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Evaluation of Physicochemical Properties of Honey Bees *(Apis Mellifera)* in Godere Woreda, Gambella, Ethiopia

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Abstract This study was conducted to evaluate the physico-chemical properties of honey bees in Godere woreda, Gambella region, Ethiopia. 12 selected honey samples from three locations of the study area (Metti town, Ashine and Cabo) were evaluated for proximate composition (moisture, protein, fat, total ash contents and energy values) and sugar contents (fructose, glucose, sucrose and reducing sugar contents). The honey samples were analyzed following the techniques proposed by the Ethiopian Conformity Assessment Enterprise (ECAE), European Union (EU) and Codex standards for honey. The overall mean of moisture, total ash, protein, fats and CHO contents of the honey samples were 18.76±1.09, 0.34±0.05, 0.37±0.03, 80.02±1.03g/100g, respectively with average energy value of 1,362.32±18.16 KJ/100g. The average sugar contents of the samples were 38.64±0.61g/100g (fructose), 36.37±2.14g/100g (glucose), 74.19±1.88g/100g (reducing sugar) and 2.66±0.23g/100g (sucrose). All the physicochemical parameters laid with-in the limits of national and international standards set by Ethiopian Conformity Assessment Enterprise for analyzing honey samples (ECAE), Codex Alimentarius Commission and EU Council.

Keywords: *honey bee, physico-chemical properties, proximate composition, sugar content*

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1. Introduction

Natural honey is one of the most widely sought products due to its unique nutritional and medicinal properties, which are attributed to the influence of the different groups of substances it contains. It is also used for industrial purposes and it is an important commodity in the international market; serving as foreign exchange earner for many countries [1,2]. Besides being healthy and easy to digest, this natural product is full of carbohydrates, vitamins, minerals and enzymes [3].

Honey contains different quantities of minerals ranging from 0.02 g/ 100 g to 1.03 g/100 g, with potassium being the most abundant element comprising approximately one-third of the total mineral content [4]. Macromineral elements, such as potassium, calcium, and sodium, and trace minerals, such as iron, copper, zinc and manganese play a critical role in biological systems. These elements maintain normal physiological reactions, induce general metabolism, germination, circulatory systems and influence reproduction as catalysts of various biochemical reactions (Stanis˙kien *et al*., 2006).

Ethiopia is one of the top 10 of honey producers in the world and it is the largest one in Africa [5]. The total volume of honey production in 2011 was estimated to be 39.89 million kilograms (kg) [6]. The country's potential for honey production, the variety of natural honey flavors associated with the country's diverse sources of bee forage and Ethiopian honey's desirable qualities, such as low moisture content, have been widely recognized. (Tadesse and Phillips, 2007). Traditional beekeeping and honey hunting is still practiced at different areas of the country including in Godere woreda, but the honey produced by this system contains wax, pollen and other impurities, which affect the quality and market value of honey [7].

2. Materials and Methods

2.1. Sample Collection and Preparation

Twelve honey samples harvested in the year 2015/16, were collected purposively from local modern and traditional honey producers each separately from different locations of the study area. All the samples were collected freshly in air tight sterile containers (labeled with numbers, place and date of collection) and stored at ambient temperature, in a dry room free of odors and ants and then and it was analyzed in the laboratories [8]. Unwanted materials such as wax sticks, dead bees and particles of combs were removed by straining the samples through sieves before analysis [9].

2.2. Chemical Composition Analysis

Chemical compositions of the honey samples were determined using the methods of AOAC [10,11].

2.2.1. Moisture Content

Moisture content was determined using AOAC [11]; the official method 925.10.

$$\text{Moisture Content } (\%) = \left[\frac{(M_2 - M_3)}{(M_2 - M_{1)}}\right] * 100$$

Where:

- M_1- weight of crucible(g)
- M_2 - weight of crucible and sample(g)
- M_3- crucible and dried sample weight (g)
- $(M_2\text{-}M_3)$ - weight of water in the sample (g).

