

Chemical and Physical Properties of Adulterated Honey and Developing Means of Identifying Adulterants

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Abstract: Honey is defined as a naturally sweet mixture produced by bees (Apis mellifera) from the nectar or from secretions of living parts of plants or excretions of plant sucking insects on the living parts of plants transform and combine with specific substances of their own (such as enzymes), deposit, dehydrate, store and leave in the beeswax honeycombs to ripen and mature. It is typical sugary foodstuff; according to present-day regulations, apart from other forms of honey no other or additives can be added to it. The aim of this study was to develop identification means of adulterated honey using chemical and physical analysis. Adulteration of honey was obtained by mixing and homogenizing major honey adulterants so far known like sugar, banana, "sheb", molasses and water with different ratio. Moisture content, electrical conductivity, PH, ash, free acidity (FA) and Hydroxymehylfurfural of adulterated honey in all treatments group (P<0.05). Highest FA content (458.33 ± 1.66 meq/kg) recorded in (honey: sugar: water: sheb) treatment. Adulteration affects the purity and quality properties of honey, by using the ash content, free acidity and electrical conductivity adulteration can be identified and color parameter is not recommended for identification of adulterated honey.

Keywords: adulteration, adulterant, honey, chemical properties

1. INTRODUCTION

Honey is defined as a naturally sweet mixture produced by bees (*Apis mellifera*) from the nectar of flowers, from secretions of parts of the living plants or excretions of plant-sucking insects on the living part of plants that the honey bees collect, transform and combine with specific substances of their own (such as enzymes), deposit, dehydrate, store and leave in the beeswax honeycombs to ripen and mature (Codex Alimentarius, 2010). Various physical types (pressed, centrifuged and drained) and forms (comb, chunk, crystallized or granulated, creamed and heat-processed) of honey are on the market (Crane, 1990). According to the definition of the Codex Alimentarius and other international honey standards honey shall not have any added food ingredient than honey to it nor shall any particular constituent be removed from it.

Adulteration usually refers to mixing other matters (substance(s) of an inferior and sometimes harmful quality with food or drink that contains naturally similar substance (Cordella, 2003; Pilizota and Tiban, 2009). Adulteration of honey with cheaper sweetening materials has been widely reported in various literatures (Martin et al., 1997). Honey adulteration appeared on the world market since 1970s when high-fructose corn syrup was introduced by the industry (Pilizota and Tiban, 2009). Recently honey is ranked third in the list of globally adulterated products (Zhou et al., 2018). Due to the superior nutritional and health value and unique flavor, natural honey is preferred by consumers; hence, the price of bee honey is much greater than other sweetening commodities, such as refined cane sugar and corn syrup. The high price of natural honey encourages workers in the honey industry, including beekeepers and merchants, to adulterate honey worldwide, which leads to deterioration of honey quality, but increases honey quantity that is sold at the same price of natural authentic honey (Mouazen and Al-Walaan, 2014). Other adulterating materials such as molasses inverted by acids or enzymes from corn, sugar beet and syrups of natural origin such as maple (Pilizota and Tiban, 2009); banana, corn flour and materials like soil had been reported (Meseret and Taye, 2017).

In Ethiopia also, honey adulteration has become a serious problem affecting both the local and the export market of the country (Gemechis, 2016). Problems of honey fraud negatively influence market

growth by damaging consumer confidence (Cabañero et al.2006). Recent report by (Meseret and Taye, 2017) indicated that about 60% of honey merchants claimed they bought adulterated honey, the major (about 69%) adulterant material being sugar though other materials were listed too. However, the study conducted before did not include the chemical analysis of adulterated and pure honey, except undertaking some observations of physical changes adulterated honey displays. However, to guarantee authenticity of honey and protect the consumer from commercial exploitation, the quality of honey must be controlled analytically (Mendes et al., 1998). The European Commission encourages the use of analytical methods to determine the authenticity of honeys (Morales et al., 2008). Therefore, this research work aims to characterize honey adulterated with different major adulterants based on specific physical properties and chemical compositions acquired with conventional analytical methods.

