Physiochemical characteristics of honey obtained from traditional and modern hive production systems in Tigray region, northern Ethiopia

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ABSTRACT
This study was conducted with the objective to determine the physiochemical characteristics of Tigray honey (n= 38) and to compare the quality honey of Tigray with those described in the Quality Standard Authority of Ethiopia, Codex Standard and European Honey Directive. Honey samples were collected from traditional hives (n =19) and modern hives (n=19) production systems. Physiochemical characteristics evaluated were: color, moisture, reducing sugar, sucrose, acidity, hydroxymethylfurfural (HMF), mineral content, pH, water insoluble solids and specific gravity. All parameters were analyzed following the techniques proposed by the Codex Standard for Honey. A statistical analysis using GMP5 soft ware was utilized to determine the differences in quality between honey from traditional and modern hive production systems. The average content of mineral, moisture, acidity, invert sugar, pH, sucrose, specific gravity and water insoluble solids in traditional hive honey samples were 22%, 18.25%, 29.89 meq kg\(^{-1}\), 70.95%, 4.13, 2.37%, 1.41 and 0.07%, respectively. On the other hand, the corresponding values for honey samples from modern hive production system were 0.18%, 18.60%, 29.12meqkg\(^{-1}\), 71.42%, 4.04, 2.71%, 1.40, and 0.03%, respectively. Consequently, no significant differences (p> 0.05) were found in all the quality parameters except for water insoluble solids. The mean value for HMF was 11.18 mgkg\(^{-1}\) for the processed honey and the color of the honey varied from white to amber with light amber (55.26%) being the predominant color of the honey in Tigray. All the honey samples met the Codex Standard, QSAE and EU physiochemical quality Standard limits for honey except water insoluble solids in traditional hive production system. Water insoluble solids were found in 26.32% of the samples examined which will have to be improved through intensive training and experience sharing.

Key word: Honey, Quality, Physiochemical characteristics, Tigray.

1. INTRODUCTION
Honey is the natural sweet substance produced by certain species of bees, from the nectar of plants, from secretion of living part of plant. The bee collect this sugary substance of their own, deposit, reduce the water content, store and leave it in honey combs or honey pots to ripen and mature for their own consumption (Codex Alimentarius, 1989). Honey is composed primarily of the sugars, glucose and fructose while its third greatest component is water. It also contains numerous other types of sugars, acids and minerals. The sugars found in honey can be classified as monosaccharide (glucose, fructose), sucrose and oligosaccharides (White, 1980).
Ethiopia has about 10 million bee colonies which make the nation with the highest bee density country in Africa. Ethiopia is a major producer of beeswax, only surpassed by Mexico and China, with more than 2000 tones per annum and is one of the five biggest wax exporters to the world market (Ayalew, 2001). Total honey production in the country is estimated to be more than 43,000 tones per year (C.S.A., 1995). With this, Ethiopia is the largest honey producing country in Africa and stands in 10\textsuperscript{th} place (Rinhard and Admasu, 1994). Honey is the most important primary product of beekeeping both from a quantitative and economic point of view. About 80\% of the total honey produced goes into ”tej” making, a local beverage, also known honey wine or honey beer, that is made in the homes of farmers or in urban area and only very small quantity goes for export mainly to Saudi Arabia and Yemen (Mammo, 1976; ARSD, 2000).

Beekeeping in Tigray is mostly traditional and has long years of experience which is parallel to the history of agriculture. In most cases, beekeeping has remained traditional and never rewarding. Because of this, the yield of honey and other hive products have been constantly the same over the past years. It did not exceed 45 kg per modern hive per year and not more than 7 kg from traditional hive per year (REST, 2004).

The honey bee (apis melifera) is encountered in all agro-ecological zones of Tigray. Farmers recognized three types of bees: In the highlands, the black bee and the red striped black bee whereas in the lowlands the yellow colored bees are found. The highland bees are docile and less productive while the lowland bee is aggressive and more productive (BoANRD, 1997). There are about 201,000 bee colonies and 2,300 tones of honey production in a year (BoANRD, 2008).

The Regional State of Tigray is running a powerful development program in beekeeping and its products so that poor crop production zones of the region are involved mainly in beekeeping interventions to maintain food security and to empowering higher production at first to satisfy family needs as well as to meet the requirements of both domestic and international market.