2.2.2. Determination of Protein

The protein content was determined using AOAC, [11]; 920.87. Total nitrogen content and the protein content was calculated using the 6.25 conversion factor for protein nitrogen [9].

$$\text{Nitrogen } (\%) = \left[\frac{(V - B) * N * 14.007)}{W}\right] * 100$$

Where:

- B = H_2SO_4 solution (ml) for the blank test
- V = H_2SO_4 solution (ml) for the test material
- N = Normality of standard sulfuric acid (0.1N).
- W = sample weight on dry matter basis
- 14.007 = molecular weight of nitrogen.

2.2.3. Determination of Crude Fat

The Crude fat was evaluated based on the Soxhlet extraction method of AOAC, [11]; 920.39.

$$\text{Crude fat } (\%) = \left[\frac{(M_2 - M_1)}{M}\right] * 100$$

Where:

- M = Weight of the initial sample (g)
- M_1 = Weight of the extraction flask (g)
- M_2 = Weight of flask and dried crude fat (g).

2.2.4. Determination of Total Ash

Total Ash content was determined using AOAC, [11]; 923.03.

$$\text{Total Ash } (\%) = \left[\frac{(M_3 - M_1)}{(M_2 - M_{1)}}\right] * 100$$

Where:

- M_1 = mass of the crucible(g)
- M_2 = mass of crucible and the sample(g)
- M_3 = mass of crucible and dried sample(g)
- $(M_2\text{-}M_1)$ = initial sample weight (g)
- $(M_3\text{-}M_1)$ = Weight of ash in (g).

2.2.5. Carbohydrate Contents (CHO)

Total percentage of carbohydrates was determined using the difference method (Onyeike *et al.,* 1995)

$$\%\mathbf{CHO} = 100\% - \left[\begin{matrix}(\%\text{ moisture}) + (\%\text{ fat}) \\ + (\%\text{ protein}) + (\%\text{ ash})\end{matrix}\right].$$

2.2.6. Energy Value (kcal/100gm)

Energy value was evaluated according to Onyeike *et al.,* 1995.

$$\text{Energy } (\text{kcal}/100\text{g}) = \left[(4 * \%\text{protein}) + (4 * \%\text{CHO}) + (9 * \%\text{fat})\right].$$

2.3. Free Acidity

It was determined by potentiometric titration. 10g of honey was dissolved in 75mL of distilled water, and alcoholic solution of phenolphthalein was added. The solution was titrated with 0.1N NaOH.

The milliequivalents of acid per kg of honey were determined as 10 times the volume of NaOH used in titration.

2.4. Hydroxy Methyl Furfural (HMF)

5 g of honey were dissolved in 25 mL of distilled water. The absorbance was measured at 284 and 336 nm against a filtered solution treated with $NaHSO_3$.

$$\text{HMF } (\text{mg}/\text{kg}) = (\text{Abs}_{284} - * \text{Abs}_{336}) * 149.7 * 5 * (\text{D}/\text{W})$$

Where

- D = dilution factor and
- W=sample weight (g)

2.6. Determination of Sugar Content

- **Reducing Sugar**

Reduction of sugar was carried out using the Layne-Enyon method. 2.6g of honey was weighed and transferred to 500 mL volumetric flask. Five ml of standardized Fehling A and B solutions was transferred to a 250 ml Erlenmeyer, with 7 mL of water and 15 mL of honey solution. The Erlenmeyer was heated and 1 mL methylene blue 0.2 % was added. Titration was conducted by adding the diluted honey solution until the indicator was decolorized. Determining sucrose content was carried out by inversion, adding 10 mL of diluted HCl, 50 mL diluted honey solution and water to a 100 mL volumetric flask, heating in water bath, then cooling and diluting to mark. Finally the Lane-Enyon method was applied and sucrose content was obtained by difference.

