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Physicochemical Characterization of Honey from Debre-Nazret Kebelle of Tigray Region, Ethiopia

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Abstract: The study was conducted in Debre-Nazret kebelle of Tigray region from August 27, 2012 to March 30, 2013. It was aimed at characterizing as the honey quality parameters and documenting the honey bee flora of the study area. Honey quality was checked from 21 samples and analyzed for eight honey quality parameters /moisture, ash, acidity, pH, hydromethyfurfural (HMF), diastase, electrical conductivity (E.C) and Water Insoluble Solids (WIS)/. Quantitative results for Ash, acidity, pH, HMF, electrical conductivity and moisture content was compared with the world set standard. All except for WIS samples were within limits of Codex Alimentarius Commission, world, FAO/WHO standards and Ethiopian Standard Authority /moisture (86 %), ash (100 %), acidity (100 %), pH (94.4%), HMF (100 %), diastase (100 %) and electrical conductivity (100 %)/ of the samples were within the acceptable range of the world set standard for honey quality. In this study a total of 32 honey bee flora plants species grouped in to 25 families was documented. The honey bee keeping constraints and honey production status of the study area was assessed from the randomly selected informants of the sampled sites. There is a need to conduct a research on the honey quality by using a parameter of pollen analysis from the sampled study sites.

Key words: Physicochemical · Quality · Characterization · Debre-Nazret And Tigray

INTRODUCTION

Honey is natural sweet substance produced by *Apis mellifera* bees from the nectar of plants or from secretions of living parts of plants or excretions of plant-sucking insects on the living parts of plants which the bees collect and transform by combining it with specific substances of their own, deposit, dehydrate, store and leave in honeycombs to ripen and mature [1]. It is a complex mixture and presents very great variations in composition and characteristics based on geographical and floral types or the nectar foraged by bees [2]. Honey characterization is based on the determination of its chemical, physical or biological Properties [3].

Adequate nectar and pollen resources are critical in maintaining honey bee health [4]. A deficiency in quantity and quality of pollen and nectar can lead to demographic decrease of bee colonies leading to low colony populations, which collect pollen and nectar [5]. Moreover, recent work shows the importance of pollen quality and diversity on the immune response of honey bees [6]. The role of these two nutrients is so critical that the beekeeper often must provide supplements as sugar syrup or pollen supplement to prevent nutritional deficiency and colony failure [6]. Production of honey and other products depend on availability of floral resources (Bee forage) and is a very important field for beekeepers. Most of the methods of obtaining information about plants used in an area are based on direct field observations of foraging honeybees. Bee colony performance as well as production of honey, wax and other hive products depends on bee foraged plants from which honey bees obtain nectar and pollen as main food. These food sources provide the nutritional requirements of the bee colonies: nectar as sources of honey provides heat and energy for honey bees and pollen provides protein, vitamins, fatty substances and other nutrients [7]. The composition of honey depends on the plant species visited by the honeybees and the environmental processing and storage conditions [8, 9]. Honey contains, water and certain minor constituents such as proteins, enzymes, amino and organic acids, lipids, vitamins,

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volatile chemicals, phenolic acids, flavonoids and carotenoid-like substances and minerals. The different quality criteria of honey and the methods used for the determination were reported by the International Honey Commission (IHC) [10]. The contents of these components in honey are the most important quality criteria of honey and show some important deterministic quality properties of the honey [11]. Careless handling of honey can reduce its quality. Amongst the factors that most influence quality is high temperature, length of storage and moisture content greater than 21%. They lead to fermentation, high levels of hydroxymethylfurfural (HMF), loss of enzymatic activity, changes from flavor, darkening and microbial growth [12]. Detection of honey adulteration is required to ensure quality and human safety. The HMF marker is an organic compound derived from dehydration of certain sugars [13, 14]. Honey contamination by heavy metals (Especially Pd, Cd and Cu) and trace and/ or toxic (Zn) are produced by location of colonies of industrial zones or other areas with considerable air pollution with toxic chemicals. Agricultural use of toxic chemicals is another common and very likely source of contamination; further contamination may results from dirty water source and non-floral sugar source [15]. Containers previously used for toxic chemicals, oil or petroleum products or vessels doesn't manufacture for food preservation should never be used for storing honey.