Efforts have been increasingly improved with regard to beekeeping systems in our region by using modern beehives intensively, increasing the rate of swarm population, maintenance and cultivation of honey bee’s floras, providing of ancillary equipments, extension programs, and expanding apiary sites; the honey production will increase substantially in quantity. As Tigray produces thousands of tones of honey, government institutions, private processing plants that can process, pack and seal to domestic and international market are emerging (TFAP, 1996).
Honey shows great variety in color, flavor and aroma, because different plants contribute their own characteristic constituents. Moreover, honey is produced under many different climatic conditions. However, the main constituents in all honeys are the same (Crane, 1975). International honey standards are specified (QSAE, 2005), European Honey Directive (1974) and in the Codex Alimentarius Standard for honey (2002, 1993).

Honey is a semi liquid product, which contains a complex mixture of carbohydrates, mainly glucose and fructose. Other sugars are present as traces, depending on floral origin. Moreover, organic acids, lactones, amino acids, minerals, vitamins, enzymes, pollen, wax, and pigments are present which are important properties used to determine quality of the honey (Crane, 1990, 1975; Yaniv and Rudich, 1996; Silici, 2002). Honey having high water content is more likely to ferment. The mineral content indicates botanical origin of honey; the blossom honey has lower mineral content than dew honey (Vorwohl et al., 1989).

Honey storage has a remarkable influence on increase in invert sugar content. A prolonged storage period of honey causes invert sugar to increase. Honey fermentation leads to an increased acidity and because of this a maximum acidity value has proven to be useful. Less the HMF in honey, better is the quality of honey. The effects of HMF in honey depend on heat process, post harvesting and storage time (White et al., 1961).

Although thousands of tones of honey are produced per annum in the region, the physiochemical parameters of Tigray honey have never been studied. One of the means to get information about the quality is physical analysis of honey. In this study, the physiochemical parameters namely, mineral content(%), moisture content(%), acidity (meqkg⁻¹), hydroxymethylfurfural (mgkg⁻¹), invert sugar(%), sucrose(%) , pH, specific gravity, and color for honey samples produced in Tigray were determined and compared against QSAE (2005), Codex (2001, 1993) and EU (1974) standard limits.

2. MATERIALS AND METHODS

2.1. Description of study area
This study was conducted in Tigray, a northern state of Ethiopia. It is bordered by the Eritrea in the north, the Sudan on the West and the Amhara and Afar Regional States of FDRE in the South and East, respectively. The land surface area is approximately 80,230 square kilometers (TFAP, 1996). Tigray falls into four main agro-ecological zones: Arid in the extreme eastern part, semi
arid in the north western part, tepid to cool moist in the center part of the region in which above 50% of the region is found, and hot to warm sub moist zone in the south-western part and along the western part of the eastern escarpment (TFAP, 1996).

Hot to warm arid lowland: This agro-ecological zone is found in the eastern part of the region below 1400 meters above sea level (masl). The mean annual rainfall ranges from 200-600 mm and mean annual temperature between 21-28°C.

Hot to warm semi-arid lowland and plateau remnants: This zone is found on the extreme western part of Tigray between 500 and 1600 masl. The mean annual rainfall ranges 300 – 800mm and the mean annual temperature varies between 21 and 28°C.

Hot to warm sub-moist lowland and plateau remnants: This agro-ecological zone is found in the south-western part of the region and along the western part of the escarpment. The altitude ranges from 400-2000 masl and the mean annual rainfall and the mean temperature ranges 400-200mm and 21-28°C, respectively.

Tepid to cool sub-moist low to high altitude: About 50% of the region is under this agro ecological zone. It occupies the central and north western part of the region with altitude varying from 1000 to 3000 masl and mean annual rainfall varies from 500 to 1600mm and the mean annual temperature ranges from 21-28°C in the lower altitudes and from 11-12°C in the higher elevated areas (BOANRD,1997). No periodic and comprehensive work has been done on the relative humidity for the region but according to the report by the Regional State of Tigray (2007), the relative humidity for ten woredas’ was 56%.

