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Research Article

Pollen analysis and Physicochemical Characterization of Honey Samples from Owo Local Government Area, Ondo State, Nigeria

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Abstract

Bees (*Apis mellifera* var. *adansonii*) produce honey, a naturally occurring sweet viscid liquid, from blossom nectar and it has both medicinal and antibacterial properties. Pollen, physical and chemical analyses are used to ascertain floral sources, the purity of honey, its botanical, ecological, and geographic provenance, the main honey-producing season, and the processing facilities that honey bees most frequently visit while gathering pollen and nectar. Four samples of honey were acquired from the Owo Local Government Area in Ondo State, Nigeria, and treated to the aforementioned evaluation. Out of thirty six(36) pollen types that originated from 25 plant families, one (1) was identified at the family level, twenty-six (26) at the generic level, eight (8) at the species level, and one (1) was unidentified. The described species come from numerous genera of trees, shrubs, grass, and herbs. Ilale, Isuada, Ipeme, and Alaguntan each had pollen grain counts of 61, 169, 172, and 236, respectively. Alaguntan provided the richest sample, with two hundred and thirty-six pollen counts. Ipeme, Isuada, and Ilale samples were next, with one hundred and seventy-two, one hundred and sixty-nine, and sixty-one pollen counts, respectively. In varying amounts, the predominant pollen types include those of *Elaeis guineens is* followed by Poaceae, *Ageratum conyzoides*, *Hymenocardia acida*, *Phyllanthus* sp., *Tridax procumbens*, *Coffea* spp., *Talinum triangulare*, *Morellia senegalensis*, *Solanum melongena*, and *Mimosa pudica*. The honey samples were all multi-floral. To confirm the safety of the analysed samples of honey, the investigation used pollen densities and quantities. Tests for protein, electrical conductivity, moisture content, ash content, pH level, and specific gravity were all part of the characterization studies. Due to the impact on texture and stability, which are crucial during honey extraction and storage, all of the results for the samples were found to meet the specifications for honey laid out by international regulatory agencies.

Keywords: Honey, Pollen analysis, Physico-chemical examination, multi-floral.

INTRODUCTION

Since ancient times, people have used honey, a complex liquid made by honeybees (*Apis mellifera* var. *adansonii*), as a source of energy, a natural sweetener, and a curative that inhibits the growth of disease-causing organisms (National Honey Board, 2002; Aled et al., 2012; Maddocks et al., 2012; Nwankwo et al., 2014; Ng and Lim, 2015; Adeonipekun et al., 2016; Ng et al., 2017).

In addition to other chemicals required for typical human growth and development, honey contains macro- and micronutrients such as water, carbohydrates, minerals, amino acids, organic acids, proteins, volatile substances, enzymes, and phenolic compounds. The nectar sources have a significant impact on the content of honey (Fernandez-Torres et al., 2005). High-quality honey has been proven to restore damaged intestinal mucosa and facilitates the emergence tissues (Kek et al., 2014; Nolan et al., 2019).

The Greeks also utilized honey as gout, fever, pain, and wound healing treatment. A rising incidence of diseases in recent years has led to an increase in the use of natural honey in place of processed sugar and related products. For instance, Nigerian supermarkets and outdoor markets sell a variety of honey products. The chemical, physical, sensory, and microbiological properties of honey affect its quality (Khalil et al., 2012).Assessing the pollen content of the honey is among the strongest factors to check the provenance and type of flora that produced the honey (Jasicka-Misiak et al., 2012). Several scientists in Nigeria have researched on various elements of honey pollen analysis. Sowunmi (1976), who examined the phytogeographical background of the honey, made the first indigenous attempt. Agwu et al. (1989), Agwu and Abaeze (1991), Sowunmi (2001), Ige and Modupe (2010), Agwu et al., (2013), Oyeyemi (2017) and Essien et al. (2022) are a few other noteworthy contributions. In various regions of the world, research into the use of palynological and physicochemical techniques to determine whether a sample of honey is authentic or contaminated is still ongoing (Saxena et al., 2010; Anklam, 2010; Ramirez-Arriaga et al., 2011; Rateb and Hussein, 2012). According to Lawal et al. (2009)'s report, it was discovered that honey samples produced in various parts of Turkey, where floral diversity was relatively significant, did not share the same quality. This can be explain by the various geographical areas from where honey samples were gathered and, consequently, the variety of plant species that go into honey production. Regulations that govern honey packaging in the European Community advocate using labels to specify the product's floral and geographic backgrounds as well as its customer satisfaction (CODEX, 2001). The authentication of the honey's botanical source responds to consumer needs and ensures product quality, prohibiting frauds. As a consequence, it becomes imperative to characterize honey samples in precise details. Numerous honeys produced in massive quantities in Spain, including Eucalyptus, Heather, Lavender, Thyme, Citrus, Rosemary, and honeydew, have been well evaluated (Ferna´ndez-Torres et al., 2005).