Like all other Ethiopian regions, Tigray region has various agro ecological zones that are suitable for the growth of different bee flora and development of apiculture. However, the loss of endangered plant species or deforestation in Tigray region, has undoubtedly affected the bee forage availability, diversity and flowering pattern and finally honeybee products and productivity in the region [16]. In order to have a beneficial effect honey must be free of any contaminating agents; any contaminants such as heavy metals present in honey above the admitted levels by pollution standards are threats to human health. The current international honey markets trend, regarding quality is more demanding. Therefore, it is necessary to promote all feasible activities in order to produce residue free honey [17]. The detection of compounds, not previously found in honey imports by the European Union has generated increased control of residues that might be present in honeys [18]. Therefore this study was aimed at 1. Evaluating honey quality and quantity status of the area 2. Identifying major constraints on honey bee keeping in Debre-Nazret and 3. Compare the physicochemical characterization results of national and world standards.

MATERIALS AND METHODS

Description of Study Area

Debre-Nazret (Fig. 1): Debre-Nazret is part of the south eastern zone of Tigray about 27 km west of Mekelle, capital of Tigray Regional state. It is situated between 13° 26'N - 13° 30'N latitude and 39° 16' - 39° 20'E longitude at an altitudinal range of 1508 to 2160 meter above sea level.

Annual Temperature and Rainfall of Debre-Nazret: The average temperature of the area ranges from 16.3 to 17.2°C and the mean rainfall of the area ranges between 31.5 and 80 mm (Fig. 2).

This (Fig. 2) data was taken from the National Meteorological Services Agency (NMSA) of the country Ethiopia, Tigray state branch, Mekelle City.

Land use of the Study Kebelle: The study area has different land use systems as agricultural land, natural forest, rehabilitated forest, pasture land and unfarmed land (Table 1). This land use system is important for the rural land management activities of the natural resources. Some of the unfarmed land of the study area is given for landless unemployed youth honey bee farmers associations of the kebelle. Some of the unfarmed land of the study area is located towards the river Giba crossing the kebelle and is conducive environment for honey bees with regard to water availability.

Sampling Design of the Research

Study Site Selection: The study was conducted in honey beekeeping potential area, Debre-Nazret kebelle of the Tigray region of Ethiopia. Before the actual survey and site selection, information was gathered from secondary data and preliminary survey from informants. Based on the information obtained from these data, a semi-structured questionnaire was developed and pre-tested for its consistency and applicability to the objectives of the study. Thus the study site was selected purposely. The information from agricultural extension workers and other honey traders convinced the researcher to select the study site purposely.

Data Sources and Methods of Collection: Only white honey samples were collected purposely from the study sites. During the peak harvesting time, twenty one honey samples were collected from the white honey producing study areas and nearby markets. Physicochemical properties were analyzed in triplicates.