2.2. Method of sampling and analysis

The sample preparation procedure was according to the Codex Alimentarius (1993). The honey was collected randomly from different location of the Tigray Region and it was crystallized. The honey was brought to the processing plant in a closed plastic container. Each container contained about 22kg of honey was placed in a large thermostatically controlled water bath without submerging. It was heated for 30 minutes at 60°C until it was completely liquefied. All honey poured into a large drum having 600kg capacity. After filling of the large drum, the honey was allowed to pass through 80 micron filters. This was done with the purpose to sieve wax, honey and foreign materials such as sticks, bees and particles of the comb. It was then mixed thoroughly by a mixer and again filtered and homogenized. A representative sample usually 5-20 g of honey was taken depending on the method of analysis required. The samples were collected
from each modern and traditional hive separately and results of the analysis were recorded. Raw honey intended to measure HMF was not heated. However, the honey was found to be crystallized and it was difficult to obtain raw honey in liquid form. Nine samples were taken for measuring HMF value from the processed honey to compare the effect of processing on raw honey.

Acidity, mineral content, water insoluble solids, specific gravity, reducing sugar, sucrose content, pH and hydroxymethylfurfural were analyzed according to the methods of Codex Alimentarius Standard (1993). Moisture content was determined using pocket refractometer.

2.3. Data Collection and analysis

In this study, considerable efforts are made to obtain honey samples from all over areas of the region and some general information about the composition of honey was collected. A total of 38 samples representing 19 from traditional and 19 from modern hives gathered from various locations of the Tigray region. The area of collection was not repeated and the samples of honey were collected from apiary site and farmers. The honey was collected in a tightly closed plastic container and made free from foreign matters like dead bees, brood, insects, and particles of the comb. Strict detection methods of adulteration at field level were applied during the collection of honey. The total honey from which samples were taken once after homogenized was about 22,800 kg. The areas from where the honey was collected are given in Table 1. The data were analyzed by the statistical method at 95% Confidence interval using GMP5. Sucrose content, mineral content, and water insoluble solids for both traditional and modern production systems were compared using Pi-chart.

Table 1. Areas of sample collection.

<table>
<thead>
<tr>
<th>Axum</th>
<th>Wukro</th>
<th>Abergellie</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adigrat</td>
<td>Hawzien</td>
<td>Haik meshale</td>
</tr>
<tr>
<td>Enderta</td>
<td>Kukuftu I</td>
<td>Hashenge</td>
</tr>
<tr>
<td>Offula</td>
<td>Kukuftu II</td>
<td>Golomekeda</td>
</tr>
<tr>
<td>Aheferom</td>
<td>Della</td>
<td>Bora</td>
</tr>
<tr>
<td>D/tembien</td>
<td>Lemlem</td>
<td>Shire</td>
</tr>
<tr>
<td>K/tembien</td>
<td>Alagae</td>
<td>S/samre</td>
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<tr>
<td>Lalay maychew</td>
<td>G/afeshum</td>
<td>G/mohoni</td>
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<tr>
<td>Adwa</td>
<td>T/maychew</td>
<td>Mekelle</td>
</tr>
</tbody>
</table>
3. RESULTS
The color of honey was measured in millimeters on a Pfund scale according to the U.S. department of agriculture classifications (White, 1975b; Crane, 1980).

Table 2. Classification of honey.

<table>
<thead>
<tr>
<th>USDA color standard</th>
<th>Pfund scale(mm)</th>
<th>% (n/Nx100)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water White</td>
<td>0 to 8</td>
<td>not found</td>
</tr>
<tr>
<td>Extra white</td>
<td>&gt;8 to 17</td>
<td>not found</td>
</tr>
<tr>
<td>White (n-3)</td>
<td>&gt;17 to 34</td>
<td>7.89</td>
</tr>
<tr>
<td>Extra light amber (n-10)</td>
<td>&gt;34 to 50</td>
<td>26.32</td>
</tr>
<tr>
<td>Light amber (n-21)</td>
<td>&gt;50 to 85</td>
<td>55.26</td>
</tr>
<tr>
<td>Amber (n-4)</td>
<td>&gt;85 to 114</td>
<td>10.53</td>
</tr>
<tr>
<td>Dark amber</td>
<td>&gt;114</td>
<td>not found</td>
</tr>
</tbody>
</table>

Figure 1. Traditional hives (pi-Chart).