Through the use of pollen and physicochemical analysis, the current study sought to identify the qualities of honey samples and assess the veracity of honey from the study area.

MATERIALS AND METHODS

Study Area

The study area is located on the Northern edge of the Yoruba Hills, between latitude 70^{o}11’46"N and longitude 50^{o}35’12"E. Nigeria's Ondo State is situated in the southwest. There are two seasons (the dry season and the wet season) which define the region's climatic conditions. The wet season begins in March and lasts until roughly November. The dry season, which lasts throughout February, officially begins in December. In Owo, the month of August seems to have the lowest temperature of the year. Honey flows in these regions between September and April. Despite a high level of anthropogenic activity, pollen assemblages provided evidence that the vegetation of the study region was of the tropical rainforest vegetation type (Essien and Ige, 2019).

Collection of honey samples

Four samples of honey were used in this investigation, and they were from Ilale, Isuada, Ipele, and Alaguntan communities in Owo Local Government Area of Ondo State, Nigeria. In sealed, labeled plastic bottles, the honey samples were shipped to the Palynology Research Laboratory at Adekunle Ajasin University, Akungba-Akoko.

Sample preparation, Mounting and Microscopic Examination

We carefully weighed 10g of filtered honey from each sample using a weighing balance, and the colour was noted. Each sample of honey was vigorously shaken to fully mix and distribute the chemical and botanical components. The vigorously shaken sample was then diluted with 35 ml of warm (40–50^{o}C) dilute sulphuric acid solutions (3ml in 100ml of water) in accordance with the procedure of Agwu et al. (2013).The honey-acid solution was thoroughly shaken before being centrifuged for 5 minutes at a speed of 2000 rpm. The supernatant was then decanted. For each sample of honey, the density of the retrieved polleniferous granules was determined. The recovered particles were treated with ten milliliters of ethyl acetate to eliminate water prior to acetolysis. Fresh acetolysis mixture was made by mixing concentrated sulphuric acid and acetic anhydride in a 9:1 ratio. The strategies of Erdtman (1969) and Agwu et al. (2013) were used to do the acetolysis. In boiling water set at boiling point (100^{0}C), the solution was mixed with the sediment and suspended for five minutes before being removed to cool for ten to twelve minutes. After 5 minutes of centrifuging and stirring, the supernatant was collected. The recovered granules then were spun twice, washed two times using distilled water, and decanted. The recovered residues were kept in a glycerin and ethanol solution in storage plastic vials (2:1).

Each vial's contents were vigorously shaken before two drops of each suspension were put on a microscope slide and covered with an 18x18mm^2 cover slip. To keep the mount from drying out, it was sealed off with colourless nail polish. The 18mm^2 surface was covered equally with the sample, and no samples leaked from the sides. The prepared slide was subsequently examined under an x40 Olympus microscope for counting and identification. Reference descriptions and photomicrographs from books and journals, including Sowunmi (1978; 1995), Agwu and Akanbi, (1985), Agwu et al. (2013), Shubharani et al. (2013), Essien (2014) and Essien and Ige (2019), were used for identification, counting, and categorization. Pollen grain frequencies are denoted by the Louveaux et al. (1970) classification of highly frequent (above 45 percent), frequent (16-45 percent), rare (3-15 percent), and sporadic (less than 3 percent).

Physicochemical Analysis

In order to quantitatively assess the moisture content, protein, ash content, specific gravity, pH, and electrical conductivity of four samples of honey, physicochemical analysis was performed on the samples. The associations of official analytical chemists' established procedures were used to evaluate these parameters (A.O.A.C, 1990).

