

# Research Article **Evaluating the Antioxidant Properties of Unifloral Honey** (*Apis mellifera* L.) from Ethiopia

# Ofijan Tesfaye D

Oromia Agricultural Research Institute, Haro Sebu Agricultural Research Center, Oromia, Ethiopia

Correspondence should be addressed to Ofijan Tesfaye; apistesfaye@gmail.com

Received 31 March 2023; Revised 1 June 2023; Accepted 28 June 2023; Published 15 July 2023

Academic Editor: Mohamad Djaeni

Copyright © 2023 Ofijan Tesfaye. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

The antioxidant properties of natural honey primarily rely on the floral origin from which nectar is collected by bees. Thus, the current activity evaluated the antioxidant properties of honey based on its floral type. The honey floral origin was verified by the melissopalynological technique. Antioxidant properties were determined by using standard procedures and analyzed by SAS. Six unifloral honey types with their harvesting month were identified. Accordingly, *Guizotia* (74% of pollen frequency), *Coffea arabica* (68%), *Vernonia* (90%), *Croton macrostachyus* (64%), *Schefflera abyssinica* (100%), and *Eucalyptus* (100%) were cropped in November, February, February, May, April, and June separately. Statistically, a variation (p < 0.05) in antioxidant parameters was displayed between unifloral honeys. *Vernonia* honey exhibited the maximum total phenol (77.2 ± 0.7), total flavonoid (65.0 ± 3.8), and total antioxidant content (65.4 ± 0.3). On the other hand, *S. abyssinica* honey recorded the least total phenol content (24.1 ± 0.4), total flavonoid content (18.6 ± 2.7), and total antioxidant content (5.6 ± 0.5). Statistical analysis showed a positive correlation between all the tested antioxidant parameters. Thus, the current study indicated that all the tested Ethiopian unifloral honey had good sources of antioxidants with the most *Vernonia* honey followed by C. *macrostachyus* whereas *S. abyssinica* honey had the least followed by *Eucalyptus*.

## 1. Introduction

Free radicals are formed in our body during a chemical reaction and lead to damage our cell. It's inhibited by antioxidants produced during our daily natural food. As indicated by Wu and Cederbaum [1], free radicals are too sensitive as they form a bond with other substances, particles, or individual electrons to produce a constant compound and reactive oxygen species take place. This causes many diseases like the formation of cancer, infection, getting old, pathogenesis and development of diabetes [2], circulatory illness, immunity failure, degenerative diseases of the nervous system , heart and lung diseases, and eye problem [3]. Eating natural antioxidants from natural products like honey is active in the hindrance of prolonged illnesses that have amplified in current time [2, 4].

Honey is a normal nutritive antioxidant whose composition is accountable for the redox properties, namely, flavonoids, phenols, enzymes, vitamins, and minerals [5]. Honey is synthesized by bees either from a single plant (its honey is called unifloral honey) or multiple plant species (its honey is known to be multifloral honey), and honey antioxidant activities are determined by its plant and geographical origin, humidity, temperature, climate, and environment condition [6]. The unifloral and multifloral honey sample is authenticated by the melissopalynological method, and taxon of the pollen is typically used to point out the plant nectar origin collected by bees to synthesize honey [7]. Ethiopia has more than ten million colonies, and above eight hundred recognized bee plant [8]. Moreover, the suitability of geographical position, plant diversity, and climatic conditions in Ethiopia makes the topmost honey producer in Africa and tenth from the worldwide [9]. The goal of this work was to screen the antioxidant properties of unifloral honey harvested in Ethiopia.

# 2. Materials and Methods

2.1. Sample Assortment. It was collected as of different areas of the country based on the accessibility of unifloral honey,

No.	Location	Latitude	Longitude
1	Haro Sebu	8° 44′ 59.99″ N	35° 19′ 60″ E
2	Nedjo	10° 49′ 21″ N	35° 14′ 19″ E
3	Gedo	9° 01′ 0″ N	37° 27′ 0″ E
4	Holota	9° 4′ 0″ N	38° 30′ 0″ E
5	Bore	6° 21′ 35″ N	38° 37′ 20″ E
6	Haro Sebu	8° 44′ 59.99″ N	35° 19′ 60″ E

TABLE 1: Latitude and longitude of each locations of unifloral honey sampled.

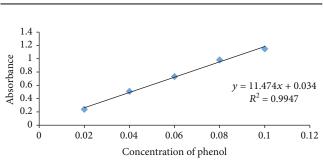


FIGURE 1: Calibration curve for phenol.

and their latitude and longitude are indicated in Table 1. Accordingly, C. arabica and C. macrostachyus honey from Haro Sebu, Guizotia honey from Nejo, Vernonia honey from Gedo, Eucalyptus honey from Holota, and S. abyssinica honey from Bore were collected from farmer beekeepers' apiary site based on their honey harvesting calendar. An overall 30 kg (sample size) of honey trials for each unifloral honey type, from 30 dissimilar apicultures (around 1 kg per farmer) of the study site, has taken. Then after, the representative trials were transported to the University of Addis Ababa, Lab. of Food Science, using sterilized beaker. At the time of cropping, visual remarking of hive environs was done besides group discussion with skilled apicultures of every site to acquire the nectar origin of the samples and flowering bee plants available in the site.