Figure 2. Modern hives (pi-Chart).
Table 3. Biochemical composition of honey samples and statistical results (Values within a column with different superscripts (a,b) were significantly different (p<0.05)).

<table>
<thead>
<tr>
<th>Production system</th>
<th>Acidity (meq/kg&lt;sup&gt;-1&lt;/sup&gt;)</th>
<th>Mineral content (%)</th>
<th>Water insoluble solids (%)</th>
<th>Moisture content (%)</th>
<th>Specific gravity (number)</th>
<th>Total reducing sugar (%)</th>
<th>Sucrose content (%)</th>
<th>Sugar (%)</th>
<th>Sugar:water (%)</th>
<th>Undetermined (%)</th>
<th>pH (number)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samples (N =38)</td>
<td>Range</td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>18-43</td>
<td>0.02-0.44</td>
<td>0.005-0.22</td>
<td>17-21</td>
<td>1.39-1.42</td>
<td>67.7-80</td>
<td>1.27-4.24</td>
<td>70.19-81.61</td>
<td>3.35-4.53</td>
<td>0.096-12.59</td>
<td>3.24-4.42</td>
</tr>
<tr>
<td></td>
<td>29.5±5</td>
<td>0.20±0.1</td>
<td>0.05±0.1</td>
<td>18.4±0.8</td>
<td>1.4± 0.01</td>
<td>71.2±2.5</td>
<td>2.54±0.7</td>
<td>73.7±3</td>
<td>4±0.24</td>
<td>7.6±2.7</td>
<td>4.1±0.2</td>
</tr>
<tr>
<td>Traditional</td>
<td>29.89±5</td>
<td>0.22±0.1</td>
<td>0.07±0.1&lt;sup&gt;b&lt;/sup&gt;</td>
<td>18.25±0.6</td>
<td>1.41±0.01</td>
<td>70.95±2</td>
<td>2.37±0.6</td>
<td>73.32±2</td>
<td>4.02±0.2</td>
<td>8.13±2</td>
<td>4.13±0.2</td>
</tr>
<tr>
<td>hive (n=19)</td>
<td>29.12±5</td>
<td>0.18±0.1</td>
<td>0.03±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>18.60±0.9</td>
<td>1.40±0.01</td>
<td>71.42±3.1</td>
<td>2.71±1.8</td>
<td>74.14±3.3</td>
<td>3.99±0.3</td>
<td>7.03±3.3</td>
<td>4.04±0.2</td>
</tr>
<tr>
<td>Modern</td>
<td>29.5±5</td>
<td>0.3485</td>
<td>0.0080</td>
<td>0.2004</td>
<td>0.06</td>
<td>0.5844</td>
<td>0.1298</td>
<td>0.3635</td>
<td>0.6933</td>
<td>0.223</td>
<td>0.1949</td>
</tr>
<tr>
<td>hive (n=19)</td>
<td></td>
<td></td>
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<tr>
<td>P values</td>
<td>0.6374</td>
<td></td>
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5. DISCUSSION

The most important aspect of honey color lies in its value for marketing determination of its end users. Darker honeys are more often for industrial use such as waffles, ice cream, barbecue sauces and hearty backed goods, while light color honey is marketed for direct consumption. Consumers’ preferences are determined by the color of honey and thus, next to general quality determinations, color is the single most important factor determining import and whole sale prices (White, 1975b; Crane, 1980).

In the present study, the color of the honey varied from white to amber. Of all the samples 55.26% light amber, 26.32% extra light amber, 10.53% amber and 7.89% white were measured (Table 2). It was observed that 89.47% honey was predominantly of light color in Tigray and this could be marketed for direct consumption. White et al. (1962) observed that variations are almost entirely due to the plant source of the honey, although climate may modify the color somewhat through the darkening action of heat. The average specific gravity of honey was 1.41 which was highly viscous. Because of high viscosity, its flow was reduced upon pumping, settling, filtration, mixing and bottling.