Determination of pH

The pH of a sample of honey was measured using a pH meter (HANNA Instrument model HI 2211) from a solution of 20g of honey in 75ml of distilled water in a 250ml beaker. It was stirred using a stirrer. The pH electric meter was immersed in the solution, read, and recorded.

Determination of moisture content

Gravimetric measurement was the method used. The weight of a previously moistened can was measured using a weight sample (5.0g). The sample in the can was dried in a 105°C oven for three hours. After chilling in a desiccator, it was weighed. The sample was returned to the oven to complete drying. After several hours of drying, cooling, and weighing the sample again, there were no longer any weight losses (i.e., constant weight was obtained).
The weight of the moisture lost was calculated and reported as a percentage of the weight of the sample analyzed. It was expressed in the manner of:

$$\% \text{ Moisture content} = \frac{W_2 - W_3}{W_2 - W_1} \times \frac{100}{1}$$

Where;
W_1 = Weigh of empty moisture can
W_2 = Weight of empty can + sample before drying
W^3 = Weight of can + sample dried to constant weight.

Determination of Protein

James' Kjeldah technique was used to accomplish this. The total nitrogen was determined and multiplied by 6.25 for the protein reactions. Precisely 0.5g of the substance was added to 10 ml of concentrated H_2SO_4 in a digestion flask. A selenium catalyst tablet was added, and it was heated under a fume cupboard until a clear solution was created (i.e., the digest). Before being used for the analysis, the digest was boiled in a volumetric flask and diluted to 100 cc.10 ml of the digest were mixed with an equivalent volume of a 45 percent NaOH solution in a kjeldahl distillation apparatus. 50cc of the distillate in total were collected and titrated from green to a deep crimson end point against 0.02 N EDTA. In 10 ml of 4 percent boric acid with 3 drops of mixed indicator (broocressol green/methyl red), the liquid was distilled. A blank reagent was also broken down, distilled, and titrated. The nitrogen content, and consequently the protein content were calculated using the formula below:

1 Mole of IN H_2SO_4 = 14 mg N_2

$$\% \text{ Protein} = \% N_2 \times 6.25$$

$$\% N = \frac{100 \times N \times 14 \times V}{W \times 1000 \times Va} - T - B$$

W = Weight of sample (0.5g)
N = Normality of titan (0.02N H_2SO_4)

Vt = Total digest volume (100 ml)
Va = Volume of digest analysed (10 ml)
T = Sample titer value
B = Blank titer value

Determination of Ash content

This was accomplished using the furnace's gravimetric incineration. A ceramic crucible that had already weighed exactly 50g was filled with the sample. The sample was heated to 550°C for three hours in a muffle furnace, reduced to ashes, cooled in a desiccator, and weighed again. The weight of the ash recovered was calculated by difference and expressed as a percentage of the weight of the sample under study, as shown in the formula below:

$$\% \text{ Ash} = \frac{W2 - W1}{W1 \text{ of sample}} \times \frac{100}{1}$$

Where;
W_1 = Weight of empty crucible
W_2 = Weight of crucible + ash

Determination of Electrical conductivity

The pH meter was used to test the electrical conductivity in millivolts after 20g of honey was dissolved in 100 ml of distilled water and properly mixed to produce a solution

.

Determination of Specific gravity

The ratio of the sample weight to the weight of an equivalent volume of water was used to calculate the specific gravity.

$$SG = \frac{W_{Sb} - W_b}{W_{Wb} - W_b}$$

Where;
W_b = Weight of pycnometer.
W_{sb}= Weight of the sample + pycnometer
W_{wb}= Weight of the water + pycnometer

RESULTS AND DISCUSSION

Pollen analysis

The relative abundance (quantity) of pollen in each sample has served as a metric for the purity and reliability of the honey samples. A total of thirty-six (36) pollen types belonging to twenty-five (25) plant families were encountered in the study; one (1) was identified to family level, twenty-six (26) to generic level, eight (8) to species level, and one (1) was unidentified (Table 1). The encountered species are among the diversity of flora (grasses, trees, shrubs, and herbs) present in the local vegetation of the study area. Pollen grain counts varied from 61 to 236 for Ilale, Isuada, Ipeme, and Alaguntan, respectively. There were 26 different pollen types found in Alaguntan, 20 in Ipeme, 19 in Isuada, and only 12 in Ilale; the pollen type with the fewest numbers.

