira 10. Litchi 11. Goran 12. Bakki 13. Olive 14. Balparmark 15. Mustard 16. Mustard (Old) 17. SR (Creamed Honey) 18. Fake Honey 19. Mustard (New) 20. Foreign Honey 21. Multifloral (Abroad) 22. Forest Honey 27. Pran Honey 28. Dabur

Generally, honey is mildly acidic with an average pH of 3.9. This acidity is due to the minor acid content of honey, mainly amino acids and organic acids that are responsible for the characteristic taste of honey. It is also important to mention that honey from tropical countries is generally characterized by lower acidity. This is due to the water content of these samples resulting in increased fermentation with a further decrease in the pH value. Relatively more acidic values ($pH < 3.24$) indicate improper storage or impure samples.The acidic pH of honey is desirable since acidification has been shown to promote healing by causing oxygen release. pH significantly differed among all the other parameter with different sample at 5% level of significance (Table 7).

Table 7: One way ANOVAbetween groups with mean difference and standard error (SE).

LSD			
Groups	Groups	Mean Difference	SE
pH	Brix	-71.96481*	3.84924
	Moisture	-17.69222*	3.84924
	Fructose	-36.81111*	3.84924

*. The mean difference is significant at the 0.05 level.

CHAPTER V

SUMMARY AND CONCLUSION

In the present study, different honey samples were analyzed for determining pH, moisture, optical rotation, brix and fructose. The results revealed that all honeys had pH, moisture, optical rotation, brix and fructose ranging within 1 to 6; 15.47 to 28.13%; -1.84 to -5, deg cm2 g-1; 69.9 to 82.77%; and 34.03 to 64.33 % respectively. Considering pH, the highest pH (6.0) was recorded in Mustard honey & Foreign honey (Sample no.15 & 20) and lowest pH (1) in Fake honey (Sample no. 18). Low pH impade the presence and growth of microorganisms as well as prevent the growth of many species of bacteria. A one-way ANOVA compares three or more than three categorical groups to establish whether there is a difference between them. pH significantly differed among all the other parameter with different sample at 5% level of significance. In terms of moisture, the highest moisture content (29 %) was obtained in olive honey (Sample no. 13.) and lowest moisture content (15.47 %) was found in mustard honey (Sample no. 15.).Some honey samples were steamed manually and processed in processing plant and their moisture content was found low in moisture content and remained below 20%. Some honey samples were collected as fresh condition and their moisture content was high. Regarding optical rotation, the highest specific rotation (- 5, deg cm2 g-1) was recorded in mustard honey. And the lowest specific rotation (-1.84, deg cm2 g-1) was in litchi honey. Maximum honey samples showed negative specific rotation. So, these honey samples were obtained from floral origin (nectar honey).Considering the brix, the highest brix (82.77) was recorded in mustard honey (sample no.15) and lowest brix (69.9) in olive honey (sample no. 13).Regarding fructose, the highest amount of fructose (64.33 %) was recorded in Khoilsa (Alwoan) (sample no. 6.) and the lowest amount of fructose (34.03 %) was in Mustard honey (sample no. 15.).All samples were significantly different from each other at 5% level of significance.

In conclusion, maximum honey samples investigated fulfilled the quality criteria and those are good for human consumption. The results also revealed excellent acceptability of the honey samples, hence are suitable for human uses.

REFERENCES

Abdullah, F. and Abdulaziz, M. A. (1998). The prophylactic and curative effect of cedar honey induced ulcers in rabbits. *J. Theor. Appl.Sci*.**1**: 26-31.

Achouri, I., Aboussaleh, Y., Sbaibi, R., Chemissi, H. and Bengueddour, R. (2015). Comparison of the physico-chemical quality of honey Ziziphus sp (Sider) and Acacia sp (Samar) consumed in the United Arab Emirates (UAE). *Int. J. Agric. Res. Innov. Technol.***10**(1): 184-191.

Adekanmbi, O. H., Walter, O. J. and Ikegbunam, N. C. (2019). Pollen analysis and heavy metals detection in honey samples from southern Nigeria. *Int. Sci. J*.**26**: 176-190.

Aljohar, H. I., Maher, H. M., Albaqami, J., Al-Mehaizie, M., Orfali, R. and Alrubiaa, S. (2018). Physical and chemical screening of honey samples available in the Saudi market: An important aspect in the authentication process and quality assessmen. *SAUDI PHARM. J.* **26**(7): 932–942.

Almasaudi, S. B., Al-Nahari, A. A. M., El-Ghany, E. S. M. and Harakehg, S. (2017). Antimicrobial effect of different types of honey on Staphylococcus aureusSaudi*. J. Biol. Sci.***24**(14): 1255-1261.