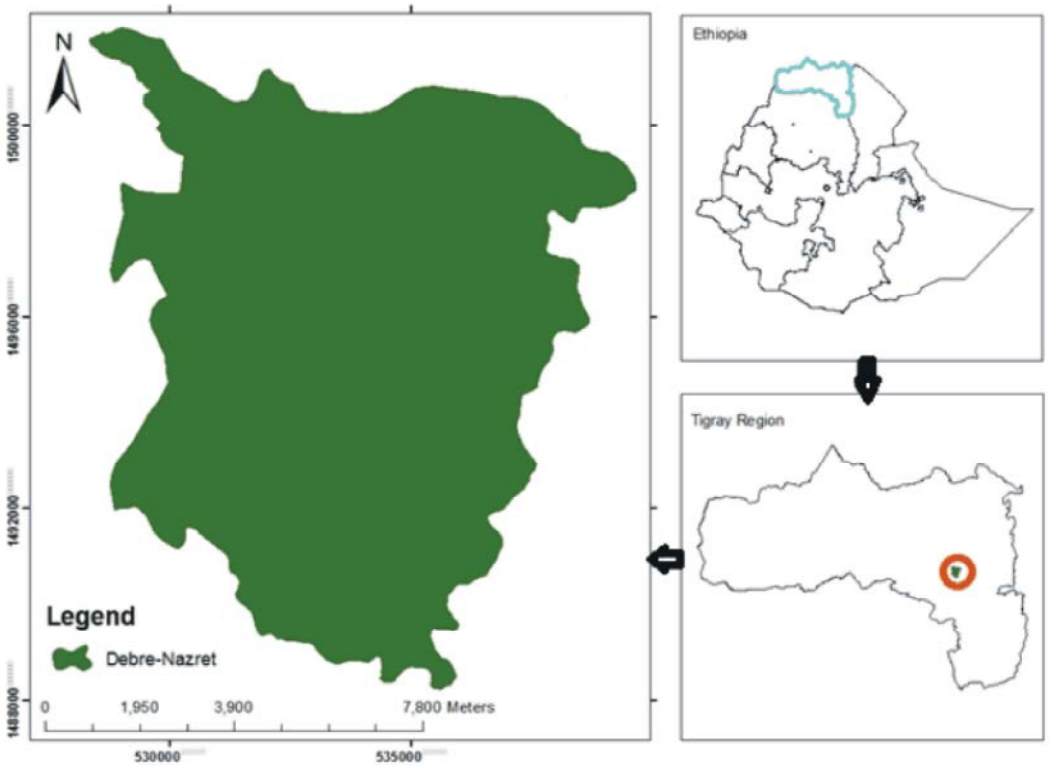

Fig. 1: Map of Debre-Nazret

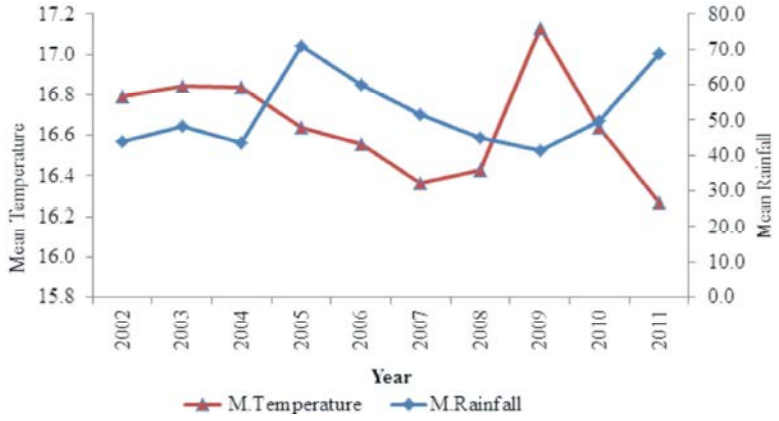

Fig. 2: Mean annual Temperature (C°) and Rainfall (Millimeter) of Debre-Nazret
Source: National Meteorological Services Agency (2012), Ethiopia

Table 1: Land use of Debre-Nazre

	Agricultural land		Natural forest		Rehabilitated forest		Pasture land		Unfarmed land		Total	
Peasant Associat	ha	%	ha	%	ha	%	ha	%	ha	%	ha	%
Mshlam	557(17)	32	2000(60)	41	150(5)	19	400(12)	24	200(6)	6	3307	100
Kolal	237(6)	13	2090(55)	43	98(3)	12	295(8)	17	1045(28)	31	3765	100
Tegoga	492(21)	28	600(26)	12	250(11)	31	658(29)	39	300(13)	9	2300	100
Mamtali	484(15)	27	170(5)	4	300(10)	38	332(11)	20	1848(59)	54	3133	100
Total	1770	100	4860	100	798	100	1685	100	3392	100	12505	

Note: ha = hectare, source: Administrative and Agricultural office of the area

Determination of Honey Quality: To determine the physicochemical characterization quality of honey samples, standard procedure developed and modified from International Honey Commission [19] was used.

RESULTS

Socio- Economic Characteristics of Households: This section provides a summary of the honey beekeeping practices of sample respondents in Debre-Nazret kebelle based on the questionnaire survey result and samples collected. The results were presented and discussed on the basis of response of the households.

Household Characteristics: From the total 65 sample households interviewed about 88.5% were male headed and the rest (11.5%) were female headed. Eventhough female participation is limited in number it showed improvement when compared to the study made by Tessega Belie [20] in Burie district, Amhara Region of Ethiopia. This limited female participation might be due to the culture the region has on bee farming. People of the area believe that the sector is the responsibility of men.The average family size of the sample farmers in the study area was 4.65 persons per household, with maximum and minimum family size of 9 and 1 person, respectively. The mean age of the respondents was 44.39 years (Table 2). The fact that there were landless beekeepers does show how the sector is useful for a society where land is limited and where there are no other options for survival. As can be seen in table 2, the ownership of beehives vary significantly which shows possession difference that calls for a necessary material and/or financial provision necessity to the society.

Of the total households interviewed, 84.6% are married while 9.2%, 3.1% and 3.1% are single, divorced and widowed, respectively. The household samples were also interviewed to describe their participation and involvement in the community (Table 3).