Table 1. The pollen spectrum of honey samples from the study area

S/N	POLLEN TYPES	LOCALITIES				
		ILALE	ISUADA	IPEME	ALAGUNTAN	TOTAL
1.	ANACARDIACEAE					
	Lannea spp.	1	0	0	0	1
	Mangifera indica	0	0	4	2	6
2.	ANNONACEAE					
	Annona senegalensis	0	0	3	7	10
3.	ARECACEAE					
	Elaeis guineensis	20	62	55	109	246

Continuation of table 1

4.	ASTERACEAE					
	Ageratum conyzoides	0	2	8	3	13
	Tridax procumbens	0	3	4	2	9
5.	BOMBACACEAE					
	Ceiba pentandra	0	0	4	0	4
6.	COMBRETACEAE					
	Combretum spp.	7	0	9	0	16
7.	COMMELINACEAE					
	Aneilema spp.	0	0	3	0	3
8.	CONVOLVULACEAE					
	Merremia spp.	2	1	0	0	3
9.	EUPHORBIACEAE					
	Euphorbia hirta	0	0	0	4	4
	Manihot spp.	0	3	1	0	4
10	FABACEAE: SUB-FAMILY					
	1. CEASALPINIOIDEAE					
	Cassia fistula	0	1	0	4	5
	Caesalpiniapulcherrima	0	0	0	3	3
	Delonix regia	6	0	0	10	16
	Mimosa pudica	0	1	7	2	10
	Zygia latifolia	1	0	0	0	1
	2. MIMOSOIDEAE					
	Albizia zygia	0	3	0	5	7
	Gliricidia sepium	0	2	1	5	9
11	IRVINGIACEAE					
	Irvingia gabonensis	1	0	0	3	4
12	MALVACEAE					
	Sida acuta	0	0	0	1	1
13	MELIACEAE					
	Trichillia prieureana	0	3	1	7	11
14	MORINGACEAE					
	Moringa oleifera	0	0	3	0	3
15	MYRTACEAE					
	Syzygium guinense	0	0	0	3	3
16	PHYLLANTHACEAE					
	Hymenocardia acida	0	9	5	6	20
	Phyllanthus spp.	0	11	5	3	19
17	POACEAE	15	45	32	25	117
18	PORTULACACEAE					
	Talinum triangulare	2	2	12	5	21
19	PROTEACEAE					
	Protea elliottii	0	0	0	1	1
20	RUBIACEAE					
	Coffea spp.	0	10	5	7	22
	Morellia senegalensis	1	2	6	7	16
21	RUTACEAE					
	Citrus sp.	3	0	0	2	5
22	SAPINDACEAE					
	Paullina pinnata	0	0	0	3	3
23	SOLANACEAE					
	Solanum melongena	0	6	4	7	17
24	VERBENACEAE					
	Vitex grandifolia	0	1	0	0	1
25	Indeterminata	2	2	0	0	4
	Total Pollen counts	61	169	172	236	638

The four samples of honey were all pure. Based on pollen weights larger than 0.4g, this was determined (Agwu and Akanbi, 1985). The predominant pollen types in the four honey samples include those of *Elaeis guineensis*, Poaceae, *Ageratum conyzoides*, *Hymenocardia acida*, *Phyllanthus* species, *Tridax procumbens*, *Combretum* species, *Coffea* species, *Talinum triangulare*, *Morellia senegalensis*, *Solanum melongena*, *Mimosa pudica*, and *Manihot* species (Table 2). Bees prefer the plant that grows a lot; the essential pollen types spanned from "frequent to sporadic". The relative abundance of the pollen types present in the honey samples are shown in Figure 1 and the predominant pollen types, percentage occurrences and classification of the honey are presented in Table 2.