Aloisi, P.V. (2010). Determination of quality chemical parameters of honey from chubut (argentinean patagonia). *Chil. J. Agric. Res.* **70**(4): 640-645.

Anthony, C. R. M. and Balasuriya, D. N. (2016). Electronic honey quality analysis. *J. Inst. Eng*.**49**(3): 41-47.

Azonwade, F. E., Paraiso, A., Dossa, C. P. A., Dougnon, V. T., N’tcha, C., Mousse, W. and Moussa, L. B. (2018). Physicochemical Characteristics and Microbiological Quality of Honey Produced in Benin. *J. Food Qual*. **50**(3): 1-13.

Bang, L. M., Buntting, C. and Molan, P. (2004). The effect of dilution on the rate of hydrogen peroxide production in honey and its implications for wound healing. *J. Altern. Complement Med.* **9**(2): 267-273.

Bogdanov, S. (1999). Honey quality and international regulatory standards: review by the International Honey Commission. Bee World. **80**(2): 61–69.

Bogdanov, S. (2009). Harmonised Methods of the International Honey Commission, http://www.ihc-platform.net/ihcmethods2009.

Bogdanov, S., Jurendic, T., Sieber, R. and Gallmann, P. (2008). Honey for Nutrition and Health: A Review. *J. Am. Coll. Nutr*. **27**(6): 677-689.

Bogdanov, S., Kilchenmann, V. and Fluri, P. (1998). Influence of organic acids and components of essential oils on honey tastes. *Am. Bee j*. **139**: 61-63.

Bogdanov, S., Martin, P. and Lullmann, C. (2002). Harmonised methods of the International Honey Commission. FAM, Liebefeld: Swiss Bee Research Centre.

Brudzynski, K. (2020). A current perspective on hydrogen peroxide production in honey. A review. *Food chemistry*, **120**(2): 678-685.

Can, Z., Yildiz, O., Sahin, H., Akyuz Turumtay, E., Silici, S. and Kolayli, S. (2015). An investigation of Turkish honeys: their physico-chemical properties, antioxidant capacities and phenolic profiles. *Food chemistry*, **180**: 133–141.

Caroli, S., Forte, G., Iamiceli, L. and Galoppi, B. (1999). Determination of essential and potentially toxic trace elements in honey by inductively coupled plasma-based techniques. *Talanta*, **50**: 327- 336.

Cervantes, M. A. R., Novelo, S. A. G. and Duch, E. S. (2000). Effect of the temporary thermic treatment of honey on variation of the quality of the same during storage. *J. Apiacta*.**20**(8): 280-286.

Chakraborti, T. and Bhattacharya, K. (2014). Quality assessment of some Indian honeys in storage through HMF content and invertase activityInt. *J. Pharm. Pharm. Sci*.**6**(2): 827–830.

Chen, c. (2019). Relationship betweenWater Activity and Moisture Content in Floral Honey. *J. food Sci*.**30**(8): 1-18.

Chua, L. S., Rahaman, N. L. A., Adnan, N. A. and Tan, T. T. E. (2013). Antioxidant Activity of Three Honey Samples in relation with Their Biochemical Components. *J. Anal. Methods Chem.* **53**(3): 915-923.

Cimpoiu, C., Hosu, A., Miclaus, V. and Puscas, A. (2013). Determination of the floral origin of some Romanian honeys on the basis of physical and biochemical properties. *Food chemistry*. **100**: 149-154.

Coco, F. L., Valentini, C., Sapra, V. and Ceccon, L. (1996). High performance liquid chromatographic determination of 2-furaldehyde and 5- hydroxymethyl-2-furaldehyde in honey. *J. Chromatogr.* **749**: 95-102.

Codex Alimentarius Commission, (2001). Revised Codex Standard for Honey Codex Stan 12-1981, Rev. 1 (1987), Rev. 2 (2001), *Codex Standard*. **12**: 1–7.

Dele, O. S. (2017). Quality Assessment of Honey Sourced from Natural and Artificial Apiaries in Ekiti State, Nigeria. *Turkish J. Agric. Food Sci. Tech.* **5**(10): 1125-1129.

Draiaia, R., Rezki, A. R., Nacer, K. B. and Chefrour, E. (2014). Quality of Some Algerian Honey: Study of Botanical and Some Physicochemical Parameters. *Middle-East J. Sci. Res.* **22**(9): 1363-1371.

Elbanna, K., Attalla, K., Elbadry, M., Abdeltawab, A., Gamal-Eldin, H. and Ramadan, M. (2014). Impact of floral sources and processing on the antimicrobial activities of different unifloral honeys. *Asian pacific J. Trop. Dis.***4**: 194–200.