2.2. Floral Source Analysis. The technique of Louveaux et al. [7] was used. Hence, 10g of honey was added in 20 mL of sterile distilled water. The honey solution was centrifuged at 3800 rpm for 10 minutes, and the supernatant was poured out. Then, 20 mL of distilled water was again added to completely dissolve the remaining sugar crystals and centrifuged at 3800 rpm again for 5 minutes, and the supernatant was removed completely. The sediment was spread evenly using a sterile micro spatula on the microscope slide, and the sample was dried for a while. Thereafter, one drop of glycerin jelly was added to the coverslip, and the pollen grains were identified using a pollen atlas [8] which was prepared for plant identification from honey sample. Moreover, pollen morphology types were verified by comparison with reference slides of pollen assorted directly from the live flower plants in the study area. Then after, bee plant species from honey sample was identified; their contribution to bees (pollen, nectar, or both) and life form was known at field and

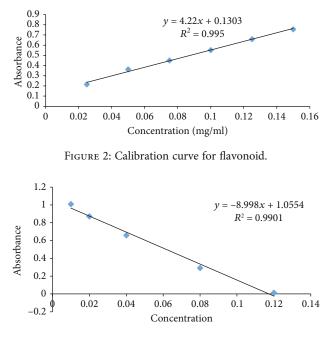


FIGURE 3: Calibration curve for ascorbic acid.

different literatures [7, 8]. The percentage of pollen types in each honey sample was calculated based on the total number of different types of pollen grains counted in each sample. Accordingly, if >45% of counted pollen grain was from specific plant species, it was categorized under predominant pollen (monofloral honey); if 16-45%, secondary pollen; if 3-15%, important minor pollen; and if <3%, minor pollen, while honey sample with no predominant pollen was used as mixed honey type [7]. The pollen count was determined under a light microscope (Swift Instrument International, serial number 8750038, Japan, high power 400x) linked to a computer.

#### 2.3. Antioxidant Properties of Honey

2.3.1. Total Phenolic Contents. The phenol content of unifloral honey was assessed by the Folin-Ciocalteu method [5]. Honey stock solution was formed by dissolving 2g of the honey in 25 mL of distilled water and strained by Whatman no. 1. Then, 0.5 mL aliquot from stock solution was mixed with 2.5 mL of 0.2 N Folin-Ciocalteu reagent and stored for 5 min. A 2 mL of 75 g/L sodium carbonate solution was added to the solution and incubated for 2h at 25°C. Finally, the absorbance of the mixture was calculated at 765 nm using UV (PerkinElmer Lambda 950 UV/VIS/NIR Spectrophotometer). A standard chemical taken to create a calibration curve was gallic acid (0-200 mg/L) as a control. Lastly, composition of total phenol was stated as milligrams of gallic acid per one hundred grams of honey from an average result of triplicate data. The calibration formula  $(y = 11.474x + 0.034; R^2 = 0.9947)$  was derived from the calibration curve (Figure 1).