Educational Status of the Household Heads: About 23.1% of the respondents had not received any sort of education, while 33.8% percent could only read and write. The rest were at stages of literacy ranging from 1-8 grades to high school level i.e. 25.4%, 14.6% and 3.1% of the sample respondents had attended 1-4 grades, 5-8 grades and 9-12 grades, respectively. Previous study conducted in Kenya by Gichora [21] noted education, especially biological, is very necessary for beekeepers, which could enable them to understand bee's biology and behavior for better colony management. At least it is necessary to train and persuade beekeepers before distributing modern hives. These calls for the regional or zonal administrators to handle the education aspect of the beekeepers so that they could be productive and responsive citizens.

Family Wise Composition of the Honey Bee Flora: In the study area a total of 31 (Table 4) honey bee flora plant species were documented based on interviewed households. From the documented plant species the most dominant families were Asteraceae, Lamiaceae and Fabaceae (Fig. 3). Euphorbiaceae and Poaceae were the second dominant species in this document.

Constraints of Bee Farming in Debre-Nazret: Though the study areas are major honey beekeeping and honey producing potential areas, there were constraints in different times. The households sample respondents were

Table 2: Age, family size, Farmland holding and Number of bee hive of household (Total sample = 65

Socio Economic indicators	Minimum	Maximum	Mean
Age of household heads (Yrs.)	25.00	70.00	44.39
Family size	1.00	9.00	4.65
Farmland holding (Hectare)	0.00	2.00 hectare	1.12 hectare
Number honey bee hive owned	1.00	100.00	7.77

Table 3: Household heads' participation in the community

Household heads participation	Frequency	%
Political leader	17	26.15
Spiritual (Religious) leader	3	4.62
Elder (Shimagle)	10	15.38
Development group	17	26.15
Community members with no leadership	18	27.69
Total	65	100.0

Table 4: Common honey bee flora plant species in Debre-Nazret

No	Scientific name	Author	Family name	Local Name	Life form	Source (P/N)
1	*Acacia origena*	Hunde	Fabaceae	Cha'a	T	N,P
2	*Achyranthes aspera*	Linn.	Amaranthaceae	Muchele	H	P,N
3	*Agave sisalana*	Perro exEng.	Agavaceae	Eqa	H	N,P
4	*Aloe berhana*	Reynolds	Aloaceae	Ire	H	P,N
5	*Andropogon abyssinicus*	(Fresen.)R.Br	Poaceae	Demahale	H	P
6	*Anethum graveolens*	Linn.	Apiaceae	Shilan	H	P,N
7	*Argemone Mexicana*	Linn.	Papaveraceae	Medafe-Tiln	H	P
8	*Azadirachta indica*	A.Juss.	Meliaceae	Nim	T	P,N
9	*Becium grandiflorum*	(Lam.)Pichi-Sermolli	Lamiaceae	Tebeb	H	P,N
10	*Carduus nyassanus*	R.E.Fries	Asteraceae	dander	H	P,N
11	*Carica papaya*	Linn.	Caricaceae	Papaye	T	P,N
12	*Carissa spinarum*	(Forssk.) Vahl	Apocynaceae	Agamsa	S	P,N
13	*Cordia africana*	Lam.	Boraginaceae	Awhi	T	
14	*Crater ostigma plantagineum*	Hochst.	Scrophulariaceae	Fosi-Anqrbit	H	P,N
15	*Croton macrostachys*	Hochst. ex Del.	Euphorbiaceae	Tambuck	T	P,N
16	*Cynadon doctylon*	-	Poaceae	Tehag	H	P
17	*Dodonaea angustifolia*	L.f.	Sapindaceae	Tahses	S,T	P,N
18	*Erythrina abyssinica*	Lam. ex DC.	Fabaceae	Zwaw	T	P,N
19	*Eucalyptus camaldulensis*	Dehnh.	Myrtaceae	Kelamitos	T	P,N
20	*Euclea racemosa subsp. schimperi*	(A.DC.) Dandly	Ebenaceae	Kuliow	S	P,N
21	*Euphorbium candelabrum*	Trem. ex Kotsch	Euphorbiaceae	Qulqwal	T	P,N
22	*Ficus vasta*	Forssk.	Moraceae	Da'ro	T	N
23	Guizotia abyssinica	(L.f.) Cassini	Asteraceae	Nihug	H	P,N
24	*Hagenia abyssinica*	(Bruce)J.F.Gmel	Rosaceae	Habbi	T	P,N
25	*Helianthus annus*	Linn.	Asteraceae	Suf ferenji	H	P,N
26	*Heliotropium cinerascens*	D.C.	Boraginaceae	Aman gmel	S	
27	*Hypoestes forskaolii*	(Vahl.) R.Sch.	Acanthaceae	Girbya	H,S	P,N
28	*Leucaena leucocephala*	(Lam.)De *Wit.*	Fabaceae	Lukina	S	P,N
29	*Leucas abyssinica*	(Benth.)Briq.	Lamiaceae	Siwakerni	S	P,N
30	*Mangifera indica*	Linn.	Anacardiaceae	Mango	T	P N
31	*Musa X paradisiaca*	Linn.	Musaceae	Muz	H	P,N
32	*Ocimum lamiifolium*	*Hochst.exBenth.*	Lamiaceae	Damakher	S,H	P,N