The average composition of reducing sugar in both traditional and modern production systems was 70.95± 1.99 and 71.42 ± 3.09, respectively, which is above 65% as recommended by QSAE (2005), Codex (1993) and EU (1974). There was no significance difference (p>0.05) between the traditional and modern production systems (Table. 3). A similar report was reported in reducing sugar (Nuru, 1996) in Ethiopian honey which accounted about 70% on an average. Silici (2002)
and White et al. (1962) also reported their observation close to this result, in which the average for reducing sugar were 67.60 % and 69.47 % in Turkey and U. S. honey, respectively. Earlier studies in Pakistan (Latif et al., 1956) also reported similar results with 71 to 76.9% reducing sugars. Consequently, there was no substantial difference in percentage of reducing sugar of Tigray honey as compared to the literature. Maurizio (1959) reported that sugar spectrum of honey depends upon the sugar present in the nectar and enzymes present in the bee and the nectar.

Generally, the Tigray honey was essentially with highly concentrated water solution of two sugars, glucose and fructose. Levulose and dextrose were the simple sugars which make higher percentage of honey. As shown in Table 3, the average value for total reducing sugar was 71.18%, while it varied among samples between 67.7 and 80.0%. The high percentage of the simple sugars could be responsible for much of the physical and nutritional characteristics of honey. The major constituents, close to 99.73%, of the honey were moisture content, reducing sugar, undetermined value and sucrose (Fig. 1). While the remaining 0.27% were mineral content and water in soluble solids. The average value for sugars (reducing sugar and sucrose content) was 73.73%. This was also four times as much as moisture content which indicated that the honey was an over saturated sugar solution. This was evident in the crystallization of the honey a few days or weeks post harvest or process. Super-saturation of honey is the cause for its crystallization. This super saturation occurs as a result of the presence of so much sugar in honey, usually more than 70%, relative to the water content, which is less than 20%.

Glucose tends to precipitate and the solution changes to a more stable saturated state (Crane, 1980). About 81.57% of the honey was dry matter. The largest portion of the dry matter was reducing sugar (about 87.26%). Doner (1977) also reported that glucose and fructose are the main sugars in honey, which accounted about 85% of solids in honey which is inline with the present study. Honey contains also disaccharides, oligosaccharides and acids which is 7% of its composition (NHB, 1995). In the present study, these sugar were not analyzed. Unanalyzed sugars accounted 7.58% (Fig. 1) for both the production systems with no significance difference (p>0.0.05).

The mean non-reducing, sucrose, content of Tigray honey was found inline with the QSAE (2005), Codex (1993) and EU (1974). The average values for traditional and modern production
systems were 2.37±0.55 and 2.71±1.79, respectively, and there was no significance difference (p>0.05).

Moisture content of honey is practically the most important quality parameter, because the rate of fermentation and its shelf life span are greatly determined by the amount of moisture contents. Honeys only with less than 18% water can be stored with little or no risk of fermentation. However, honey with over 19% water will ferment (White et al., 1962)

Moisture values for all the samples were less or equal to the maximum limit (21%) established by the QSAE (2005), Codex (1993) and EU (1974). The average moisture content was 18.43% for both production systems with no significance difference (p>0.05). This was lower as compared to the standard. As a result, Tigray honey must be less susceptible to fermentation irrespective of the yeast count. HBRTC (1980) reported lower moisture content of (19.40%) Tigray honey which is slightly higher than the present study indicating better quality of the honey. Zeleke (1989) reported a mean of moisture content of 20.5% that ranged from 19.1 to 21.5%, in his study samples in Hararghe Administrative Region. This moisture content is clearly higher than the value in the present study which follows that Tigray honey is qualitatively better than honey from Hararghe region. This is mainly due to the prevailing atmospheric humidity both pre- and post removal of honey from the hives as the climatic condition of Tigray is characterized by low humidity of air. In more humid climates, even sealed cells can contain honey with more than 24% or even 28% moisture content (Krell and Crane, 1990). According to the United States Standard for Grades of Extracted Honey (1985), 78.95% of the samples analyzed had moisture values ranged from 17-18.6% which qualified grade “A” and the remaining 21.05% were ranged from grater 18.6 – 21% and placed in grade “B”, “C” and “D”, respectively.