2. MATERIALS AND METHODS

2.1. Sample Preparation

The major known adulterant materials were prepared in different percent (ratios) with pure honey in laboratory. These major adulterant materials were identified previously (Meseret and Taye, 2017). The major adulterant materials identified in the country were commercial sugar, banana, molasses, sheb (shebeb). Three replicates were taken to determine the chemical properties of each adulterated honey samples. The analyses were conducted in Holeta Bee Research Center and the percentage of honey with major identified and known honey adulterants were prepared as follows.

Adulteran ts with honey	Ratios of adulterants to honey (treatments)							
Honey: sugar	0:1	1:1	1:2	2:1	1:0.5	5:1	10:1	20:1
Honey: sugar : water: sheb	4:4:1:0.1	2:2:0.5:0 .1	3:4:1:0.1	5:2:0.5:0. 15	4:2:0.5:0. 25	1:2:0.5:0. 25	2:2:0.5:0 .3	2:1:1:0. 4
Honey: molasses	0:1	1:1	2:1	1:2	1.5	5:1	10:1	20:1
Honey: banana : molasses	0:1:1	1:1:1	4:1:1	1:2:2	2:1:2	10:1:1	20:2:1	
Honey :sugar: banana	0:1:1	1:1:1	0.25:0.25 :1	0.5:1:1	0.25:1:1	1:0.5:1	10:1:1	20:1:1
Honey : banana: molasses	0:1:2.5:0 .5	2:1:2.5:0 .5	1.:0.5:4:1	1:1:2:1	0.5:1:2:0. 5	1:0.5:1.5: 1	10:1:1:1	20:1:1: 1

Table1. Ratio of adulterants to honey	Table1.	Ratio	of	adulterants	to	honey
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2.2. Physiochemical Properties

2.2.1. Colour

Colors of the pure and adulterated honey samples were measured using Pfund honey color grader (Koehler Bohemia. NY). Hundred grams (100 g) of adulterated honey was poured into the sample holder of the Pfund grader. Determination was based on the matching of the honey sample colors with the color indexes present in the glass Pfund grader. Color of pure honey was determined in the same way and compared with that of adulterated honey.

2.2.2. Moisture

Moisture content was determined using Abbe refractometer (ABBE- 5 Bellingham Stanley. Ltd, United Kingdom) at 20°C. Directly after homogenization of honey, the surface of the prism was evenly covered with the sample of honey and after 2 minutes the reading of the refractive index was recorded. Distilled water (1.3330) was used as a reference. The refractive index reading was converted to moisture content (g/100 g) using (AOAC, 1990).

2.2.3. Hydroxymethylfurfural

Hydroxymethylfurfural (HMF) content was determined using 6800 UV-Vis spectrophotometer (JENWAY, United Kingdom). Different reagents were prepared as follow: Carrez solution I was prepared by weighing 15 g of potassium hexacyanoferrate (II), K₄Fe (CN) 6•3H₂O and dissolving it in water. Then, the solution was made up to 100 ml. Carrez solution II, 30 g of zinc acetate, Zn(CH₃.COO)₂.2H₂O was diluted and made up to 100 ml., 0.20 g of solid sodium hydrogen sulphite, NaHSO₃, diluted to 100 ml. Carrez solution is clarification reagents to remove these interfering compounds from the analytes. Five gram (5 g) of honey was weighed into a 50 ml beaker and dissolved the sample in 25 ml of water and transferred quantitatively into a 50 ml volumetric flask. A 0.5 ml of Carrez solution I was added with 0.5 ml of Carrez solution II, mixed well and made up to the mark with water and filtered through filter paper (general purpose); the first 10 ml of the filtrate was rejected. Five milliliters was pipetted in each of two 2 test tubes and 5 ml of water was added to one of the test tubes and mixed well (the sample solution). Five milliliters of sodium bisulphite solution, 0.2% was added to the second test tube and mixed well. By subtracting the absorbance measured at 284 nm for HMF in the honey sample solution against the absorbance of reference (the same honey solution treated with sodium bisulphite, 0.2%) at 336 nm as described in harmonized method (AOAC, 1990) method. HMF content of honey was calculated using the following equation:

HMF in mg/kg honey = $(A_{284}-A_{336}) \times 149.7 \times 5 \times D/W$

Where A284= absorbance at 284 nm, A336= absorbance at 336 nm, Factor = 149.7=(126/1683)(1000/10)(1000/5), 126 = molecular weight of HMF, 16,830 = molar absorptive of HMF at 284 nm, 1000 = conversion of g into mg, 10 = conversion of 5 into 50 mL, 1000 = conversion g honey into 1000 g, 5 = nominal sample weight, D= Dilution factor and W= weight in g of the honey sample

2.2.4. Free Acidity and PH

Free acidity of honey is the content of all free acids, expressed in milli equivalents per kilogram honey (meq of acid/kg) was determined using pH meter (METTLER TOLEDO, CHINA) and titrating until pH value of 8.3. Ten gram of honey sample was dissolved in 75 ml distilled water in 250 ml volumetric flask and the standardized 0.1M NaOH and the dissolved honey sample was titrated with standardized 0.1M NaOH to pH 8.3 using pH glass electrode attached to pH meter as end point indicator. The pH value was determined by using glass electrode after calibration with standard buffer solution pH 4, 7 and 10 (AOAC, 1990) method.

2.2.5.Ash

Ten gram of the honey samples (m_0) was weighed into a pre-weighed crucible (m_2) and two drops of olive oil was added to the honey sample. Then, ash dish (m_1) with honey was placed on hotplate to remove water from the honey at low heat rising to 350 -400°C and after the preliminary ashing, and the dish was placed in the preheated a muffle furnace at 600°C until ashing complete (Bio Base JKKZ .5.12GJ Muffle Furnace, Shandong. ldt, China) and was heated for 1 hour and the ash dish cooled down in the desiccators (m_1) and weighed as described in harmonized IHC (Bogdanov, 2009). The proportion of ash (W_A) in g/100 g honey was calculated using the following formula:

$W_A = (m_1 - m_2)/m_0)*100$

Where: m_0 = weight of honey sample, m_1 = weight of crucible with ash, m_2 = weight of dish.

2.2.6. Electrical Conductivity

The electrical conductivity of a solution of 20 g dry matter of honey in 100 ml distilled water was measured using an electrical conductivity cell (BANTE Instrument- 520 conductive and temperature meter, China). A 0.745 g of potassium chloride, was dried at 130°C, dissolved in freshly distilled water in a 100 ml flask and filled to volume with distilled water. Forty milliliters of the potassium chloride solution was transferred to a beaker and the conductivity cell connected to the conductivity meter, the cell was rinsed thoroughly with potassium chloride solution and immersed in the solution, together with a thermometer and reading of the electrical conductance of the solution in mS after the temperature equilibrated to 20° C was taken as described in harmonized IHC (Bogdanov, 2009). The cell constant K, was calculated using the following formula:

K=11.691x1/G

Where: K=the cell constant in cm^{-1} , G= the electrical conductance in mS, measured with the conductivity cell, 11.691= the sum of the mean value of the electrical conductivity of freshly distilled water in mS.cm⁻¹ and the electrical conductivity of a 0.1M potassium chloride solution, at 20°C.

3. RESULTS AND DISCUSSION

3.1. Color

The color of honey is a useful parameter for the characterization of the product. Color is the single most important factor determining import and wholesale prices (Belay *et al.*, 2015). The color of the honey and sugar adulteration, grouped as dark to light amber, the adulterated honey with the sugar :water and sheb color were light amber, the color of adulterated honey from honey and molasses amber to dark, adulterated honey from honey : banana : molasses was found to be amber to dark and adulterated honey prepared from honey : sugar: banana color found to be white. The variation among the adulterated honey types was due to the adulterating material of the honey. The color of the honey samples from the various regions could not be the single determinant of adulteration.