Note: P=Pollen, N=Nectar, T=Tree, H= Herb, S=Shrub

Table 5: Honey production status of the sampling Households

Tegoga			Mshlam		
Product (kg)	Frequency	Percent	Product (kg)	Frequency	Percent
10-50	14	43.75	10-50	15	45.45
60-100	6	18.75	60-100	7	21.21
110-150	1	3.125	11-150	1	3.03
160-200	2	6.25	160-200	2	6.06
210-250	1	3.125	210-250	1	3.03
260-300	3	9.375	260-300	1	3.03
310-350	2	6.25	310-350	3	9.09
360-400	1	3.125	360-400	-	-
410-450	-	-	410-450	2	6.06
460-500	2	6.25	460-500	1	3.03
Total	32	100	Total	33	100

interviewed to confirm what major constraints they were faced with. As indicated in Figure 4 the most important constraint was drought. Shortage of honey beekeeping equipment and other training on how to use the honey beekeeping materials was the second important constraint (Fig. 4).

The honey characterized shown in Table 6 was directly sampled from the beehive sites. The sample was taken while the farmers were harvesting from their respective beehive in around their homes. The two sampling areas, Tegoga and Mshlam were selected for their high number of beehive colonies availability.

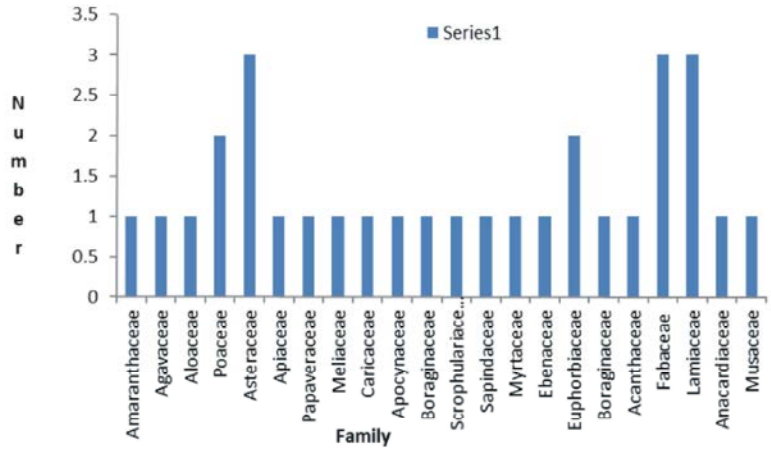

Fig. 3: Family wise composition of the documented honey bee flora species

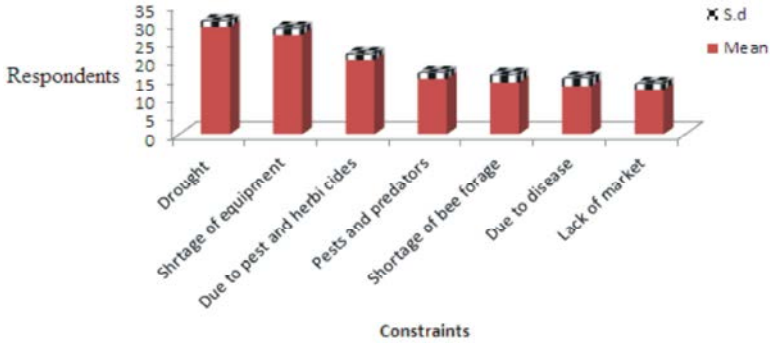

Fig. 4: Major constraints of bee farming in Debre-Nazret

Table 6: Physicochemical characterizations of honey from Debre-Nazret, (N=9)