2.3.2. Total Flavonoid Content (TPC). The procedure by Chua et al. [5] was used. For this, a mixture of 5 g honey

Site	Harvesting time	Scientific name	Family name	Vernacular name (Afaan Oromo)	Life form	Resources for bees	Pollen grain counted (%)	Frequency class	Honey type
		Guizotia spp.	Asteraceae	Tufo/nuugii	Herb	P & N	74.1	ЪР	
		Bidens spp.	Asteraceae	Maxxannee	Herb	P & N	5.9	IMP	
		Trifolium spp.	Fabaceae	Siddisa	Herb	P & N	4.1	IMP	
		Eucalyptus spp.	Myrtaceae	Baargamoo	Tree	P & N	4.1	IMP	
		Grass type	Poaceae	Gosamargaa	Herb	P & N	3.0	IMP	
	M	Plantago lanceolata	Plantaginaceae	Qorxobbii	Herb	Р	2.6	MP	Monofloral
INeajo	INOVEINDEL	Sesamum indicum	Pedaliaceae	Saalixa	Herb	P & N	1.8	MP	(Guizotia spp.) honey
		Allophylus abyssinicus	Sapindaceae	Sarara	Tree	P & N	1.6	MP	
		Vicia faba	Papilionaceae	Baaqelaa	Herb	P & N	1.5	MP	
		Lepidium sativum	Brassicaceae	Feexoo	Herb	P & N	1.1	MP	
		Brassica carinata	Brassicaceae	Raafuu	Herb	P & N	0.2	MP	
		Satureja paradoxa	Lamiaceae	Xinaaddama	Herb	P & N	0.04	MP	
		Coffea arabica	Rubiaceae	Buna	Shrub	P & N	68.0	ЪР	
		Vernonia spp.	Asteraceae	Gosaeebichaa	Shrub	P & N	17.46	SP	
		Terminalia spp.	Combretaceae	Birdheessa/dabaqqa	Tree	P & N	5.16	IMP	
		Guizotia spp.	Asteraceae	Tuufoo/hadaa	Herb	P & N	3.67	IMP	
		Apodytes dimidiata	Icacinaceae	Qumbaala	Tree	P & N	2.15	MP	
		Bidens spp.	Asteraceae	Keelloo	Herb	P & N	1.57	MP	
Haro Sebu	February	Hypoestes triflora	Acanthaceae	Darguu	Herb	P & N	1.22	MP	Monofloral (C. arabica) honey
		Plantago lanceolate	Plantaginaceae	Qorxobbii	Herb	P & N	0.60	MP	110110
		Galiniera saxifraga	Rubiaceae	Mixoo	Shrub	P & N	0.53	MP	
		Justicia schimperiana	Acanthaceae	Dhummuuggaa	Shrub	P & N	0.35	MP	
		Bersama abyssinica	Melianthaceae	Lolchiisaa	Tree	P & N	0.35	MP	
		Rumex nervosus	Polygonaceae	Dhangaggoo	Shrub	P & N	0.15	MP	
		Euphorbia ampliphylla	Euphorbiaceae	Adaamii	Tree	P & N	0.13	MP	
		Croton macrostachyus	Euphorbiaceae	Bakkanniisa	Tree	P & N	64.42	ЪР	
		Syzygium guineense	Myrtaceae	Baddeessaa	Tree	P & N	14.03	IMP	
		Eucalyptus spp.	Myrtaceae	Baargamoo	Tree	P & N	12.57	IMP	
		Acacia spp.	Myrtaceae	Laaftoo	Tree	P & N	7.81	IMP	
Haro Sehu	Mav	Justicia schimperiana	Acanthaceae	Dhummuuggaa	Shrub	P & N	0.55	MP	Monofloral
	67777	Rumex nervosus	Polygonaceae	Dhangaggoo	Shrub	P & N	0.38	MP	(C. macrostachyus) honey
		Coffea arabica	Rubiaceae	Buna	Shrub	P & N	0.27	MP	
		Guizotia spp.	Asteraceae	Tuufoo	Herb	P & N	0.16	MP	
		Bidens spp.	Asteraceae	Keelloo/ Maxxannee	Herb	P & N	0.03	MP	

# International Journal of Food Science

3

	Honey type				Vernonia					S. abyssinica (monofloral) honey	<i>Eucalyptus</i> (monofloral) honey	PP = predominant pollen (if >45% counted pollen grain was from specific plant species); SP = secondary pollen (16%-45%); IMP = important minor pollen (3%-15%); MP = minor pollen (<3%); P & N = pollen and nectar.
	Frequency class	ЪР	IMP	MP	MP	MP	MP	MP	MP	ΡΡ	Ы	5%); MP = min
	Pollen grain counted (%)	90	3.02	2.6	1.4	1.3	1.04	1.04	0.5	100	100	inor pollen (3%-15
TABLE 2: Continued.	Resources for bees	P & N	P & N	P & N	P & N	P & N	P & N	P & N	P & N	P & N	P & N	= important mi
	Life form	Shrubs	Tree	Herb	Tree	Herb	Herb	Shrub	Herb	Tree	Tree	6%-45%); IMP
	Vernacular name (Afaan Oromo)	Eebicha/Reejjii	Baargamoo	Darguu	Qumbaala	Qorxobbii	Qoreeharree	Dhangaggoo	Tuufoo	Gatamaa	Baargamoo	P = secondary pollen (1
	Family name	Asteraceae	Myrtaceae	Acanthaceae	Icacinaceae	Plantaginaceae	Asteraceae	Polygonaceae	Asteraceae	Araliaceae	Myrtaceae	specific plant species); S
	Scientific name	Vernonia spp.	Eucalyptus camaldulensis	Hypoestes triflora	Apodytes dimidiata	Plantago lanceolate	Cardus nyassanus	Rumex spp.	Guizotia spp.	Schefflera abyssinica	Eucalyptus spp.	ounted pollen grain was from
	Harvesting time				February					April	June	ant pollen (if >45% c
	Site				Gedo					Bore	Holota	PP = predomir and nectar.