Figure1. Honey: sugar

Figure 2. Honey: molasses



Figure3. Honey: sugar: banana



Figure4. Honey: banana: molasses



Figure5. Pure honey

Figure6. Honey: Sugar: water: sheb (shebeb)

Treatment	Parameters (mean ±SE)							
	HMF	pН	EC	Ash (%)	Free Acidity	MC (%)		
	(mg/kg)		(mS/cm)		(meq/kg)			
Honey:sugar:water:sheb (4:4:1:0.1)	0.00 ± 0.00^{d}	3.25±0.16 ^b	0.83±0.06 ^b	0.39±0.03 ^b	161.00±10.54 ^b	14.5±0.00 ^{de}		
Honey:sugar:water:sheb (2:2:0.5:0.1)	0.00 ± 0.00^{d}	3.24±0.13 ^b	0.80±0.35 ^b	0.38±0.20 ^b	110.33±19.46 ^b	14.0±0.28 ^{de}		
Honey:sugar:water:sheb (3:4:1:0.1)	0.00 ± 0.00^{d}	3.39±0.15 ^b	0.59±0.09 ^b	0.25±0.05 ^b	102.33±5.24 ^b	13.0±0.28 ^f		
Honey:sugar:water::sheb (5:2:0.5:0.15)	23±8.03 ^b	3.26±0.15 ^b	1.17±0.02 ^{ab}	0.51±0.01 ^{ab}	168.33±5.24 ^b	14.9±0.10 ^{cd}		
Honey:sugar:water:sheb (4:2:0.5:0.2)	35.7±8.20ª	3.18±0.16 ^b	1.33±0.09 ^{ab}	0.68±0.05 ^{ab}	212.33±6.22 ^b	15.6±0.16 ^{bc}		
Honey:sugar:water:sheb (1:2:0.5:0.25)	0.00 ± 0.00^{d}	3.03±0.05 ^b	0.88±0.26 ^b	0.42±0.14 ^b	452.66±6.74 a	14.4±0.21 ^{de}		
Honey:sugar:water:sheb (2:2:0.5:0.3)	0.00 ± 0.00^{d}	3.26±0.11 ^b	1.94±0.71ª	1.03±0.40ª	458.33±1.66 ^a	13.9±0.16 ^{ef}		
Honey:sugar:water:sheb (2:1:1:0.4)	0.00 ± 0.00^{d}	3.27±0.12 ^b	1.09±0.38 ^{ab}	0.54±0.21 ^{ab}	252.33±112.3 ^b	19.1±0.10 ^a		
Pure honey	9.96± 0.12°	4.46±0.04 ^a	0.69±0.02 ^b	0.47±0.03 ^{ab}	14.66±2.18 °	16.02±0.26 ^a		
Ethiopian honey standard	≤40		≤0.8	≤0.6	≤40	≤21		

Table2. Analytical results of honey samples adulterated with sugar, water and sheb by different ratio

Different letters down column showed significant difference (p < 0.05)

Table3. Test results of honey samples adulterated with molasses by different ratio

Treatment	Parameters (mean ± SE)								
	HMF	pН	EC	FA (meq/kg)	Ash (%)	MC (%)			
	(mg/kg)		(mS/cm)						
Honey:molasses (0:1)	49.63±1.16°	5.80±0.00 ^a	8.23±7.7 ^{ab}	164.77±2.90 ^a	4.65±4.4 ^{ab}	7.27±1.23 ^e			
Honey:molasses (1:1)	0.00±0.00 ^e	5.59±0.05 ^{bc}	8.52±1.85 ^{ab}	107.00±5.13 ^{bc}	4.81±1.06ab	14.31±1.81 ^d			
Honey:molasses (2:1)	0.00±0.00e	5.38±0.09 ^{abc}	6.98±5.4 ^{ab}	86.00±9.00 ^{cd}	3.93±3.13 ^{ab}	19.83±0.72 ^b			
Honey:molasses (1:2)	19.6±.10.1 ^d	5.66±0.06 ^{ab}	15.83±1.3ª	127.33±2.66 ^b	9.04±0.79ª	14.63±0.39 ^d			
Honey:molasses (1:5)	130.3±.88 ^a	5.53±0.03 ^{abc}	7.96±0.18 ^{ab}	90.77±4.33°	4.49±0.46 ^{ab}	14.35±0.08 ^d			
Honey:molasses (5:1)	86.5±3.75 ^b	4.58±0.59 ^{bc}	9.10±2.14ab	64.77±.33 ^{de}	5.15±1.23 ^{ab}	19±0.00bc			
Honey:molasses (10:1)	0.00±0.00e	4.70±0.12 ^{bc}	9.32±3.7 ^{ab}	49.33±1.76 ^{ef}	5.27±2.16 ^{ab}	17.76±0.14 ^{bcd}			
Honey:mollases (20:1)	0.00±0.00 ^e	4.54±0.11°	1.31±0.40 ^{ab}	40.00±5.03 ^f	0.67±0.23 ^b	24.16±0.29ª			
Pure honey	9.96±0.12 ^{de}	4.46±0.04°	0.69 ± 0.02^{b}	14.66 ± 2.18	0.47 ± 0.03^{b}	16.02±0.26 ^{cd}			
Ethiopian	≤40		≤0.8	≤40	≤0.6	≤21			
honey									
standard									