		Parameters							
Honey sample	Result	PH	Acidity	Moisture	Ash	E.C	Diastase	WIS	HMF
Modern Hive (n=3)	Max	4.12	31	22	0.3	0.51	9.76	0.19	15.12
	Min	3.95	22	17	0.14	0.21	6.14	0.06	14.55
	Mean	4.05	26.12	19.4	0.22	0.37	8.10	0.12	14.84
Traditional Hive (n=3)	Max	4.45	32.7	23	0.3	0.41	8.3	0.38	15.12
	Min	3.89	17.33	17.5	0.09	0.25	4.95	0.21	14.87
	Mean	4.16	25.39	20.13	0.20	0.32	6.60	0.26	15.00
Market honey (n=3)	Max	3.01	22.3	22	0.14	0.3	4.22	0.17	14.96
	Min	4.2	30.3	17	0.29	0.51	8.12	0.29	15.13
	Mean	3.82	26.34	19.4	0.22	0.38	6.25	0.25	15.01

N.B:E.C. = Electrical Conductivity, HMF= Hydroxymethylfurfural, WIS= Water Insoluble Solids

Metal Determination of Honey of the Study Area: Metal determination was made in Ezana mining development laboratory in Mekelle, capital of Tigray. Metals copper (Cu), lead (Pb), cadmium (Cd) and zinc (Zn) were determined in the honey samples (Table 6). Concentration of zinc (Zn) in Tegoga and Mshlam site of Debre-Nazret kebelle was 0.93 and 0.65 mg/kg respectively; this is below the allowable limit of zinc concentration standard set by different organizations. This result is similar with research study conducted in different districts of the Tigray region by [23] and [24]. There is a report that, metal content of honey ($mgkg^{-1}$) to be 4.25,2, 0.11 and 0.05 for Zn, Cu, Pb and Cd respectively in Chile (Latin America) [23].

DISCUSSION

PH of honey samples of the study areas varied from 2.99 to 4.45 with mean value of 3.95. Accessible researches showed that standard pH value of honey to be between 3.2 and 4.5 [1]. About 94.4% of the results for

Table 7: Physicochemical characterizations of honey sampling study area, sampled during harvesting time (N=12)

	Tegoga (n=6)			Mshlam (n=6)		
Parameters	Maximum	Minimum	Mean	Maximum	Minimum	Mean
PH	4.45	3.40	4.01	4.30	2.99	3.88
Acidity	31.00	22.00	25.94	32.70	17.33	25.96
Moisture	23.00	18.20	19.97	22.00	17.00	19.38
Ash	0.30	0.10	0.21	0.30	0.09	0.22
E.C	0.51	0.25	0.39	0.51	0.21	0.33
Diastase	9.76	4.22	6.98	9.04	4.38	6.98
WIS	0.29	0.06	0.20	0.38	0.08	0.22
HMF	15.13	14.87	14.99	15.21	14.66	15.01

Note: E.C. =Electrical Conductivity, HMF= Hydroxymethylfurfural, WIS= Water Insoluble Solids

Table 8: Mean Honey characteristics result of study area, National and World standards

		Standards		
Determined characteristics	Study area result (Mean)	National	World	WHO/FAO
PH	3.95	-	3.2-4.5	-
Acidity,meq kg^{-1}	25.95	40/kg	5-54	40/kg
Moisture,%	19.66	21max	18-23	21-23
Ash,%	0.21	0.6max	0.25-1	0.6-1
EC,$mScm^{-1}$	0.36	-	0.1-3	-
Diastase (Gothe units)	7.00	3min	3-10	-
Water insoluble solids	0.21	0.1max	0.26-0.84	-
HMF meq kg^{-1}	15.00	40max.	40 -80	80max

Source: International Honey Commission [19] and Ethiopian Quality Standard Authority [22]

Note: E.C. = Electrical Conductivity, HMF = Hydromethylfurfural

Table 9: Metal content of honey samples of Debre-Nazret (sample size=12)

		Content of elements in mg/kg with sites	
No	Element	Tegoga	Mshlam
1	Zinc (Zn)	0.65 ± 0.14	0.93 ± 0.07
2	Copper (Cu)	0.23 ± 0.03	0.13 ± 0.04
3	Lead (Pb)	0.03 ± 0.006	0.10 ± 0.01
4	Cadmium (Cd)	0.02 ± 0.005	0.03 ± 0.008

pH value of the honey samples of this study fall within the conventional standard quality honey (Table 8). Acidity (A) of the sampled honey ranged from 17.33 - 32.7 meq/kg with mean of 25.95 meq /kg. The suggested acidity of a honey range from 8.7- 46.8 meq/kg of honey [1]. All results of acidity obtained in this study fall within the recognized standard quality of honey samples (Table 6 and 7). This result has shown that the honey samples cannot allow microbial growth within this range of acidity. The taste of the honey is good.