Erejuwa, O. O., Sulaiman, S. A. and Wahab, M. S. A. (2012). Fructose Might Contribute to the Hypoglycemic Effect of Honey. *Int. J. Mol. Sci.***17**(2): 1900-1915.

Escuredo, O., Miguez, M., Fernandez-Gonzalez, M. and Seijo, M. C. (2013). Nutritional value and antioxidant activity of honeys produced in a European Atlantic area. *Food Chemistry*. **138**(3): 851-856.

Fallico, B., Zappala, M., Arena, E. and Verzera, A. (2004). Effects of conditioning on HMF content in unifloral honeys. *Food Chemistry*, **85**(2): 305–313.

Ghorbani M., Saei-Dehkordi S. S., Mohebbi A. and Pak A. (2017). Evaluation of some chemical quality characteristics of honey produced in Iran. *J. Food Qual. Hazards control.* **4**: 113-118.

Gul, A. and Pehlivan, T. (2018). Antioxidant activities of some monofloral honey types produced across Turkey. *Saudi J. Biol. Sci.***25**: 1056–1065.

Gulfraz, M., Ifftikhar, F., Asif, S., Raja, G. K., Asad, M. J., Imran, M., Abbasi, K. and Zeenat, A. (2010). Quality assessment and antimicrobial activity of various honey types of Pakistan. *Afr. J. Biotechnol.* **9**(41): 6903-6906.

Hasan, S. H. (2013). Effect of storage and processing temperatures on honey quality. J. Univ. *Babylon Pure Appl. Sci.***21**(6): 2244–2253.

Hossain, M. S., Kabir, M. R., Reybroeck, W. and Alam, M. Z. (2015). Study on the quality of honey collected from three floral sources of Bangladesh. *Int. J. Sci. Res.***5**(7): 2250-3153.

Huidobro, J. F., Santana, F. J., Sanchez, M. P., Sancho, M. T. and Muniategui, S. (1995). Diastase, invertase and ß-glucosidase activities in fresh honey from north-west Spain. *J. Agril. Res.***34**(1): 39–44.

Kamal, A., Raza, S., Rashid, N., Hameed, T., Gilani, M., Qureshi, M. A. and Nasim, K. (2002). Comparative study of honey collected from different flora of pakistan. *J. Biol. Sci.* **2**: 626-627.

Kıvrak, S. and Kıvrak, I. (2017). Assessment of phenolic profile of Turkish honey. *Int. J. Food sci.* **20**: 864–876.

Kowalski, S. (2013). Changes of antioxidant activity and formation of 5-hydroxymethylfurfural in honey during thermal and microwave processing. *J. Agril. Food Chem.* **141**(2): 1378-1382.

Krishnasree, V. and Ukkuru, P. M. (2017). Quality Analysis of Bee Honeys. *Int. J. Current Microbiol. Sci.**6**(2): 626-636.*

Kruzik, V., Gregrova, A., Rajchl, A. and Cizkova, H. (2017). Study on honey quality evaluation and detection of adulteration by analysis of volatile compounds. *J. Apicul. Sci.* **61**(1): 17-27.

Kuçuk, M., Kolayli, S., Karaoglu, L., Ulusoy, E., Baltaci, C. and Candan, F. (2007). Biological activities and chemical composition of three honeys of different types from Anatolia. *Food chemistry*. **100**: 526–534.

Kugonza, D. R. and Nabakabya, D. (2008). Honey Quality as Affected by Handling, Processing and Marketing Channels in Uganda. *Tropicultura*. **26**(2): 113-118.

Kump, P., Neemer, M. and Najder, J. (1996). Determination of trace elements in bee honey, pollen and tissue by total reflection and radioisotope X-ray fluorescence spectrometry. Spectrochimica Acta Part B. Atom. *Spectroscopy*. **51**: 499-507.

Mandal, M. D. and Mandal, S. (2011). Honey: its medicinal property and antibacterial activity. . *J. Apicul. Sci.* **1**(2): 154-160.

Molan, P. C. (2001). Why honey is effective as a medicine? Its use in modern medicine, In Munn, P; Jones, R (eds).5-13.

Molar, P. C. (2012). Potential of honey in the treatment of wounds and burns. J.*Food Sci. Technol.***2**: 13–19.

Obiegbuna, J. E., Osajiele, B. O. and Ishiwu, C. N. (2017). Quality Evaluation of Awka Market Honey and Honey from Beekeepers in Two Floral Regions of Anambra State, Nigeria. *Am. J. Food sci. Technol*. **5**(4): 149-155.