This study revealed no significant difference in pH between the traditional (4.13±0.15) and modern (4.04 ±0.21) production systems. The honey pH values varied from 3.24 to 4.42 and the mean value for all the samples (both traditional and modern systems) was 4.08±0.18. This result is also within the quality regulation level proposed by Codex (1993) and EU (1974) in which the honey pH value varied from 3.4 to 6.1 with a mean value of 3.9. This indicates that the honey was fairly acidic and this could be in part responsible for the excellent stability of honey against microorganisms and natural flavor (Doner, 1977; White, 1975a). The variations in pH might
mainly be resulted due to different acids found in different floral types. Hussain (1989) observed a significant difference in honey pH of different floral types. The average total acidity of all the samples (29.5± 5.12) in the present study was recorded within the prescribed limits of 40 meqkg⁻¹ as proposed by QSAE (2005), Codex (1993) and EU (1974). However, the range of the samples was from 18-43 meq kg⁻¹ which showed great variations which might be a consequence of the source of nectar. White et al. (1962) reported a similar result in which the total acidity was 29.12 meq kg⁻¹ in 490 samples of honey.

The average mineral content was recorded 0.20±0.12 for both production systems and this indicated good quality of honey as compared to the standard. Although there was no significant difference in mineral content between traditional and modern production systems, the traditional hives tend to have higher mean value (0.22 ±0.11) than the modern hives (0.18 ± 0.13). The mineral content of all the samples analyzed varied from 0.02 to 0.44% and this is within the quality regulation level (≤0.6%) proposed by QSAE (2005), Codex (1993) and EU (1974). At national level, a mineral content of honey ranges from 0.01-1.16% with mean a value of 0.23% (Nuru, 1996). The average amount of mineral in honey is about 0.17% with a variation of range of 0.02 to 1%. The marked difference in ash content could be attributed to the soil where the honey bee plants grow (White et al., 1962).

The average value of water insoluble solids of all the samples (0.05±0.04) was recorded within the prescribed limits of ≤0.1% as recommended by QSAE (2005), Codex (1993) and EU (1974). The lower percentage of water insoluble solids of Tigray honey as compared to the standard is another additional quality of the honey. The mean value of water insoluble solids for traditional hives (0.07±0.06) was recorded twice as modern hives (0.03±0.02). Consequently significant difference (p<0.05) was recorded between traditional and modern production systems. The water insoluble solids content in modern production system for all the samples were ranged from 0.005-0.067% which was within the quality regulation level (≤ 0.1%) where as for traditional production systems, the range varied from 0.005-0.22% and 26.32% of the samples had higher than the permitted maxima in the quality standard. The higher percentage of water insoluble solids in traditional hives could be directly attributed to the poor honey handling and a sign of improper handling during harvest. The common method of honey harvesting in Tigray Region is pressed method which means the wax, honey, pollen and with some other impurities like dead
bees, larvae and propolis were all removed in one mass and crushed to gather which could have contributed to the high content of water insoluble solids.

The most commonly monitored parameters for determining honey freshness include hydroxymethylfurfural, HMF level and diastase and invertase activity (Oddo et al., 1999; Bogdnov and Martin, 2002). The HMF level ranged from 2.9-26 mg kg\(^{-1}\) with a mean value of 11.18 mg kg\(^{-1}\) for the processed honey. This was also within the limit (40 mg kg\(^{-1}\)) prescribed by QSAE (2005), Codex (1993) and EU (1974). This can in fact be regarded as a good quality of honey. However, the increment of this value could be due to the heat treatment during the processing of honey. Honey does not naturally contain HMF, but it can be derived from the loss of two molecules of fructose. Its presence indicates long storing periods and heating (White, 1992). It can be concluded that this study had shown that honey produced from traditional and modern production systems met the physiochemical quality requirements established in International market. However, water insoluble solid was higher in traditional hives which decreases honey quality and this may influence its market value. Although honey produced in Tigray Region had the physiochemical parameters that are desirable in the International market, harvesting, bottling, and storing honey in this Region has to be improved through training and using available technological inputs. It is strongly recommended that training on apiculture farming, especially, harvesting, handling and storage will substantially improve the quality of honey which is vital income generating means for millions of families in Tigray.

5. REFERENCES
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