

Botswana Journal of Agriculture & Applied Sciences

Leading Agriculture through Science and Innovation

Please cite this article as: **Bafo, W., Seifu, E., Molapisi, M. and Sonno, K. (2019).** Meliponiculture and physicochemical properties of honey produced by the African stingless bee *Plebeina hildebrandti* Friese in Kalakamati village, Botswana. *Botswana Journal of Agriculture and Applied Sciences* 13 (Issue 1 – Special): 33–42.

The online version of this article is located on the World Wide Web at:

<https://bojaas.buan.ac.bw>

The views expressed in this article are those of the author(s) and not the publisher. The accuracy of any instructions, formulae, and drug doses should be independently verified with primary sources. The publisher shall not be liable for any loss, actions, claims, proceedings, demand, or costs or damages whatsoever or howsoever caused arising directly or indirectly in connection with or arising out of the use or misuse of this material.

This article may be used for research, teaching, and private study purposes. Any substantial or systematic reproduction, redistribution, reselling, loan, sub-licensing, systematic supply, or distribution in any form to anyone is expressly forbidden.

Meliponiculture and physicochemical properties of honey produced by the African stingless bee *Plebeina hildebrandti* Friese in Kalakamati village, Botswana

Bafo, W., Seifu, E.^{*}, Molapisi, M. and Sonno, K.

Department of Food Science and Technology, Botswana University of Agriculture and Natural Resources, Gaborone, Botswana.

*Corresponding Author: eseifu@buan.ac.bw; eyassu.b@gmail.com; Tel: +2673650147; Fax: +2673552908

WB conducted the study (proposal development, data collection and report write-up). ES served as major supervisor of the student, conceived the research idea, designed the study, contributed in proposal development and write-up of final report, drafted the manuscript. MM served as co-supervisor of the student. KS contributed during laboratory analysis of samples and co-edited the manuscript.

ABSTRACT

This study was conducted to assess the harvesting practices in Kalakamati village of honey produced by the African stingless bee *Plebeina hildebrandti* Friese and to determine the physicochemical properties of this honey. To understand stingless bee honey harvesting practices, a semi-structured questionnaire was prepared and 15 individuals who have experience in collecting stingless bee honey were interviewed. The physicochemical properties of four honey samples were analysed following standard procedures. The stingless bee honey had average moisture (28.4%), reducing sugar (61%), apparent sucrose (2.4%), free acidity (36 meq/kg), pH (4.77), total ash (0.42%), hydroxymethylfurfural (HMF) (17.82 mg/kg) and water-insoluble solids content of 0.07%. All the parameters comply with the Tanzanian standard for stingless bee honey whereas all but moisture content agrees with the European standard for honey from *Apis mellifera*. Moisture content of the stingless bee honey in the present study is higher than the 21% EU standard for *Apis mellifera* honey. In Kalakamati village, honey from stingless bees is collected from the bush and stingless bees are not kept by farmers. According to the respondents, honey from stingless bees is consumed as it is, is used as a substitute for sugar in tea and for its medicinal value to treat cough, stomach ache and flu. The results suggest that honey produced by stingless bees in Kalakamati village is of good quality as it generally meets international quality standards. Honey production from stingless bees could contribute to poverty alleviation and food security in the study area if appropriate interventions are made.

Keywords: Botswana, honey quality, meliponiculture, stingless bee, *Plebeina hildebrandti*

INTRODUCTION

Beekeeping is an important agricultural activity in the world. Honey produced from beekeeping is the major product of honeybees and it has important nutritional characteristics and provides significant economic contributions. Honey is a sweet substance that bees make from nectars or juices and exudates that are found on living parts of plants (Nkoba, 2012). Honey produced by honeybees of the genus *Apis* is the most commonly utilized type, worldwide. Honey is also produced by bumblebees, stingless bees, and other hymenopteran insects such as honey wasps, though the quantity is generally lower and they have slightly different properties compared to honey from the genus *Apis* (Eardley, 2004). Africa has numerous types of wild honey bees which exist everywhere on the continent, from the equatorial evergreen rain-forest to the desert oasis.