- **Glucose content**

Honey sample was determined by enzymatic oxidation with glucose oxidase reagent (Randox Laboratories Ltd., UK). 20 µL of the sample or standard was allowed to react with 2.0 mL of the reagent, mixed well and incubated for 10 min at 37°C. The absorbance of the sample ($\mathbf{A}_{\text{sample}}$) and standard ($A_{\text{standard}}$) was read against a reagent blank within 60 min, and the glucose concentration was calculated as follows:

$$\text{glucose content}\left(\frac{\text{mg}}{\text{dL}}\right) = \left(\frac{\text{Asample}}{\text{Astandard}}\right) * \text{conc. standard.}$$

• **Fructose Content**

Fructose content was determined using the resorcinol reagent method [11].

2.7. Statistical Data Analysis

The Data obtained from the experimental laboratory analysis was analyzed and interpreted using SAS JMP™ Version 5 (*SAS Institute Inc., Cary, NC, USA*) software and Microsoft Excel. Means Comparisons were performed using one-way analysis of variance (ANOVA) for all the data. Comparisons for each data were carried out using Student's t test at α= 0.05, to see whether the actual difference in each value means was greater than the difference that would be significant.

3. Results and Discussion

3.1. Biochemical Composition

The Biochemical composition and energy values of all the honey samples collected from the study area were shown in Table 1 and Table 2.

3.1.1. Moisture Content

The overall mean moisture content of the honey samples collected in the present study was 18.76±1.09 g/100g ranged between 17.28 and 20.05 g/100g (Table 1 and Table 2) which was in an acceptable range of the world honey market. The overall mean value of all the honey samples (18.76 ± 1.09 g/100g) was greater than 16.00 ± 2.19 g/100g and 14.35 ± 1.50 g/100g which was reported by Buba *et al.,* [9] and Areda [12], respectively. According to the studies conducted by Awraris *et al.,* [13], on physico-chemical properties of honey produced in Masha, Gesha, and Sheko Districts in Southwestern Ethiopia, the overall mean moisture content was 22.86 ± 1.03 which is much greater than the result found in in the present study. Amabye T. and Mekonen F. [14] reported that the maximum moisture content of honey samples of Eastern Zone Areas in Tigray, Ethiopia was ranged 15.00± 2.74 to 17.33 ± 2.56 g/100g which were less than the results of this study.

Honey samples collected from traditional hives showed high moisture content than samples collected from modern hives (Table 1 and Table 2). The moisture content is the most essential quality component of honey, because the rate of fermentation, its shelf life span and processing characteristics are greatly determined by the amount of moisture content [15]. The different moisture content of honey depends on harvesting season, the degree of maturity that honey reached in the hive, type of hive used, environmental temperature and moisture content of original plant. Moisture content of honey can naturally be as low as 13% or as high as 23 % depending on the source of the honey, climatic conditions and other factors [16].

Table 1. Physicochemical properties (g/100g) and Energy Values (KJ/100g) (N=12, n=3)

Parameters	Mean ± SD					
	THM	MHM	THA	MHA	THC	MHC
Moisture	19.26±0.25^{b}	18±0.15^{c}	19.77±0.15^{a}	18.05±0.16^{c}	20.05±0.18^{a}	17.28±0.12^{d}
Ash	0.32±0.04^{cd}	0.38±0.03^{ab}	0.28±0.03^{d}	0.41±0.04^{a}	0.31±0.03^{cd}	0.34±0.02^{bc}
Protein	0.42±0.03^{d}	0.61±0.04^{a}	0.49±0.03^{c}	0.56±0.05^{ab}	0.52±0.03^{bc}	0.49±0.04^{c}
Fats	0.37±0.01^{ab}	0.39±0.03^{ab}	0.35±0.03^{ab}	0.41±0.04^{a}	0.33±0.03^{b}	0.35±0.06^{ab}
CHO	79.63±0.05^{c}	80.62±0.06^{b}	79.11±0.05^{c}	80.44±0.12^{b}	78.79±1.0^{7d}	81.54±0.07^{a}
Energy	1,354.30±2.35^{d}	1,374.81±1.09^{b}	1,346.01±0.85^{e}	1,371.71±1.01^{c}	1,340.40±0.92^{f}	1,386.69±0.61^{a}
pH	3.57±0.11	3.69±0.12	4.12±0.18	3.98±0.19	4.52±0.14	3.52±0.04
Acidity (meq/kg)	29.11±0.33	22.54±0.95	27.48±0.35	23.63±0.16	21.72±0.27	22.37±0.13
HMF (mg/kg)	6.86±0.27	9.34±0.56	13.27±0.67	12.93±0.84	9.18±0.39	7.84±0.41
WIM	0.062±0.02	0.071±0.07	0.065±0.05	0.048±0.02	0.068±0.01	0.075±0.05