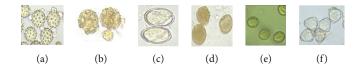


FIGURE 4: Pollen pictures of bee floras that provide unifloral honey in Ethiopia: (a) *Guizotia* spp. (predominant pollen, 74.10%), (b) *Vernonia* spp. (predominant pollen, 90.00%), (c) *C. arabica* (predominant pollen, 68.01%), (d) *C. macrostachyus* (predominant pollen, 64.42%), (e) *S. abyssinica* (predominant pollen, 100%), and (f) *Eucalyptus* spp. (predominant pollen, 100%).

in 50 mL distilled water was used as a honey stock solution, and out of this, 5 mL was dropped in 5 mL of 2%  $AlCl_3$  solution and incubated for 10 minutes. Then, its absorbance was read at 415 nm by spectrophotometer. Then, for calibration curve formulation, a standard chemical which is quercetin (0-200 mg/L) as a control was chosen. This procedure was triplicated and stated as milligrams of quercetin per 100 grams of honey from the averaged result of triplicate data. The calibration equation (y = 4.22x + 0.1303;  $R^2 = 0.995$ ) was derived from the calibration curve (Figure 2).

2.3.3. The Antioxidant Composition. It was founded by calculating the ascorbic acid equal antioxidant capacity (AAEAC) using usual procedures [10]. For this, 0.5 milligrams of DPPH was dissolved in twenty-five millilitres of methanol to get DPPH solution. As well, 30 mg of honey was mixed in millilitre methanol. Then after, 0.75 mL honey solution was mixed in 1.5 mL of DPPH solution. After the solution was incubated at room temperature for 15 min, its absorbance was measured at 517 nm. A mixture of 0.75 mL of a methanolic honey solution with 1.5 mL of methanol was used as a blank. A calibration curve was produced from a standard chemical: ascorbic acid (0-200 mg/L) (control). The procedure was triplicated and stated as milligrams of ascorbic acid per 100 grams of honey. The calibration equation  $(y = -8.998x + 1.0554; R^2 = 0.9901)$  was derived from the calibration curve (Figure 3).

2.4. Statistical Analysis. Average and standard deviations were calculated using SAS Software. Significant variation between unifloral honeys was determined using one-way ANOVA. Antioxidant parameters were used for mean separation by least significant difference.

# 3. Results and Discussion

3.1. Floral Source Result. All floral honey source with their characteristics such as harvesting time, life form, and resource released for bees and pollen frequency category is depicted in Table 2. Pollen pictures of bee floras that provide unifloral honey are illustrated in Figure 4. All the time, honey sample includes various pollen grains which provide a good fingerprint of the geographical and botanical origin where the nectar is collected from [11]. After the pollen grain was counted and the percentage calculated, all honey types were categorized as unifloral honey since their pollen frequency was greater than 45%. Accordingly, there are six unifloral honey types, namely, (1) *Guizotia* honey from the Nedjo area harvested through November, (2) *C. arabica* honey from the Haro Sebu area harvested through February,

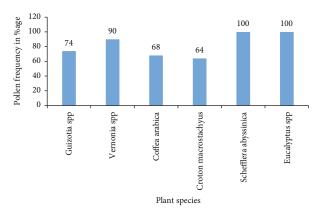


FIGURE 5: Percentage of pollen grain frequency of unifloral honey types.

(3) Vernonia honey from Gedo harvested through February, (4) C. macrostachyus from Haro Sebu area harvested through May, (5) S. abyssinica honey from Bore harvested through April, and (6) Eucalyptus honey from Holota harvested through June. The percentage pollen frequency from Guizotia, C. arabica, Vernonia, C. macrostachyus, S. abyssinica, and Eucalyptus honey samples was 74, 68, 90, 64, 100, and 100, respectively (Figure 5).

Similarly, in Ethiopia, *Guizotia*, *Vernonia*, *C. arabica*, and *S. abyssinica* honeys are harvested from November to December, February, February through March, and April through May [12], respectively. Not all flowering plants equally contribute to the bees. Nectar quality (sugar content), potentiality, and abundance of the plant in a given area contribute to cropping a unifloral honey (predominant pollen source). From this study, secondary pollen contributor (*Vernonia* plant) has occurred in *C. arabica* honey. The study area is well known in coffee production and when flowered is abundantly found and stays for a short period of flowering (less than 10 days). This is concurrent with [13] who observed an overlapping of the *Vernonia* plant flowering period with *C. arabica* from honey samples harvested from February through March.

#### 3.2. Antioxidant Properties

3.2.1. Total Phenolic Content (TPC). The present study screened honey samples between plant sources, and a very significant disparity (p < 0.05) of TPC was obtained in all the tested honey types. TPC is articulated as milligrams of gallic acid per 100g of honey. It ranged from a mean of 24.1 ± 0.4 by *S. abyssinica* to 77.2 ± 0.7 by Vernonia (Table 3). The current result is that the indication of Vernonia honey has a high antioxidant content followed by C.