Africa is also believed to be the origin of stingless bees, which have dispersed to other tropical and subtropical parts of the world including South America where

meliponiculture (the practice of keeping stingless bees) is extensively practiced. This belief is based on paleontological and bio-geographical data and is also supported by the fact that their primitive species with a well-developed sting system are found in Africa (Nkoba, 2012). Throughout the tropics many species of stingless bees have been kept by farmers for their products (honey, propolis and pollen) and also for pollination of fruits and vegetables (Roubik et al., 1995).

Botswana is a semi-arid/arid country with daily mean maximum temperature of 32°C (in summer) and mean annual rainfall varying from less than 250 mm in the southwest to over 650 mm in the extreme north (Turner and Makhaya, 2014). The first official beekeeping in Botswana was started in Kagae in west central Kalahari by the Ministry of Agriculture in the 1970s, in an effort to develop agricultural production in this area (Turner and Makhaya, 2014). In Botswana, the agricultural sector is the main source of food, income and employment for the majority of the rural population and thus the Government

promotes beekeeping. Despite the varied agro-ecology which is suitable for apiculture, the country is not self-sufficient in honey production and imports large quantities of honey annually (Hussein, 2001).

The African stingless bee *Plebeina hildebrandti* Friese is the dominant stingless bee species found in Botswana. It belongs to the family Apidae and the genus *Meliponula* and is native to Africa (Wille, 1983; Eardley, 2004). *Plebeina hildebrandti* Friese lives in perennial colonies with a queen, drones and hundreds of workers. They nest underground and provide honey which in some communities is considered to have medicinal properties (Eardley, 2004). Honey hunting or honey harvesting (the gathering of honey from wild bee colonies) is still practiced by people in parts of Africa, Asia, Australia and South America. In Botswana, it is mostly practiced by people living in rural areas.

The apicultural sector in Botswana is relatively well developed. Since the 1980s, the government of Botswana has established programmes aimed at supporting beekeeping as a way to enhance the apiculture sector (Turner and Makhaya, 2014). However, little attention if any has been given to meliponiculture, and to date, no research and development work has been carried out on honey production from stingless bees in the country.

Kalakamati village is located in the North East District of Botswana. The natural vegetation is predominately *Colophospermum mopane* (mophane) woodland which is not a bee plant species. However, during good rainy seasons, beekeeping in the area is possible as farmers grow crops such as groundnuts and watermelons that are good bee forages (FAO, 2005). Although the arable fields are small, they are often supplemented by backyard gardening. The low production in the livestock and arable farming sectors in this region makes beekeeping an option for farmers to increase their agricultural output and hence it can be an attractive venture (FAO, 2005). The district produces a considerable amount of honey from honeybees (*Apis mellifera*) (FAO, 2005).

In addition to honey from *A. mellifera*, farmers in Kalakamati village harvest honey from stingless bees and use it for different purposes. People use stingless bee honey as a food product with essential nutrients and also use it for its medicinal properties. As is the case in many African countries, there is limited information about the characteristics of honey produced by stingless bees found in Botswana and to date, no study has been conducted to assess the physicochemical properties of honey produced by stingless bees in the country. Stingless bee honey is harvested from the wild and farmers do not keep stingless bees in their backyards or cattleposts as is the case with honeybees. This might be due to the fact that meliponiculture has received little research and development attention and lack of awareness and knowledge about stingless bees.

Characterization of the physicochemical parameters of stingless bee honey is important to ensure product quality and for developing specific legislation for honey produced by these bees in Botswana. The objectives of this study

were to assess the production practices of stingless bees (meliponiculture) in Kalakamati village and to determine physicochemical properties of honey produced by stingless bees (*Plebeina hildebrandti* Friese) in the study area.