*THM = traditional hive from Metti; MHM = modern hive from Metti; THA= traditional hive from Ashine; MHA = modern hive from Ashine; THC = traditional hive from Cabo; MHC = modern hive from Cabo; N = number of samples, n= number of replicate

*Values presented are mean ± SD of three determinations. Mean values with different superscript along a row are significantly different (P<0.05); SD = standard deviation; CHO = total carbohydrate, WIM = water insoluble matte

Table 2. Comparison of Range and average means with standards (N=12, n=3)

Parameters	Unit	Mean ± SD	Range (Min – Max)	Standards*	
				national	International
Moisture	(g/100g)	18.76±1.09	17.28 - 20.05	17.5 – 21	18 – 23
Ash	(g/100g)	0.34±0.05	0.28 - 0.41	<0.6	0.25 - 1.0
Protein	(g/100g)	0.52±0.07	0.42 - 0.61	----	-----
Fats	(g/100g)	0.37±0.03	0.33 - 0.41	----	----
CHO	(g/100g)	80.02±1.03	78.79 - 81.54	-----	-----
Energy	(KJ/100g)	1,362.32±18.16	1,340.40 - 1,386.69	---	---------
pH	---	3.9±0.38	3.52 - 4.52	----	3.2 - 4.5
Acidity	meq/kg)	24.48±3.06	21.72 - 29.11	<40	<50
HMF	(mg/kg)	9.91±2.64	6.86 -1 3.27	<40	<80
WIM	g/100g	0.06±0.01	0.048 - 0.075	0.5	-----

SD = standard deviation; N = number of samples, n= *number of replicate*

*Source: Quality and Standards Authority of Ethiopia [20].

High moisture content could accelerate crystallization in certain types of honey and increases its water activity of the honey to ferment and deteriorate its quality. Therefore, the higher moisture content of honey samples collected from the traditional hives in the study area could be due to higher humidity, inappropriate honey harvesting time before ripening and storage conditions [17].

Most of the samples showed low moisture contents (average value 18.76 ± 1.09g/100g), and only one sample was slightly exceeded (20.05 g/100g) the limit of 20.0 g/100 g established by international norms. A maximum value of 20.0 g/100g was established by the Codex Alimentarius Commission and EU Commission as the international standard for honey moisture contents.

3.1.2. Total Ash Content

The total ash content of honey samples in the present study was ranged between 0.28 g/100g to 0.41 g/100g with an overall mean value of 0.34±0.05g/100g which is within the acceptable range between 0.01-1.2% reported by the Quality and Standards Authority of Ethiopia (QSAE) and 0.6% maximum limit reported by the International Honey Commission [18], and European Honey Directive for mineral content of honey. Mineral content of honey is highly dependent on the soil type where the nectar producing plant is located, and the type of flower used by bees for nectar [19]. The mean results of the ash contents (0.34±0.05 g/100g) were greater than mean values of honey bee in Tigray region (0.20±0.12 g/100g) that was reported by Gebreegziabher *et al., [15]*, but similar to Guji Zone in Oromia region (0.34±0.38 g/100g) as reported by Areda, [12] and North-East Nigeria (0.42 ± 0.09 g/100g) research which was conducted by Buba *et al.,* [9]. According to the study results of Awraris *et al.,* [13], the overall mean value of honey samples Produced in Masha, Gesha, and Sheko Districts in Southwestern Ethiopia was 0.22 ± 0.16 g/100g which was less than 0.34±0.05 g/100g that was found in the present study.