		TABLE 3: Antioxidant properties between the tested unifloral types.	int properties betv	veen the tested uni	loral types.					
Dometare		Unifloi	ral types (average	Unifloral types (average $\pm$ standard deviation)	( <i>uc</i>		>		A IGN D'N CN	
r atamcters	Vernonia honey	Vernonia honey C. macrostachyus honey Guizotia honey C. arabica honey Eucalyptus honey S. abyssinica honey	Guizotia honey	C. arabica honey	Eucalyptus honey	S. abyssinica honey	4	гог	Г - V	Ś
TPC (mg GAE/100 g of honey)	$77.2 \pm 0.7^{a}$	$70.3 \pm 1.6^{b}$	$57.6 \pm 0.3^{\circ}$	$47.9 \pm 2.2^{c}$	$26.3 \pm 1.1^{e}$	$24.1 \pm 0.4^{\mathrm{f}}$	49.2	2.1	49.2 2.1 <0.0001 2.4	2.4
TFC (mg QE/100 g of honey)	$65.0 \pm 3.8^{a}$	$53.4 \pm 0.5^{b}$	$31.5\pm0.7^{ m d}$	$39.7 \pm 2.2^{\rm d}$	$26.2 \pm 0.2^{e}$	$18.6 \pm 2.7^{\mathrm{f}}$	40.4	3.8	40.4 3.8 <0.0001	5.4
AC (mg AAE/100 g of honey)	$65.4 \pm 0.3^{a}$	$27.4 \pm 1.1^{b}$	$9.0 \pm 4.5^{c}$	$6.4 \pm 1.4^{c}$	$5.9 \pm 0.1^{c}$	$5.6 \pm 0.5^{c}$	19.9	3.5	19.9 3.5 <0.0001 10	10
Different superscripts in a row vary statistically at a 1% probability level. Note: mg GAE/100 g of honey = milligrams of gallic acid equivalent in 100 g of honey; mg QE/100 g of honey = milligrams of quercetin equivalent in 100 g of sample; mg AAE/100 g of honey = milligrams of accorbic acid equal in hundred grams of honey; $X =$ mean; LSD = least significant difference; P-V = p value; CV = coefficient of variation in %	tatistically at a 1% pr E/100 g of honey = m	obability level. Note: mg GAE, illigrams of ascorbic acid equal	/100 g of honey = m in hundred grams o	illigrams of gallic acid of honey; X = mean; LS	equivalent in 100 g of D = least significant di	honey; mg QE/100 g of l fference; P-V = <i>p</i> value; C	noney = V = coel	milligra fficient o	ums of quero of variation	cetin in %
a.										

oes.
5
Ð
floral
l unifi
tested
the
between
it properties
pro
idant
р
oxi
Ξ
An
3
ΓE

*macrostachyus* while *S. abyssinica* honey is a weak antioxidant compound content.

The total phenol substance in honey is examined by TPC which is a fast and simple technique [5]. Moreover, Al et al. [14] demonstrated that TPC was an adequate parameter for an overall phenol approximation in honey and is directly interconnected to the antioxidant action of honey. Furthermore, number of phenols found in the honey sample highly relies on the floral source from which honey is synthesized and is one of the greatest vital classes of substances existing in honey [15].

The TPC of the current study is less than that of Malaysian honey (110.4-196.5 milligram GAE in hundred gram honey) [5] and Sudanese honey  $(201.1 \pm 2.5 \text{ milligram})$ GAE in hundred gram honey) [16]. However, it is found higher than in Germany (4.6 mg/100 g honey) [17] and Slovenia (4.48 mg GAE/100 g honey) [18]. This finding was found within the reported ranges of Northeast Brazilian honey (27.0 to 92.7 mg GAE/100 g) [19] and Ethiopian honey  $(233.3 \pm 24.0 \text{ mg GAE/kg to } 693.3 \pm 26.8 \text{ mg GAE/kg})$  [20]. Nevertheless, as in my study, comparable and higher TPC was investigated by V. amygdalina (693.3  $\pm$  26.8 mg GAE/kg) followed by C. macrostachyus  $(574.2 \pm 40.8 \text{ mg})$ GAE/kg) [20] which is an observer of phenolic content in honey is an indication of its floral source. On the other side, the difference in TPC might be due to floral and environmental origin, method of honey harvesting, duration of honey storage, laboratory procedure, and chemical and reagents used during the laboratory analysis.