Table 2. Predominant pollen types, percentage occurrences and classification of the honey

Localities	Class of honey	Pollen type	Percentage composition	Frequency class
Alaguntan	Multi-floral	*Elaeis guineensis*	17.08	Frequent
		Poaceae	3.92	Rare
		Delonix regia	1.56	Rare
		*Coffea*sp.	1.09	Rare
		Morellia senegalensis	1.09	Rare
		Annona senegalensis	1.09	Rare
		Solanum melongena	1.09	Rare
		Trichillia preuriana	1.09	Rare
		Hymenocardia acida	0.94	Sporadic
		Albizia zygia	0.78	Sporadic
		Talinum triangulare	0.78	Sporadic
		Irvingiagabonensis	0.47	Sporadic
		Syzygium guineense	0.47	Sporadic
Ipeme	Multi-floral	*Elaeis guineensis*	8.62	Rare
		Poaceae	4.68	Rare
		Talinum triangulare	1.88	Rare
		Combretum spp.	1.41	Rare
		Agerattum conyzoides	1.25	Rare
		Mimosa pudica	1.09	Rare
		Morellia senegalensis	0.94	Sporadic
		Hymenocardia acida	0.78	Sporadic
		Ceiba pentandra	0.62	Sporadic
		Mangiferaindica	0.62	Sporadic
		Solanum melongena	0.62	Sporadic
Isuada	Multi-floral	*Elaeis guineensis*	9.71	Rare
		Poaceae	7.03	Rare
		Phyllanthus spp.	1.72	Rare
		Coffea spp.	1.56	Rare
		Hymenocardia acida	1.41	Rare
		Solanum melongena	0.94	Sporadic
		Manihot spp.	0.47	Sporadic
		Tridax procumbens	0.47	Sporadic
		Gliricidia sepium	0.47	Sporadic
		Trichillia preuriana	0.47	Sporadic
Ilale	Multi-floral	*Elaeis guineensis*	3.13	Rare
		Poaceae	2.35	Rare
		Combretum spp.	1.09	Rare
		Delonix regia	0.94	Sporadic
		Citrus spp.	0.47	Sporadic
		Merremia spp.	0.31	Sporadic
		Talinum triangulare	0.31	Sporadic

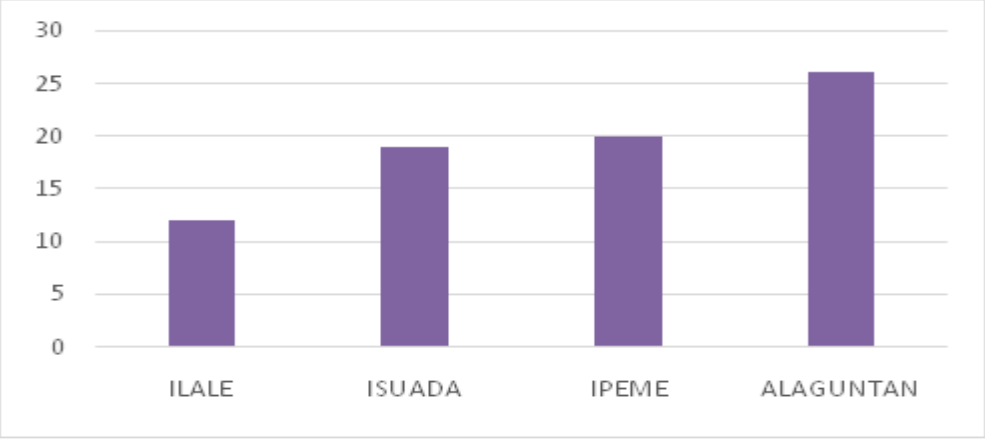

Figure 1. Histogram showing the abundance of pollen types in the study locations

Pollen grains of diverse forms, sizes and morphological features were found in the honey samples, indicating that they were all multi-floral honeys made from a range of nectar sources. The four samples of honey used for this investigation

were all multi-floral and distinguished by a variety of pollen types. Indicator species that honeybees commonly visit for pollen and nectar sources have been identified in this study. In designed to motivate competitive honey production in the study locality and Nigeria in general, such plants could be properly conserved and managed for long-term exploitation.
It has been reported by Richard (1999) that lighter-coloured honeys are supplied directly for consumption whereas darker-coloured honeys are frequently used in industrial applications. In this study, the four samples of honey that were analysed varied from light brown to yellowish brown to dark brown in colour. Each sample gives a visual representation of pollen in honey, the colour of diluted honey, and the amount of honey (Table 3).