Different letters down column showed significant difference (p < 0.05)

Table4. Test results of honey samples adulterated with sugar and banana by different ratio

Treatment						
	HMF	pН	Ash (%)	EC	FA	MC (%)
	(mg/kg)			(mS/cm)	(meq/kg)	
Honey :sugar:banana (0:1:1)	0.00 ± 0.00^{b}	4.9±0.10 ^{ab}	0.43±0.12 ^{ab}	0.90±0.22 ^{ab}	33.66±2.18 ^{ab}	23.23±1.6 ^{bc}
Honey:sugar:banana (1:1:1)	0.00 ± 0.00^{b}	4.52±0.12 ^{bc}	0.40±0.12 ^{ab}	0.84±0.20 ^{ab}	35.00±0.57 ^{ab}	18.61±0.59 ^{de}
Honey:sugar:banana (0.25:0.25:1)	0.00 ± 0.00^{b}	4.50±0.17 ^{bc}	0.36±0.14 ^b	0.77±0.24 ^b	41.66±4.84 ^{ab}	24.8±0.86 ^{bc}

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Honey:sugar:banana (0.5:1:1)	0.00 ± 0.00^{b}	5.30±0.18 ^a	3.08±1.53 ^a	5.50±2.66 ^a	66.33±16.8ª	38.27±0.14 ^a
Honey:sugar:banana (0.25:1:1)	0.00 ± 0.00^{b}	4.70±0.14 ^{ab}	0.37 ± 0.04^{b}	0.79 ± 0.07^{b}	39.33±5.17 ab	19.45±0.93 ^{cd}
Honey:sugar:banana (1:0.5:1)	0.00 ± 0.00^{b}	4.57±0.13 ^{abc}	$0.52 \pm .08^{ab}$	1.04±0.14 ^{ab}	39.00±1.15 ^{ab}	26.23±0.48 ^b
Honey:sugar:banana (10:1:1)	2.46±0.15 ^b	4.03±0.21°	0.65±0.16 ^{ab}	1.27±0.28 ab	28.66±0.88 ^b	24.53±0.16 ^{bc}
Honay : sugar:banana (20:1:1)	5.53±0.28 ^{ab}	4.05±0.18°	0.17±0.01 ^b	0.43±0.01 ^b	38.33±7.75 ^{ab}	24.33±0.33 ^{cd}
Pure honey	9.96±0.12 ^a	4.46 ± 0.04^{bc}	0.47 ± 0.03^{b}	0.69 ± 0.02^{b}	14.66±2.18 ^b	16.02±0.02 ^e
Ethiopian honey	≤40		≤0.6	≤0.8	≤40	≤21
standard						

Different letters down the column showed significant difference (p < 0.05)