3.2.2. Total Flavonoid Content (TFC). It is stated as milligrams of quercetin per 100 g of honey. The presence of flavonoid significantly contributes to the overall antioxidant action of honey, hence take positive properties on human healthiness [21, 22]. An important disparity (p < 0.0001) was cropped among all honey types. As with the TPC, the lowest and highest TFC results of the current study ranged from 18.6 ± 2.7 by *S. abyssinica* honey to  $65.0 \pm 3.8$  by *Vernonia* honey (Table 3). Comparably, [23, 24] demonstrated that honey samples with higher phenolic substance will similarly yield high flavonoid. The flavonoid from the current finding was greater than honey from Turkey (1.1 to 9.2 milligram QE/100 g) as of *A. mellifera* [25] and marketable honey from Portugal ( $1.7 \pm 0.8$  milligram QE/100 g in citrus) from *A. mellifera* [26].

3.2.3. Antioxidant Content (AC). AC of the current study is defined as milligrams of ascorbic acid equal in hundred grams of the sample. As those of TPC and TFC, Vernonia honey demonstrated the highest AC ( $65.4 \pm 0.3$ ) followed by *C. macrostachyus* ( $27.4 \pm 1.1$ ). However, statistically similar (p > 0.05) and less AC was obtained by *Guizotia*( $9.0 \pm 4.5$ ), *C. arabica* ( $6.4 \pm 1.4$ ), *Eucalyptus* spp. ( $5.9 \pm 0.1$ ), and *S. abyssinica* honey ( $5.6 \pm 0.5$ ) (Table 3). The surprisingly highest result was obtained from Vernonia honey in TPC, TFC, and AC.

The AC of the current result was comparable to those from Malaysian honey that recorded an average of 14.23 to 26.64 mg AEAC/100 g [27], Pakistani natural honey (8.30–

TABLE 4: Pearson correlation between TPC, TFC, and AC.

	TPC	TFC	AC
TPC	1		
TFC	0.80375*	1	
AC	0.8737*	0.77178*	1

\* is significant at a 1% probability.

22.10 mg AEAC/100 g [28]), multifloral Burkina Fasan honey (10.20–37.87 mg AEAC/100 g [29]), and Czech honey from 14.15 to 40.71 mg AEAC/100 g [30] while less than Manuka honey (84.47 mg/100 g) [27].

Comparably, Adgaba et al. [20] have recorded higher antioxidant capacities by *V. amygdalina* and *C. macrostachyus* while multifloral and *Guizotia scabra* honey produced relatively lower. The disparities in phytochemicals of the particular honey floras and environmental origin could bring variation in antioxidant properties. Similarly, differences in antioxidant activities of different honeys based on floral and environmental origin and seasonal factors are well testified [31, 32].

3.2.4. Correlation between TPC, TFC, and AC. The correlation matrix is depicted in Table 4. A strong and significant positive correlation was observed among TPC and TFC (r = 0.80, p < 0.0001) and TPC and AC (r = 0.70, p < 0.01). Besides, TFC and AC showed a moderate and important positive association (r = 0.69, p < 0.001). From the current result, the antioxidant properties of any honey type are determined by its phenolic, flavonoid, and antioxidant contents. As the phenolic content of a given honey type increases, then its flavonoid content is also increased and their aggregation could exhibit high antioxidant compounds of honey. The beneficial effect of honey on the health of humans is determined by its phenolic and flavonoid contents. The total phenolic substance in honey is sensitive to polyphenol entities, ascorbic acid, and vitamin E [33].

Comparable correlation with the current study was obtained between TPC and TFC (r = 0.776) and TFC and AC (0.730) [24]. In contradiction with this finding, there is no correlation between TPC and AC (0.165) from Algeria [24] and Czech [30]. A. mellifera honey was observed. However, a highly positive correlation between AEAC and TPC (r = 0.968) from Malaysian raw honey [27] and a positive association between flavonoid, phenolic, and antioxidant activities in Brazilian honey [34] were exhibited which are similar with this result.

# 4. Conclusion

This study showed that the country has a potential for cropping different brands of honey owing to the availability of dominant honey plant diversity in a different environments. No study was carried out on the harvesting period and antioxidant properties of honey based on floral origin. Unifloral honey types, namely, *Guizotia, C. arabica, Vernonia, C. macrostachyus, S. abyssinica,* and *Eucalyptus,* could be cropped in November, February, February, May, April, and June, respectively, where they abundantly occurred and are major honey plants in Ethiopia. Based on the current result, all the tested honey types exhibited good antioxidant properties with the highest TPC, TFC, and AC by *Vernonia* followed by *C. macrostachyus* while *S. abyssinica* was the least followed by Eucalyptus honey. Moreover, TPC, TFC, and AC had a strong positive correlation and are important parameters for the antioxidant constituents of the honey sample.

### **Data Availability**

The datasets used and/or analyzed during the current study are available upon reasonable request from the relevant author.

# **Conflicts of Interest**

No potential conflict of interest was reported by the author.