Table 3. Colour of diluted honey and weight of pollen in the honey sample

Localities	Color of diluted honey	Weight of honey (gram)	Weight of pollen (gram)
Ilale	Light brown	10	0.20
Isuada	Dark-brown	10	0.23
Ipeme	Yellowish brown	10	0.35
Alaguntan	Dark brown	10	0.32

Physicochemical analysis

The physicochemical parameters tested for were: pH value, Moisture content, Ash content, Electrical conductivity, Proteins and Specific gravity. The results are presented in (Table 4).

The pH values

The pH range for blossom honeys should be between 3.2 and 4.5, according to CODEX (2001) international standards, and all of the honey samples met this requirement.

According to CODEX (2001) international regulations and all of the honey samples surpassed this specification. The four samples were all certified acidic, with pH values ranging from 3.36 to 4.04 (Table 4). The results conformed to Saxena et al. (2010)'s assertion that honey is usually acidic regardless of its source. Similar results were reported by Lawal et al. (2009) who opined that samples exhibiting acidic pH values were pure and had the ability for longer shelf life.

Table 4. Physicochemical parameters of the studied honey sample

LOCATIONS	Moisture content (%)	Specific gravity	pH value	Electrical conductivity (Mv)	Protein (%)	Ash content (%)
ILALE	14.29%	1.47	4.03	27.3%	0.95	0.02
ISUADA	17.04%	1.52	3.70	83.5%	1.11	0.05
IPEME	13.39%	1.49	3.36	98.7%	0.91	0.03
ALAGUNTAN	19.35%	1.50	3.59	88.6%	1.75	0.02

Conductivity

Electrical conductivity is frequently used in honey quality control and purity techniques since it is considered to be a reliable criterion. Every one of the samples' electrical conductivity data (Table 4) indicated that they were all within permissible limits. The electrical conductivity of the Ipeme honey sample was the highest (98.7 mV), whereas the Ilale honey sample had the lowest electrical conductivity (27.3 mV) but the highest pH level. High conductivity was reportedly correlated with high pH by Ouchemoukh et al. (2010).

Protein content

The samples of honey had a protein content that varied from 0.91 to 1.75 percent. As per the CODEX (2001) standard, this was accomplished. The findings reinforce Njokuocha and Osayi's (2005) report, which highlighted that floral origin, nectar sources, and quality, as well as pollen type in the honey sample; all have an influence on the protein composition of honey, which varies extensively in comparison to the plant sources.

Moisture content

Maturity level is inversely correlated with water content. The obtained water contents ranged from 13.3 to 19.35%. Among the analysed samples, the honey samples from Ipeme and Alaguntan had the lowest and highest water content. Getachew et al. (2014) suggest that depending on the source of the honey, the season, and other parameters, the moisture content of honey can naturally range between 13 and 23%. High moisture load in some varieties of honey may

cause the honey's water activity to ferment and hasten crystallization (Gomes et al., 2010). Honey with a value around 16 and 18% is considered to be the best honey for conservation and storage, according to Azzedine et al. (2007). Premature extraction may provide honey samples with high moisture content. Honey with a longer shelf life presumably has lower moisture content, according to Fredes and Montenegro (2006).

Ash content

The other honey samples exhibited total ash values of 0.03 percent (Ipeme), 0.02 percent (Alaguntan), and 0.02 percent (Ilale), all of which were within the regulatory agency's restrictions. Isuada had a total ash content of 0.05 percent. The Isuada honey sample had the highest ash content (0.05%), which was within the threshold of 0.50% that was considered to be suitable. These findings are in agreement with those of White (1975), who evaluated various varieties of honey and discovered that their ash contents ranged from 0.020 to 1.028 percent. The variation may originate from variables including plant physiology, weather factors, and soil properties.