MATERIALS AND METHODS

Study area

The study was conducted in Kalakamati village. The area has distinct summer and winter seasons. Winter is generally mild with a minimum temperature of 5 °C and maximum temperature of 23 °C. Summer is hot with maximum average temperature of 30 °C and minimum temperature of 17 °C. The village receives annual rainfall averages of around 500 mm that occurs from October to March (FAO, 2005). Soils are well drained and moderately deep (about a meter) and are of medium texture, developed from basic igneous and metamorphic rocks. The texture varies from loamy sand to clay loam. The village has an ephemeral river known as 'Shashe' and several streams. The direction of drainage is towards the south. The vegetation is characterized mainly by tree savanna with *Colophospermum mopane*, *Vachellia tortilis*, *Grewia monticola*, *Combretum imberbe*, *Sclerocarya birrea* and *Senegalia cinerea* trees predominating. Patchy grass cover is found on most of the communal lands and parts of the freehold land, which are not overgrazed (FAO, 2005).

Survey

A survey was conducted by administering a semi-structured questionnaire from December 2016 to January 2017 in order to determine the practices of honey production by stingless bees in Kalakamati village, to identify the major tree/shrub species used as forage by stingless bees in the area, their flowering season, honey harvest time, method of collection and handling of honey, uses of honey obtained from stingless bees, and constraints and opportunities for production of honey from stingless bees. A total of 15 individuals were selected purposively based on their experience and involvement in stingless bee honey harvesting and use, knowledge on sources of nectar and vegetation for stingless bees and the questionnaire was administered during a face-to-face interview.

Sampling techniques and sample size

A total of four stingless bee honey samples (600 ml each) were collected from the wild in Kalakamati village, each from different nests/locations. The samples were harvested from the southern, northern, and two from central parts of the village, with locations being 2-3 km from each other. The four locations were chosen to represent the village and no variation in composition was observed between the honey samples.

Physicochemical properties of honey

The moisture content of honey samples was determined by measuring the refractive index of the sample with an Abbe Refractometer, using the relationship between refractive index at 20 °C and water content, as described in the harmonized methods of the International Honey Commission (IHC, 2009). Reducing sugars content was determined by the modified Lane and Eynon (1923) method involving the reduction of Soxhlet modification of Fehling's solutions by titrating at boiling point (60 °C) against a solution of reducing sugars in honey using methylene blue as an internal indicator (Pearson, 1971). Sucrose content of the honey samples was determined according to the procedures of Pearson (1971).

Free acidity, pH, total ash and water-insoluble solids of the honey samples were determined according to the procedures of QSAE (2005). Determination of hydroxymethylfurfural (HMF) content of honey samples was based on the measurement of absorbance of HMF at 284 nm using UV Spectrophotometer (Evolution-201-UV Visible Spectrophotometer, Thermo Scientific). In order to avoid the interference of other components at this wavelength, the difference between the absorbance of a clear aqueous honey solution and the same honey solution after addition of bisulphite solution was determined. The HMF content was calculated after subtraction of the background absorbance at 336 nm (QSAE, 2005).

Descriptive statistics were used to analyse the data generated in this study.

RESULTS**Honey production by stingless bees in Kalakamati village**

The practice of honey production by stingless bees in Kalakamati village is indicated in Table 1. The stingless bee species found in Kalakamati village is *Plebeina hildebrandti* Friese (Figure 1). According to the respondents, stingless bee honey is harvested in the study area mostly during and after the rainy season (November-May). Honey from the underground nest (Figure 2) is harvested by careful digging of the ground which involves use of different tools such as shovels, picks, axes, buckets, flexible stick and a sharp metal rod. The process starts by inserting a thin flexible stick into the tunnel leading to the nest until it reaches the nest sac. The length of the stick determines the depth of the nest. Digging is done using a pick whilst keeping the stick in the tunnel in order to indicate the location of the nest. As the pit gets deeper digging using a pick becomes difficult and thus a sharp metal rod is used. As the depth approaches the nest, digging is done cautiously to avoid breaking the nest which can lead to the honey being contaminated by soil.