3.1.3. Protein Content

The honey proteins are mainly in the form of enzymes. The honey bees add different enzymes during the process of honey ripening. The enzymes added include diastase (amylase), which digest starch to maltose and is relatively stable to heat and storage, and invertase (saccharase or α-glucosidase), which catalyses the conversion of sucrose to glucose and fructose.

Glucose oxidase and catalase are two other enzymes added by the honey bee, which regulate the production of hydrogen peroxide H_2O_2 that serves anti-bacterial factor in honey [9]. The protein mean value of all the honey samples from all locations in this study were significantly ($P<0.05$) different with overall mean value of 0.52 ± 0.06. The values obtained in this study were smaller than 0.67 ± 0.25 which was reported by Buba, [9] in North-East Nigeria and 0.57±.020 which was the research finding of Hira *et al.,* [22] from Pakistan. The protein content of the honey samples were within the ranges reported by Amabye T., Mekonen F. [14].

3.1.4. Fat Content

According to Singh and Kuar, [21], honey contains little or no fat, but the presence of free fatty acids like palmitic, oleic and linolenic acids have been reported in white clover honey. The fat contents of the honey samples investigated in this study fall within the range of 0.33 to 0.41 g/100 g with overall mean value of 0.37±0.03 g/100g indicating that the honey samples contain very little amount of fat.

According to Buba *et al.,* [9], the fat contents of the honey samples investigated in northern Nigeria were in the range of 0.1 to 0.5 g/100g with an average value of 0.29 ± 0.11g/100g which is smaller than the research findings of this research. The range and overall mean fat content of honey sample of the current study falls within the range of the research findings of Amabye T., Mekonen F. [14].

3.1.5. Carbohydrates

Carbohydrates are the main constituents of honey comprising about 95% of honey dry weight. The monosaccharides, fructose and glucose, are the main sugars found in honey; these hexoses are products of the hydrolysis of sucrose [23].

The honey samples in this research had a range value of 78.79 - 81.54g/100g with overall mean value of 80.02±1.03g/100g which is within the range of 77.60 – 86.20g/100g (mean value of 82.30 ± 2.03g/100g) that was reported by Buba *et al.,* [9]. According to the studies conducted by Amabye T., and Mekonen F. [14] on Phytochemical and Biochemical Composition of Honey samples in Eastern Zone, Tigray, Ethiopia the carbohydrate content was ranged from 82.10±4.31 to 83.00±1.31 g/100g which was slightly greater than the findings of this research.

3.1.6. Energy Value

Honey is primarily a high energy carbohydrate food and the honey sugars are easily digestible sugars similar to those found in many fruits. For this reason honey is regarded as a good food for both infants and adults [23]. The average of the honey samples from all locations ranged between 1,340.40 - 1386.69 KJ/100 g. honey samples found from modern hive in Cabo location had the largest energy value (1386.69 KJ/100 g) whereas the smallest energy (1386.69 KJ/100 g) was found from the traditional samples found from Cabo. According to Buba *et al.,* [9] in northern Nigeria, the average energy value of the honey samples was 1401.33 ± 33.71 which was greater than 1,362.32±18.16KJ/100g that was the average energy values found in the present study.

3.1.7. pH

In this research study the pH value of all the samples were ranged between 3.52 and 4.52 with an overall mean value of 3.9±0.38. The results were within the range that was reported by Tesfaye *et al.,* [24] who studied Physico-Chemical Properties of Honey Produced in Bale Natural Forest, Southeastern Ethiopia.

According to Gebreegziabher *et al.,* [15] who studied Physiochemical characteristics of honey obtained from traditional and modern hive production systems in Tigray region, northern Ethiopia, the overall mean value of hone was 4.1±0.2 which was greater than the value reported in this study.

3.1.8. Hydroxy Methyl Furfural (HMF)

HMF is defined as a breakdown product of fructose that is formed slowly and naturally during the storage of honey

and much more quickly when honey is heated [25], and widely recognized as an indicator of honey freshness [18]. The higher the HMF value, the lower the quality of the honey. The amount of HMF concentration increases with storage and prolonged heating of honey [17].