 Table5. Test results of honey samples adulterated with sugar

Treatment	Parameters mean±SE							
	HMF	\mathbf{P}^{H}	Ash (%)	EC	FA	MC (%)		
	(mg/kg)			(mS/cm)	(meq/kg)			
Honey:sugar (1:1)	0.00 ± 0.00^{b}	3.21±0.02 ^c	0.04 ± 0.01^{b}	0.19±0.01°	78.66 ± 0.66^{a}	13±0.28 ^e		
Honey:sugar (2:4)	47.1±6.23 ^a	3.74±0.03 ^b	0.20 ± 0.05^{b}	0.48±0.10 b	36.33±5.23 ^b	13.5±0.29 ^{cd}		
Honey:sugar (5:1)	0.00±0.00 ^b	3.86±0.11 ^b	0.17±0.02 ^b	0.44±0.04 ^b	32.00±4.16 ^b	13.16±0.16 ^e		
Honey:sugar (10:1)	1.36 ± 1.36^{ab}	4.04 ± 0.14^{ab}	0.22 ± 0.02^{b}	0.52 ± 0.04^{ab}	27.66±2.33 ^{bc}	14.33±0.16bc		
Honey:sugar (20:1)	5.63±5.28 ^{ab}	4.06±0.17 ^{ab}	0.12 ± 0.05^{b}	0.35±0.11 ^{bc}	28.66 ± 2.40^{bc}	15±0.00 ^{ab}		
Pure honey	9.96±0.12 ^b	4.46 ± 0.04^{a}	0.47 ± 0.03^{a}	0.69±0.02 ^a	14.66±2.18°	16.02±0.26 ^a		
Ethiopian honey standard	≤40		≤0.6	≤0.8	≤40	≤21		

Different letters down the column showed significant difference (p < 0.05)

 Table6. Test results of honey samples adulterated with banana and molasses

		Parameters (mean ±SE)						
	HMF							
Treatment	(mg/kg)	pН	Ash (%)	EC (mS/cm)	FA (meq/kg)	MC (%)		
Honey:banana:molasses					102.33±18.26			
(0:1:1)	35.33±11.04 ^a	5.64 ± 0.06^{a}	4.02±1.99 ^{ab}	7.14 ± 3.74^{ab}	ab	58.6±2.7 ^a		
Honey:banana:molasses								
(1:1:1)	0.00±0.00 ^b	5.42±0.07 ^{ab}	2.69±1.01 ^{ab}	4.82±1.75 ^{abc}	78±5.68 ^{abcd}	53.09±3.63ª		
Honey:banana:molasses				1.96±0.09 ^{bc}				
(4:1:1)	0.00 ± 0.00^{b}	5.19±0.07 ^{bc}	1.05 ± 0.05^{b}		56±3.51 ^{bcde}	37.83 ± 3.58^{b}		
Honey:banana:molasses								
(1:2:2)	0.00 ± 0.00^{b}	5.55±0.05 ^{ab}	3.79±1.41 ^{ab}	6.74 ± 2.46^{abc}	90.3±6.35 ^{abc}	36.90±0.26 ^b		
Honey:banana:molasses								
(2:1:2)	0.00 ± 0.00^{b}	5.58±0.12 ^a	4.78±0.03 ^a	8.47 ± 2.33^{a}	116.1±19.21 ^a	23.93±1.79°		
Honey:banana:molasses								
(10:1:1	0.00 ± 0.00^{b}	4.95±0.04°	2.96±0.56 ^{ab}	5.30±0.05 ^{abc}	42.6±1.86 ^{cde}	24.13±0.18°		
Honey:banana:molasses								
(20:2:1)	$0.00 \pm 0.00^{\circ}$	4.56±0.11 ^d	1.60±0.39 ^{ab}	2.93±0.98 ^{abc}	34.3±1.88 ^{de}	9.54 ± 0.44^{d}		
Pure honey	9.96±0.12 ^b	$4.46 \pm .04^{d}$	0.47±0.03 ^b	0.69±0.02 ^c	14.67±2.18 ^e	16.02±0.26 ^{cd}		
Ethiopian honey								
standard	≤40		≤0.6	≤0.8	≤40	≤21		

Where SE is the standard deviation of each measurement values. EC is Electrical conductivity, HMF, Hydroxymethylfurfural, FA, Free Acidity and MC, Moisture Content. Different letters down the column showed significant difference (p < 0.05)

3.2. Free Acidity

Free acidity content indicate the freshness of the honey samples and absence of unwanted fermentation in the honey sample. In tables the free acidity content increase when adulterating

material ratio increase which means, the pure honey free acidity content is significantly different from adulterated honey with sugar, banana, molasses, sheb and water (P<0.05) as showed in (Table 2) indicate fermentation the honey and highest FA content observed in honey: sugar: water: sheb (2:2:0.5:0.3) treatment 458.33 ± 1.66 meq/kg of honey. Most of adulterated samples with the highest ratio of the adulterating material free acidity content were above limit set by CA, EU and Ethiopian standards. The maximum limit for free acid set by the CA is 50 meq/kg of honey, while the EU and Ethiopian standard is 40 meq/kg of honey (Bogdanov, 2007).