Table 1. Meliponicultural practices in Kalakamati village as reported by the respondents (n = 15)

Variables	Responses
Nesting place	<ul style="list-style-type: none"> Underground Tree trunks
Time of harvesting stingless bee honey	<ul style="list-style-type: none"> All year round but mostly during and after the rainy season (November-May)
Honey harvesting	<ul style="list-style-type: none"> Honey is harvested by digging underground nests
Equipment used for harvesting honey	<ul style="list-style-type: none"> Shovel, pick, axe, buckets, flexible stick, sharp metal rod
Containers and storage conditions	<ul style="list-style-type: none"> Containers: buckets, bottles, clay pots Storage conditions: stored in cool place, refrigerators
Enemies of stingless bees	<ul style="list-style-type: none"> Honey badger, ants, ant bear, termites
Challenges in collecting/producing honey from stingless bees	<ul style="list-style-type: none"> Hard surfaces of the nesting ground, tree roots, underground rocks, depth of nest, loss of entrance tube during digging and small quantities of honey
Medicinal uses of honey from stingless bees	<ul style="list-style-type: none"> To treat cough, stomach ache, ulcer, flu and can also neutralize food poisoning
Other uses of honey from stingless bees	<ul style="list-style-type: none"> Used in tea as a substitute for sugar Used as a relish Used as insect repellent in cowpea field Can be cooked to make jam

n = total number of respondents



Figure 1. Stingless bees with large brown queen in the centre



A



B

Figure 2. Underground stingless bee nest (A = before opening; B = after harvesting of honey)



Figure 3. Honey comb of stingless bees before draining



Figure 4. Draining of honey from the comb



Figure 5. The clear stingless bee honey obtained after draining

After reaching the nest, the pot can be removed carefully from the pit, or it is opened whilst it is in the pit and then the honey combs (Figure 3) are removed and put in a bucket, after which the honey is allowed to drain (Figure 4). After harvest, honey is placed in buckets, bottles or glass bowl (Figure 5) and stored in a cool place and in some cases in a refrigerator.

Stingless bee honey is used for different purposes in the study area. It is consumed as is, added to tea as a substitute for sugar, used as insect repellent in cowpea fields and used to make jam. The respondents also indicated that honey from stingless bees is used for its medicinal properties to treat cough, stomach ache, ulcer, flu and neutralize food poisoning.

The major challenges encountered during honey harvesting in the study area include hard surfaces of the nesting ground, tree roots that make digging difficult, underground rocks, depth of the nesting place, loss of entrance tube that guides to the nest during digging and the small quantity of honey obtained. In addition to the challenges of harvesting honey, another major problem associated with meliponiculture is the prevalence of different predators of the stingless bees in the study area. The major predators in the study area include honey badger (*Mellivora capensis*), ants, ant bear (*Orycteropus afer*) and termites.

Table 2 shows the plant species used as forage by stingless bees in Kalakamati village and their flowering

season. The major plant species used as forage are *Vachellia tortilis* (mosu), *Grewia monticola* (moretlwa), *Combretum imberbe* (motwere), *Senegalia cinerea* (mosu yo montsho) and *Grewia flavescens* (mokgomphatha). The latter was reported to flower in December but all of the others flower in November.

Physicochemical properties of honey

Table 3 shows physicochemical properties of honey samples produced by African stingless bees (*Plebeina hildebrandti* Friese) in Kalakamati village. The average moisture content of honey samples observed in this study is above the limit of 20% and 21% established respectively by the Codex Alimentarius Commission (2001) and the European Union (Bogdanov et al., 1997) for *A. mellifera* honey. The reducing sugar content of the honey samples in the current study is within the limits of the Tanzanian standards (TBS, 2006) for stingless bee honeys which is a minimum of 50%; however, the value is lower than the acceptable minimum limit of 65% for *A. mellifera* honey according to the harmonized methods of European Honey Commission (Bogdanov et al., 1997).