#### Acknowledgments

I would like to thank the Ethiopian Agricultural Research Institute for financial support and Addis Ababa University, College of Natural and Computational Sciences, Department of Food Science, for antioxidant analysis. I also thank the Holota Apiculture Research Center and botany laboratory technician for the assistance in the melissopalynological analysis. This research was carried out at Addis Ababa University, Department of Food Science, for antioxidant analysis and Holota Apiculture Research Center for melissopalynological analysis.

# References

- D. Wu and A. Cederbaum, "Alcohol, oxidative stress, and free radical damage," *Alcohol Research & Health*, vol. 27, pp. 277– 284, 2003.
- [2] Y. S. Lau, X. Y. Tian, Y. Huang, D. Murugan, F. I. Achike, and M. R. Mustafa, "Boldine protects endothelial function in hyperglycemia-induced oxidative stress through an antioxidant mechanism," *Biochemical Pharmacology*, vol. 85, no. 3, pp. 367–375, 2013.
- [3] N. Koca and F. Karadeniz, "Serbest radikal olusum mekanizmaları ve antioksidan savunma sistemleri," *Gıda Mühendisliği Dergisi*, vol. 16, pp. 32–37, 2003.
- [4] K. D. Bozdogan and B. Mungan, "Effects of variety, maturation and growing region on chemical properties, fatty acid and sterol compositions of virgin olive oils," *Journal of the American Oil Chemists' Society*, vol. 93, pp. 1499–1508, 2016.
- [5] L. S. Chua, N. L. Rahaman, N. A. Adnan, and T. T. Eddie Tan, "Antioxidant activity of three honey samples in relation with their biochemical components," *Journal of Analytical Methods in Chemistry*, vol. 2013, Article ID 313798, 8 pages, 2013.
- [6] K. Elbanna, K. Attalla, M. Elbadry, A. Abdeltawab, H. Gamal-Eldin, and M. F. Ramadan, "Impact of floral sources and processing on the antimicrobial activities of different unifloral honeys," *Asian Pacific Journal of Tropical Disease*, vol. 4, no. 3, pp. 194–200, 2014.
- [7] J. Louveaux, A. Maurizio, and G. Vorwohl, "Methods of melissopalynology," *Bee World*, vol. 59, no. 4, pp. 139–157, 1978.

- [8] N. Adgaba, Atlas of Pollen Grains of Major Honey Bee Flora of Ethiopia, vol. 152, Holeta Bee Research Centre, Commercial Printing Enterprise, Addis Ababa, Ethiopia, 2007.
- [9] N. Kebede, P. Subramanian, and M. Gebrekidan, *Physicochemical Analysis of Tigray Honey: An Attempt to Determine Major Quality Markers of Honey*, Chemical Society of Ethiopia, Addis Ababa, Ethiopia, 2011.
- [10] A. Islam, I. Khalil, N. Islam et al., "Physicochemical and antioxidant properties of Bangladeshi honeys stored for more than one year," *BMC Complementary and Alternative Medicine*, vol. 12, no. 1, p. 177, 2012.
- [11] I. K. Karabagias, A. Badeka, S. Kontakos, S. Karabournioti, and M. G. Kontominas, "Characterisation and classification of Greek pine honeys according to their geographical origin based on volatiles, physicochemical parameters and chemometrics," *Food Chemistry*, vol. 146, pp. 548–557, 2014.
- [12] B. Tura and A. Admassu, "Pollen analysis of honey from Borana zone of Southern Ethiopia," *Journal of Apicultural Science*, vol. 63, no. 2, pp. 233–242, 2019.
- [13] B. Tura and A. Admassu, "Honeybee flora resources of Guji zone, Ethiopia," *Journal of Biology, Agriculture and Healthcare*, vol. 8, no. 21, pp. 1–9, 2018.
- [14] M. L. Al, D. Daniel, A. Moise, O. Bobis, L. Laslo, and S. Bogdanov, "Physico-chemical and bioactive properties of different floral origin honeys from Romania," *Food Chemistry*, vol. 112, no. 4, pp. 863–867, 2009.
- [15] D. Hailu and A. Belay, "Melissopalynology and antioxidant properties used to differentiate *Schefflera abyssinica* and polyfloral honey," *PLoS One*, vol. 15, no. 10, article e0240868, 2020.
- [16] Y. M. Idris, A. A. Mariod, and S. I. Hamad, "Physicochemical properties, phenolic contents and antioxidant activity of Sudanese honey," *International Journal of Food Properties*, vol. 14, no. 2, pp. 450–458, 2011.
- [17] N. Gheldof, X. H. Wang, and N. J. Engeseth, "Identification & quantification of antioxidant components of honeys from various floral sources," *Journal of Agricultural and Food Chemistry*, vol. 50, no. 21, pp. 5870–5877, 2002.
- [18] J. Bertoncelj, U. Doberšek, M. Jamnik, and T. Golob, "Evaluation of the phenolic content, antioxidant activity and colour of Slovenian honey," *Food Chemistry*, vol. 105, no. 2, pp. 822– 828, 2007.
- [19] A. M. de Almeida, M. B. Oliveira, J. G. da Costa, I. B. Valentim, and M. O. Goulart, "Antioxidant capacity, physicochemical and floral characterization of honeys from the northeast of Brazil," *Revista Virtual de Quimica*, vol. 8, no. 1, pp. 57–77, 2016.
- [20] N. Adgaba, A. Al-Ghamdi, D. Sharma et al., "Physico-chemical, antioxidant and anti-microbial properties of some Ethiopian mono-floral honeys," *Saudi Journal of Biological Sciences*, vol. 27, no. 9, pp. 2366–2372, 2020.
- [21] J. M. Alvarez-Suarez, F. Giampieri, A. M. González-Paramás et al., "Phenolics from monofloral honeys protect human erythrocyte membranes against oxidative damage," *Food and Chemical Toxicology*, vol. 50, no. 5, pp. 1508–1516, 2012.
- [22] M. B. Sghaier, I. Skandrani, N. Nasr, M. G. Franca, L. Chekir-Ghedira, and K. Ghedira, "Flavonoids and sesquiterpenes from *Tecurium ramosissimum* promote antiproliferation of human cancer cells and enhance antioxidant activity: a structureactivity relationship study," *Environmental Toxicology and Pharmacology*, vol. 32, no. 3, pp. 336–348, 2011.
- [23] M. Moniruzzaman, M. I. Khalil, S. A. Sulaiman, and S. H. Gan, "Physicochemical and antioxidant properties of Malaysian