The overall average HMF value of the honey samples in this study was 9.91±2.64 mg/kg ranging between 6.86 and 1 3.27mg/kg (Table 1 and Table 2) which is acceptable range in the world honey market standard. Tesfaye *et al.,* [24] who studied Physico-Chemical Properties of Honey Produced in Bale Natural Forest, Southeastern Ethiopia reported that the overall mean value of HMF was 19.52 ± 9.4 mg/kg which was greater than 9.91±2.64 mg/kg that was found in this study. The HMF value of this research finding was less than the findings of Awraris *et al*., [13] who studied on Physico-Chemical Properties of Honey Produced in Masha, Gesha, and Sheko Districts in Southwestern of Ethiopia.

3.1.9. Free Acidity

The average acidity of all the samples was ranged between 21.72 and 29.11 meq/kg with an overall mean value of 24.48±3.06 meq/kg. All of the samples met the requirement of national and international standards [20], which indicates the freshness of honey samples and absence of unwanted honey fermentation. Differences in honey acidity could be caused by differences in geographical condition, harvesting procedure and storage conditions [26], which could be the reason in the case of the results in this study.

According to the findings of Awraris *et al.,* [13] who studied Physico-Chemical Properties of Honey Produced in Masha, Gesha, and Sheko Districts in Southwestern Ethiopia, the overall mean value of acidity was 28.32 ± 14.14 meq/kg which was greater than the finding of the present study.

3.2. Sugar Contents

Carbohydrates are the major constituents of honey, corresponding to 95–99% of the dry matter. These sugars are composed mainly of fructose, glucose and sucrose [26,27]. There was significant difference in fructose content among all honey samples except for the samples obtained from Metti town (traditional hive) and Ashine (traditional hive). The highest fructose content was recorded for the honey samples obtained from modern hives collected from Cabo (39.29±0.09 g/100g) and the lowest fructose content was 37.77±0.17 that was collected from traditional hive in Metti town (Table 3).

All the honey samples had an overall mean value of 38.64±0.61 fructose as indicated in Table 3. Honey samples collected from Cabo (modern hive) got the highest glucose content (37.6±0.31). The honey samples from all locations ranged between 31.55 and 37.60 g/100g with mean value of 36.37±2.14g/100g (Table 4). The fructose contents (38.64±0.61g/100g) of the samples were significantly (P<0.001) higher than the glucose contents (36.37±2.14g/100g).

The fructose/glucose ratio and glucose/water ratio were within the range of 1.05 to 1.20 and 11.62 and 2.18 with mean values of 1.06±0.06 and 2.01 1.94±0.21, respectively.

The Sucrose contents of all the samples were significantly different (P<0.001). Honey samples obtained from Ashine (traditional hive) had the highest sucrose content (2.98±0.05g/100g) and honey samples collected from Metti town (modern hive) had got 2.34±0.11g/100g. The overall sucrose mean value of all the samples was 2.66±0.23g/100g with a range from 2.34 - 2.98g/100g. The sum of fructose and glucose (fructose + glucose) contents ranged between 69.32 and 76.89g/100 g with an average of 75.01±2.69g/100 g while the reducing sugar contents varied between 72.46 and 76.86g/100 g with an average of 74.19±1.88g/100 g.

The average mean value of all the honey samples (38.64±0.61g/100g) in this study was similar to the average mean value of honey samples (38.94 ± 0.89g/100g) from North-East Nigeria which was reported by Buba *et al.,* [9]. The overall mean value of glucose (36.37±2.14g/100g) and sucrose (2.66±0.23g/100g) in this study were greater than 31.65 ± 2.27g/100g (for glucose) and 1.84 ± 0.7g/100g (for sucrose) that was reported by Buba et al., [9].