Sucrose content of the honey samples is less than the maximum acceptable limit of 5% for *A. mellifera* honey according to the harmonized methods of European Honey Commission (Bogdanov et al., 1997) and 6% established by Tanzanian standards (TBS, 2006) for stingless bee honeys.

Table 2. Major plant species used as forage by stingless bees in Kalakamati village and their flowering season (n = 15)

Scientific name	Local name (Setswana)	Onset of flowering
<i>Vachellia tortilis</i>	Mosu	November
<i>Grewia monticola</i>	Moretlwa	November
<i>Combretum imberbe</i>	Motswere	November
<i>Senegalia cinerea</i>	Mosu yo montsho	November
<i>Grewia flavescens</i>	Mokgomphatha	December

n = total number of respondents

Table 3. Physicochemical properties of honey produced in Kalakamati village by the stingless bee *Plebeina hildebrandti* (n = 4)

Parameters	Range	Mean \pm SD
Moisture (% by mass)	27.4 – 30.1	28.4 \pm 1.23
Reducing sugars (% by mass)	59.7 – 63.09	61.0 \pm 1.46
Apparent sucrose (% by mass)	1.6 – 2.8	2.4 \pm 0.5
Free acidity (meq/kg)	27 – 43	36.0 \pm 7.16
pH	4.46 – 5.42	4.77 \pm 0.44
Total ash (% by mass)	0.39 – 0.43	0.42 \pm 0.02
Water-insoluble solids (% by mass)	0.04 – 0.09	0.07 \pm 0.02
Hydroxymethylfurfural (mg/kg)	14.02 – 21.06	17.82 \pm 2.90

n = number of samples; SD = standard deviation

The average free acidity level of honey samples analysed in the current study is less than the maximum limit (40 meq/kg) set by EU standards for honey of *A. mellifera*. It is also less than the limit (85 meq/kg) set by the Tanzanian standards for free acidity of stingless bee honey.

DISCUSSION

Honey production by stingless bees

Stingless bees, like honey bees of the genus *Apis*, live with many individuals in a nest where honey and pollen are stored. Although the amount of honey is generally smaller than in the nests of honey bees, people have used stingless bee honey for many centuries. The dominant stingless bee species that produces honey in the study area is *Plebeina hildebrandti* Friese. According to the respondents, *Plebeina hildebrandti* Friese produces honey underground and rarely nests on tree trunks. This is in line with the report of Eardley (2004) who indicated that stingless bees nest underground and in unoccupied termite mounds in Tanzania and South Africa.

The therapeutic property of stingless bee honey is reported by different authors and this is in agreement with the findings of the present study. Honey from stingless bees has antioxidant, anti-proliferative, anti-bacterial, anti-diabetic and anti-inflammatory properties and it is used to heal wounds (Garedew et al., 2003; Issaro et al., 2013; Jalil et al., 2017). Stingless bee honey contains compounds such as proteins, organic acids, vitamins, flavonoids and acetylcholine that contribute to its therapeutic properties (Garedew et al., 2003; Issaro et al., 2013; Jalil et al., 2017).

Harvesting of stingless bee honey was reported to be the major challenge in the study area. This calls the need for designing and developing an appropriate hive for the stingless bees taking into consideration their unique behavioral and biological characteristics. Domestication of stingless bees in log or box hives has often been used in Africa and elsewhere. Nkoba et al. (2016) reported that the use of vertical compartmented (icipe-4M) hives for rearing three stingless bee species including *Plebeina hildebrandti* increased honey yield and reduced postharvest colony loss in Kenya. Similarly, Sommeijer (1999) reported that use of the Utrecht University–Tobago Hive (UTOB hive, developed in Tobago by Utrecht University) for keeping stingless bees in Trinidad and Tobago improved management and inspection of colonies and allowed quick and efficient harvesting of honey. Halcroft et al. (2013) also reported that keepers of stingless bees in Australia use a variety of hive designs ranging from a simple, wooden box to a complex, insulated hive constructed from PVC.