Ozcan, M., Arslan, D. and Ceylan, D. A. (2006). Effect of inverted saccharose on some properties of honey. *Food chemistry*, **99**(1): 24-29.

Pavlova, T. (2018). Quality characteristics of honey: a review. *Proceedings of university of ruse*. **57**(10): 31-37.

Pires, R.M.C., Moura, S. G. D., Filho, F. D. C. C., Monte, A. M., Pires, L. F., Lorezon, M. C. A., Barros, R. O., Pereira,M. M. G. and Muratori, M. C. S. (2015).

Evaluation of hygienic-sanitary quality of honey from *Apis mellifera* L. obtained in semi-arid region of Piauí, Brazil. *African J. Microbiol. Res.* **9**(30): 1806-1813.

Pita-Calvo, C. and Vázquez, M. (2017). Differences between honeydew and blossom honeys: A review. *Trends Food Sci. Technol.***59**: 79–87.

Pridal, A. and Vorlova, L. (2002). Honey and its physical parameters. *Czech J. Food Sci.* **47**(10): 439–444.

Qamer, S., Ahamed, F., Ali, S. S. and Shakoori, A. R. (2013). Effect of Storage on Various Honey Quality Parameters of Apis dorsata Honey from Nepal. *Pakistan j. Zool.* **45**(3): 741-747.

Rahman, M. M., Karmoker, P. and Alam, M. Z. (2017). Quality evaluation of some selected commercial honey products available in the market of Bangladesh. *J. Fundam. Appl. Agricul.* **2**(3): 326-330.

Ribeiro, R. D. O., Marsico, E. T., Carneiro, C. D. S., Monteiro, M. L. G., Junior, C. C. and Jesusc, E. F. O. D. (2014). Detection of honey adulteration of high fructose corn syrup by Low Field Nuclear Magnetic Resonance (LF 1H NMR). *J. Food Eng.* **135**: 39-43.

Rybak-Chmielewska, H. (2007). High performance liquid chromatography (HPLC) study of sugar composition in some kinds of natural honey and winter stores processed by bees from starch syrup. *J. Apicul. Sci.* **51**(1): 23–37.

Sahinler, N. and Gul, A. (2005). Effect of heating and storage on honey hydroxymethylfurfural and diastase activity. *J. Food Technol.* **3**(2): 152–157.

Sarker, N., Chowdhury, M. A. Z., Fakhruddin, A. N. M., Fardous, Z., Moniruzzaman, M. and Gan, S. H. (2015). Heavy metal contents and physical parameters of aegiceras corniculatum, brassica juncea, and litchi chinensis honeys from Bangladesh. *Biomed Res. Int.* **10**: 1-7.

Serra, B. J. and Ventura, C. F. (1995). Characterization of citrus honey (Citrus spp.) produced in Spain. *J. Agricul. Food Chem.* **43**: 2053-2057.

Shambaugh, P., Worthington, V. and Herbert, J. H. (1990). Differential effects of honey, sucrose, and fructose on blood sugar levels. *J. Agricul. Food Chem.* **13**(6): 322-325.

Singh, I. and Singh, S. (2018). Honey moisture reduction and its quality. *J. Food sci. Technol.* **55**(10): 3861–3871.

Terrab, A., Gonzalez, A. G., Diez, M. J. and Francisco, J. H. (2003). Characterization of Moroccan unifloral honeys using multivariate analysis. *J. Eur. Food Res. Technol.* **218**: 88-95.

Tucak, Z., Periskic, M., Beslo, D. and Tucak, I. (2004). Influence of the Beehive Type on the Quality of Honey. *Collegium antropologicum*. **28**(1): 463–467.

Turkmen, N., Sari, F., Poyrazoglu, E. S. and Velioglu, Y. S. (2006). Effects of prolonged heating on antioxidant activity and colour of honey. *Food chemistry*. **95**(4): 653-657.

White, J. W. and Landis, W. D. (1980). Honey composition and properties 1st Edn., Beekeeping in the US Agriculture, USA.

Willix, D. J., P C Molan, P.C. and Harfoot, C.G. (1992). A comparison of the sensitivity of wound-infecting species of bacteria to the antibacterial activity of manuka honey and other honey. *J. Appl. Bacteriol.* **73**(5): 388-394.

Wu, D. and Cederbaum, A. (2003). Alcohol, oxidative stress, and free radical damage. *Alcohol research & health*. **27**: 277–284.

Zerrouk, S. H., Fallico, B. G., Arena, E. N., Ballistreri, G. F. and Boughediri, L. A. (2011). Quality Evaluation of Some Honey from the Central Region of Algeria. *Jordan J. Biol. Sci.* **4**: 243-248.