Specific gravity

Isuada and Ilale already had a specific gravity of 1.52, and Ilale reached 1.47. These honey sample specific gravity estimates are relatively close to the range of 1.47-1.52 (Table 4). This breaches the 1.38–1.45 range of the established Codex standard (Adams et al., 2010)

Botanical origin of honey

Due to the obvious considerable species diversity prevalent in anthropogenically disturbed mosaics of lowland rainforest and secondary grassland, pollen grains from various plants families and pollen types are detected in honey samples. The honeys from the transition zones exhibited a plethora of pollen types. Irrespective of source locations and attendant localized ecological characteristics, some species were common to some localities while others were encountered in all samples analysed. For example, *Elaeis guineensis*, Poaceae, *Delonix regia*, *Morellia senegalensis*, *Solanum melongena*, *Phyllanthus* spp. and *Talinum triangulare* are a few of the pollen types that occur in this category. The presence of a diversity of major pollen types in the four samples proved that the honey samples were of botanical origin and provided a clear indication of their geographical origin (Ige and Modupe, 2010). Similar findings were reported by Essien and Ige (2020) and Aina and Owonibi (2011).

Geographical origin of honey

The majority of honey samples in Nigeria are sourced in the Mosaic of lowland rainforest and secondary grassland. Lowland rainforest and derived savanna vegetation are characterized by *Elaeis guineensis*, Poaceae, *Morellia senegalensis*, *Tridax procumbens*, and *Phyllanthus* spp. Farmlands and suburban complexes are distinguishable by the sight of these indicator floras as reflected by their pollen. Published investigations undertaken across the globe had already established that the pollen content of a sample of honey can be used to pinpoint the geographical origin of honey. Similar findings were reported by Maurizio (1951), Sowunmi (1976), and Agwu et al. (2013).All of the aforementioned pollen types were detected in varying amounts, indicating confirmations that their geographical backgrounds being disturbed tropical lowland rainforest and secondary grasslands.

Season of honey production

The periods within which honey is produced can be linked to the flowering phases of the floras which their pollen reflects. The majority of Nigerian honey is produced during the season with no or little rainfall (that is September to April) according to ethno-linguistic evidence and market study report. Tropical plants, specifically trees, flower instantly once the sunshine is high intensely and the atmosphere becomes less humid, and the climatic conditions are naturally more suitable for the insect's motion including honey bees. For instance, *Elaeis guineensis* flowers bloom all year but peaks in October to April, Poaceae flowering from October to June, *Annona senegalensis* flowers in April, Asteraceae taxa blooms from April to December, *Hymenocardia acida* blooms from January to March, *Syzygium guineensis* blooms from November to May, *Phyllanthus* sp. blooms from January to October, and *Lannea* (January to April). The presence of pollen grain from *Phyllanthus* and Asteraceae plants in the honey samples suggests that honeybees foraged from December to June. Those from the Asteraceae family and *Elaeis guineensis* suggest that the honeybees gather their food between October to April. The findings reinforce with the report of Essien (2020) and Sowunmi (1976). As a consequence of the predominant nectar plants' flowering cycles, the honey samples from the study localities were

primarily produced between the months of October and April. Pollen analysis revealed that the honey samples acquired from sourced localities demonstrated that honeybees foraged for both native and exotic flora from the various and different floral sources used in the honey production.

CONCLUSION

Numerous indicator species were identified by pollen examination of honey specimens from four localities in Owo Local Government Area of Ondo State, Nigeria. The honey's pollen spectrum substantially matched the floristic makeup of the vegetative environment where it was produced. This study revealed that the weight and the proportion of pollen can be leveraged to discern between adulterated and pure honeys. The finding of this research also demonstrated that the marker taxa that honeybees (*Apis mellifera* var. *adansonii*) routinely visit in supplies of nectar and pollen were the principal pollen types that were observed in these catchment areas. The outcomes should be efficient in guiding the conservation and ethical exploitation of these indicator species, which will boost the region's capacity to produce large quantities of honey. The honey samples' physicochemical investigation suggests that, when compared to the Codex honey standard, these were of relatively high quality and equivalent to those reported in earlier investigations for honey quality assurance. The honey specimens' acidified pH levels confirmed overall integrity and indicated that they might have a long shelf life.

For the identification, multiplication, cultivation, conservation, and even the sustainable usage of these taxa, the creation of a pollen flora and/or atlas for the honeybee foraged plants of a study area is highly imperative. Attributable to the underlying facts that a vegetation source (or sources) seems to have a substantial impact on the physical and chemical characteristics of honey, is considered to be integral towards its establishment. It is essential to attach importance towards how contrasting honey as well as the procedures suppliers use for its extraction, processing, storage, and preservation.

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Conflict of interest

The authors declare that there is no conflict of interest.

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