Predators of stingless bees were reported to be a major problem in the study area. This finding is in line with previous reports such as Sommeijer (1999) and Bradbear (2009) who stated that the major pests of stingless bees are phorid flies (*Pseudohypocera* spp.) and ants. On the other hand, Halcroft et al. (2013) reported that the major

predators of Australian stingless bees are flies, ants, spiders, mites, wasps, birds, lizards, toads and humans.

In Kalakamati village, stingless bee honey is collected from the wild and stingless bees are not kept by farmers. However, in some countries like Tanzania and Australia, stingless bees are kept in hives like honeybees (Bradbear, 2009). Such practices could potentially be adopted in Botswana and would increase honey production from stingless bees and allow management of the colonies.

The plant species growing in the study area could influence the quality of honey produced by stingless bees as the composition of *A. mellifera* honey is known to be influenced by the flora used by bees, as well as by climatic conditions, geological region of production, environmental factors, processing techniques and storage conditions (Moo-Huchin et al., 2015; Gobessa et al., 2012). Ramalho et al. (1990) reported that plants belonging to the genus *Vachellia* and *Senegalia* are important pollen and nectar sources for stingless bees (Melipona) in neotropical habitats.

Moisture content

Moisture is a physical parameter that is related to the climatic conditions and degree of maturity of honey (Moo-Huchin et al., 2015). The moisture contents of honey samples in the present study (27.4 to 30.1%) are in line with the findings of Souza et al. (2006), Nascimento et al. (2015) and Moo-Huchin et al. (2015) who reported moisture content of stingless bee honeys produced in Venezuela, Parana State (Brazil), and Yucatan Peninsula (Mexico) to be 26 to 42%, 25 to 36% and 22.2 to 24%, respectively. They indicated that the moisture contents of stingless bee honeys are higher than those of *A. mellifera* honey. The results are also in agreement with the average moisture ($31 \pm 5.4\%$) content reported by Chuttong et al. (2016) for stingless bee honey produced in Thailand.

According to the Tanzanian standard (TBS, 2006), the maximum moisture content of stingless bee honey should be 30%. Moisture content of honey is an important factor contributing to its stability and shelf life. Higher moisture content may lead to undesirable honey fermentation by yeasts resulting in the formation of ethyl alcohol and carbon dioxide; in turn, the ethanol may break down into acetic acid and water, giving the honey a sour taste and a runny consistency, which makes it unfit for human consumption (Moo-Huchin et al., 2015; Nascimento et al., 2015). The high moisture content in stingless bee honey indicates the need to store the product under refrigerated conditions to avoid modification of the physicochemical properties and spoilage by yeasts.

Reducing sugars

Stingless bee honey has lower reducing sugar content than *A. mellifera* honey. Bogdanov and Kilchenmann (1994) reported that the reducing sugar content of stingless bee honeys varied from 48.18 to 79.94%, which is generally lower than the 68.4 to 77.5% found for *A. mellifera* honeys. The reducing sugar content observed in

the current study is in line with the findings of Nascimento et al. (2015) and Chuttong et al. (2016) who reported that stingless bee honey has lower sugar content and normally fructose is the prevalent monosaccharide, which is one of the factors responsible for its sweetness and its hygroscopicity. The taste of stingless bee honey is influenced by the low sugar content and the acidic pH, which shows a clear preference of consumers for this type of honey (Nascimento et al., 2015).

Apparent sucrose

The sucrose content of honey is influenced by the botanical origin of nectar. Bogdanov and Kilchenmann (1994) reported that the sucrose content (0.52-2.86%) of honey from *Melipona* species produced in Venezuela was found to be lower than that of *A. mellifera* honey (0.42-5.20%). The sugar composition of honey depends on the sugars present in the flora of the area and also on their transformation by the enzymes secreted by bees. Important tree species foraged by bees in the study area (Kalakamati village) belong to the genera *Vachellia*, *Senegalia*, *Combretum* and *Grewia* (Table 2) and the lower sucrose content of honey observed in the present study might be attributed to the types of plant species used by the stingless bee as a source of nectar.