3.3. HMF Content

The result of this study showed that there is significant difference in HMF content between the adulterated honey and pure honey and among the group of adulterating materials and ratios of adulterants (Table 2-5). The highest HMF content $(130.3\pm0.88 \text{ mg/kg})$ was recorded in honey adulterated with molasses in ratio of 1:5 (honey to molasses) as mentioned in Table 3. However, it was observed that HMF of the adulterated honey was found to be lower than the pure honey. This might be due to the freshness or unheated or properties of the adulterating materials such as banana, sugar and water used.

3.4. PH Value

In general, honey is acidic in nature irrespective of its variable geographical origin. The main acid is gluconic acid. pH content of pure honey were significantly different from the adulterate honey with molasses, banana, sugar, sheb, water as showed in (Table 2-6). The high acidity of honey correlates with the fermentation of sugars present in the honey into organic acid, which is responsible for two important characteristics of honey flavor and stability against microbial spoilage (Bogdanov et al., 2008) and the highest pH value recorded in honey: molasses (0:1) treatment group 5.80 ± 0.00 (Table 3).

3.5. Moisture Content

The moisture content is an important criterion for assessing the ripeness of the honey and its shelf-life. In general, high amount of water causes the honey to ferment, to spoil and to lose flavor, with following honey quality loss. Moisture content of adulterated honeys was significantly different from pure honey at (p<0.05) as shown (Table 2-6) and highest moisture content was recorded on honey: banana: molasses (0:1:1) adulteration types $58.6\pm2.7\%$ (Table 6) adulteration of honey with those material can increase the moisture content of honey this can lead to fermentation and spoilage of honey. An increase in moisture content of honey is indicative of adulteration the result of this study was supported by different pervious findings (Lawal et al., 2009, Kingsta et al. 2018) found that higher the adulterant added to pure honey.

3.6. Electrical Conductivity

The result shows that almost in all of the adulterant samples, higher conductivity records were obtained. Honey and molasses adulterated sample with different ratio found to be the highest electrical conductivity ranges 15.83 mS/cm (Table 3). Significant differences were observed between pure honey and adulterated honey treatment group (Table 2-6) and some of the results were above the limit of Ethiopian and EU standard that is greater than 0.8 mS/cm. The results also had shown that in all of the adulterated honey samples the highest is the electrical conductivity has the highest ash and acid contents which agrees with that of (Bogdanov *et al.*, 2002).

3.7. Ash Content

The ash content is a measure of the mineral elements in the honey samples and directly measure inorganic residue after carbonization (Lawal *et al.*, 2009). According to the result, most of the ash contents of above the standard limit. This indicates high mineral content which could be due to adulteration because the pure honey ash content is within the standard limit, the highest ash content were obtained from adulteration of mean $9.04\pm.79$ % honey: molasses (1:2) (Table 3) and that of pure honey were significantly different from adulterated honey as shown (Table 2-6). This result also agrees with study result of (Ribeiro *et al.*, 2014) detection of honey adulteration of high fructose corn syrup by Low Field Nuclear Magnetic Resonance (LF 1H NMR).

4. CONCLUSION AND RECOMMENDATION

In current study, well known honey samples were used to distinguish pure honey from adulterated honey with molasses, sugar, banana, "sheb" and water using chemical properties. The study indicated that

adulterated honey sample could be identified from pure honey sample by using the ash content, free acidity and electrical conductivity. However, according to this result HMF content may not identify adulteration of honey because of HMF content values affected by over heating or aging of honey. This result also showed that color parameter could not identify adulteration of honey. It can be concluded that adulteration can affect the chemical and quality properties of honey. Color parameter is not recommended for identification of adulterated honey. Therefore, users must be take care of buying fake honey based on color which is similar to pure honey but it may be adulterated honey which may affect the health and economy of the community. In addition, any honey bought from local market or mobile traders should be seen in laboratory before consuming.

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