The moisture contents of honey in this study area varied from 17 to 23 (Table 6). About 86 % of the tested honey samples had moisture content within the accepted range of International Honey Commission [19]. Moisture content is one of the most important guidelines to be considered in the quality determination of honey samples. Honey from the two sampling sites of the study area had similar results. Moisture content of honey from traditional hives was higher than that of modern honey samples (Table 6 and 7). The reason for this result might be that honey sampled from the traditional beehives was analyzed immediately after sampling.

Mineral (Ash) content of the honey samples of this study varied from 0.09 - 0.30 % with a mean of 0.21%. Mineral content of honey samples of different countries ranges from 0.02 - 1.03% [19]. Accepted mineral contents of honey should be less than 0.6% [19]. Mineral contents of all the honey samples (100%) of this study result fall on the accepted standard quality (Table 6 and 7). Electrical conductivity determined result of the honey samples ranged from 0.21 to 0.51 $mScm^{-1}$ with the mean of 0.36. Hundred percent of this study sample result fall on the world set standard (Table 8). Moreover, there was similar result reported by Kebede Nigussie *et al*. [24] conducted in different kebelles of Tigray region of Northern Ethiopia. The electrical conductivity means values of sample 1 and sample 2 was 0.39 $mScm^{-1}$ and 0.33 $mScm^{-1}$ respectively. Modern, traditional and market honey samples had 0.37, 0.32 and 0.38 respectively. The diastase activity of the samples varied from 4.22 - 9.76 Goth scale with mean value of 7. Our country national honey standard for diastase activity is not less than 3 Gothe scale. This current study

result (100%) of the determined honey samples met the standard honey quality (Table 6 and 7). Mean diastase results for the two study sites was the same (6.98%) as showed in the Table 6. Modern honey samples had higher diastase than traditional and market honey samples.

Water Insoluble Solids (WIS) of the honey samples is stretched from 0.06 to 0.38 with the mean value of 0.21. This result was below the world set standard but above the national standard indicated in Tables 6 and 7. The reason for this might be the pure whiteness of the honey i.e. the honey sample has got very small water insoluble solids. Modern honey types had less WIS value. The reason for this might be that modern honey is strained using modern honey extractor machines. In the contrary the WIS mean values for the two study sites was similar, the reason for this might be distribution of the honey bee flora types of the sampling areas is similar. Hydromethylfurfural (HMF) values of the honey samples from Mshlam varied from 14.66 to 15.21 meq kg^{-1} with the mean value of 15.00 and that of Tegoga was 14.87 to 15.13 meq kg^{-1}. The honey samples (100%) fall on national acceptable range of the standards shown in Tables 6 and 7. Concerning with the modern, traditional and market honey sample types the HMF mean values was similar. Moreover, there was no difference mean value of HMF in the two sampling sites. Likewise the honey quality analysis results of the honey samples of the study areas was compared with the national and international honey quality standards of some guidelines. Hundred percent of the sample result agreed with the national and WHO/FAO standards shown in (Table 8).

CONCLUSSIONS

In this study, most of the characterized result of the honey samples of the study area agreed with the national and international set quality standards. Debre-Nazret is endowed with diversity of honey bee flora species. Based on the informants' response, thirty two plants species grouped in to 25 families were documented in this study. From the interviewed households there was a honey bee farmer that harvested up to 500kg per one harvesting year. Of all the list of limiting factors for honey bee farmers, draught was the dominant constraint in the study kebelle. The quantitative result determined for Cu, Pb, Cd and Zn in the study area was below the allowed standards.

Recommendation: With regard to honey quality characterization, we recommend that, study has to be conducted taking pollen analysis parameter in to consideration.

We also recommend that a study has to be conducted considering different agro ecological zones of the Tigray region including the present study kebelle (Medium highland).

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