According to the research findings of Areda [12] which was conducted in Guji Zone, the sucrose and reducing sugar content of honey samples were 2.22 ± 0.9 and 60.84 ± 0.62g/100g sample which were smaller than 2.66±0.23g/100g (sucrose) and 74.19±1.88g/100g (reducing sugar) of this study. In the study of Quality of Honey in Argentina Cantarelli *et al.,* [28] found 68.08 g/100g (Reducing sugar) and 4.05g/100g (sucrose) which is not in good agreement to the work findings of this research study.

Table 3. Sugar Content (g/100g) of Honey Samples (N=12, n=3)

Parameters	Mean±SD					
	THM	MHM	THA	MHA	THC	MHC
Fructose	37.77±0.17^{d}	38.15±0.12b^{c}	37.98±0.08^{cd}	39.05±0.22^{a}	38.25±0.17^{b}	39.29±0.09^{a}
Glucose	31.55±0.08^{d}	34.61±0.13^{c}	35.47±0.26^{b}	37.05±0.26^{a}	35.35±0.27^{b}	37.6±0.31^{a}
Fructose + Glucose	69.32±0.21^{a}	72.76±0.15^{d}	73.45±0.30^{c}	76.1±0.19^{b}	73.6±0.32^{c}	76.89±0.32^{a}
Reducing sugar	72.46±0.30^{c}	73.14±0.19^{bc}	72.71±0.30^{c}	76.23±0.61^{a}	73.78±0.43^{b}	76.86±0.39^{a}
Sucrose	2.74±0.09^{ab}	2.34±0.11^{d}	2.98±0.05^{a}	2.67±0.06^{b}	2.59±0.11^{bc}	2.41±0.18^{cd}
(Fructose/Glucose) ratio	1.20±0.04^{a}	1.10±0.08^{b}	1.07±0.03^{b}	1.05±0.03^{b}	1.08±0.04^{b}	1.05±0.05^{b}
(Glucose/water) Ratio	1.62±0.06^{d}	1.92±0.08^{bc}	1.79±0.13^{cd}	2.04±0.16^{ab}	1.76±0.16^{cd}	2.18±0.17^{a}

*THM = traditional hive from Metti; MHM = modern hive from Metti; THA= traditional hive from Ashine; MHA = modern hive from Ashine; THC = traditional hive from Cabo; MHC = modern hive from Cabo; N = number of samples, n= number of replicate

*Values presented are mean ± SD of three determinations. Mean values with different superscript along a row are significantly different (P<0.05); SD = standard deviation.

Table 4. Comparison of overall Mean and ranges with standards (N=12, n= 3)

Parameters	Unit	Mean ± SD	Range	Standards*	
			(Min – Max)	National	International
Fructose	g/100g	38.64±0.61	37.77 - 39.29	-----	------
Glucose	g/100g	36.37±2.14	31.55 - 37.6	-----	-----
Fructose + Glucose	g/100g	75.01±2.69	69.32 - 76.89	>60.0	60 - 70
Reducing sugar	g/100g	74.19±1.88	72.46 - 76.86	>65	60 – 70
Sucrose	g/100g	2.66±0.23	2.34 - 2.98	<5.0	<10
(Fructose/Glucose) ratio	__	1.06±0.06	1.05 - 1.20	__	___
(Glucose/water) Ratio	__	1.94±0.21	1.62 - 2.18	__	___

SD = standard deviation; N = number of samples, n= number of replicate
*Source: Quality and Standards Authority of Ethiopia [20].

4. Conclusion

The results of the present study indicated that almost all of the honey quality parameters (moisture, protein, fat, ash, CHO and energy content; reducing sugars, sucrose, fructose and glucose contents) analyzed from all locations of the study area revealed that, all the physicochemical parameters lie with-in limits of national and international standards set by Ethiopian Conformity Assessment Enterprise for analyzing all honey samples (ECAE), Codex Alimentarius Commission and EU Council. There were significant differences ($p<0.01$) between hive types for most of the quality parameters. Honey producers should be well trained on the quality standards and the ways how to improve and assure the quality parameters.

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Statement of Competing Interest

The authors have no competing interest.

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