honeys produced by Apis cerana, Apis dorsata and Apis mellifera," *BMC Complementary and Alternative Medicine*, vol. 13, no. 1, pp. 1–12, 2013.

- [24] M. Khalil, M. Moniruzzaman, L. Boukraâ et al., "Physicochemical and antioxidant properties of Algerian honey," *Molecules*, vol. 17, no. 9, pp. 11199–11215, 2012.
- [25] Ş. Kivrak, I. Kivrak, and E. Karababa, "Characterization of Turkish honeys regarding of physicochemical properties, and their adulteration analysis," *Food Science and Technology*, vol. 37, pp. 80–89, 2017.
- [26] S. Aazza, B. Lyoussi, D. Antunes, and M. G. Miguel, "Physicochemical characterization and antioxidant activity of commercial Portuguese honeys," *Journal of Food Science*, vol. 78, no. 8, pp. C1159–C1165, 2013.
- [27] S. P. Kek, N. L. Chin, Y. A. Yusof, S. W. Tan, and L. S. Chua, "Classification of entomological origin of honey based on its physicochemical and antioxidant properties," *International Journal of Food Properties*, vol. 20, Supplement 3, pp. S2723– S2738, 2017.
- [28] N. Noor, R. A. Sarfraz, S. Ali, and M. Shahid, "Antitumour and antioxidant potential of some selected Pakistani honeys," *Food Chemistry*, vol. 143, pp. 362–366, 2014.
- [29] A. Meda, C. E. Lamien, M. Romito, J. Millogo, and O. G. Nacoulma, "Determination of the total phenolic, flavonoid and proline contents in Burkina Fasan honey, as well as their radical scavenging activity," *Food Chemistry*, vol. 91, no. 3, pp. 571–577, 2005.
- [30] J. Lachman, A. Hejtmankova, J. Sýkora, J. Karban, M. Orsak, and B. Rygerova, "Contents of major phenolic and flavonoid antioxidants in selected Czech honey," *Czech Journal of Food Sciences*, vol. 28, no. 5, pp. 412–426, 2010.
- [31] L. Estevinho, A. P. Prereira, L. Moreira, L. G. Dias, and E. Pereira, "Antioxidant and antimicrobial effects of phenolic compounds extracts of Northeast Portugal honey," *Food and Chemical Toxicology*, vol. 46, no. 12, pp. 3774–3779, 2008.
- [32] K. Jantakee and Y. Tragoolpua, "Activities of different types of Thai honey on pathogenic bacteria causing skin diseases, tyrosinase enzyme and generating free radicals," *Biological Research*, vol. 48, no. 1, p. 4, 2015.
- [33] M. Ahmed, B. Khiati, A. Meslem, S. Aissat, and N. Djebli, "Evaluation of physicochemical and antioxidant properties of raw honey from Algeria," *Journal of Microbial & Biochemical Technology*, vol. S4, no. 1, p. 6, 2014.
- [34] F. B. Salgueiro, A. F. Lira, V. M. Rumjanek, and R. N. Castro, "Phenolic composition and antioxidant properties of Brazilian honeys," *Quimica Nova*, vol. 37, pp. 821–826, 2014.