Free acidity

Nascimento et al. (2015) reported that in Brazil honeys of the stingless bees *Cephalotrigona capitata*, *Melipona marginata*, *Melipona quadrfasciata*, *Melipona scutellaris*, *Melipona seminigra*, *Scaptotrigona xanthotricha* and *Tetragonisca angustula* had an average acidity of 34.33, 22.55, 35, 27.25, 30.44, 28.78 and 27 meq/kg, respectively. However, the acidity of *Plebeina hildebrandti* honey samples observed in this study is higher than the Brazilian values. Acidity can be related to the state of maturation of the honey and it increases with fermentation (Alves et al., 2005). Stingless bee honey usually has high acidity in comparison to *A. mellifera* honey and this is responsible for the typical sour taste of honey from stingless bees (Bogdanov and Kilchenmann, 1994; Nascimento et al., 2015). The high acidity of stingless bee honey contributes to the high antibacterial activity reported for these honeys when compared with honey from *A. mellifera* and this is associated with the purported medicinal value of honey from stingless bees (Bogdanov and Kilchenmann, 1994; Garedew et al., 2003).

pH

Acidic pH inhibits the growth of microorganisms and thereby contributes to an increased shelf life of honey. The pH values of stingless bee honey reported by different authors (Bogdanov et al., 1999; Alves et al., 2005; Souza et al., 2008) range from 3.3 to 4.7. Nascimento et al. (2015) reported pH values ranging from 2.93 to 4.08 for *Meliponinae* honey produced in Brazil. The honey samples analysed in the present study showed higher pH values than those reported by the different authors indicated above. The pH of honey can be influenced by the flora used by the bees, storage conditions and the

geographical area where the honey is produced (Terrab et al., 2004).

Total ash

The ash content of honey indicates the amount of minerals found in honey, which is influenced by the botanical origin of the nectar and the type of soil where the stingless bees nest (Nascimento et al., 2015; Garedew et al., 2003). The ash content of the honey samples analysed in the present study (Table 3) are within the maximum limit (5%) established by the Tanzanian standard for stingless bee honey (TBS, 2006). Nascimento et al. (2015) reported ash contents ranging from 0.140 to 0.327% for *Meliponinae* honey produced in Brazil. On the other hand, Bogdanov and Kilchenmann (1994) reported an ash content of 0.12-0.76% for honey of stingless bees from Venezuela.

Water-insoluble solids

Honey's water-insoluble matter includes wax, pollen, honey-comb debris, bees and filth particles. Thus water-insoluble matter is used to determine the cleanliness of honey. The higher the water-insoluble solids, the less clean the honey is and vice versa. The maximum acceptable level of water-insoluble matter in honey from *A. mellifera* is 0.1g/100g as indicated by the Codex Alimentarius Commission (2001) and the harmonized methods of the European Honey Commission (Bogdanov et al., 1997). The honey samples analysed in the current study showed a water-insoluble solids content ranging from 0.04 to 0.09 g/100g with a mean value of 0.07g/100g. The results show that the honey samples used in the present study were clean suggesting good harvesting techniques.

Hydroxymethylfurfural

The hydroxymethylfurfural (HMF) levels in honey have traditionally been used as a measure of honey heating history and therefore honey quality. When honey is subjected to high temperature, inadequate storage conditions or addition of invert sugar, the HMF content increases, indicating its age and/or adulteration (Nascimento et al., 2015). The average HMF content of the honey samples observed in the present study (17.82 ± 2.90 mg/kg) agrees with the findings of Moo-Huchin et al. (2015) who reported a mean HMF content of 17.9 mg/kg for stingless bee honey produced in Mexico. However, it is higher than the HMF content of 8.7 ± 1.2 mg/kg reported by Chuttong et al. (2016) for stingless bee honey produced in Thailand. According to the Tanzanian standard (TBS, 2006), the maximum HMF content of stingless bee honey should be 40 mg/kg and the EU standards also specifies maximum HMF content of 40 mg/kg for *A. mellifera* honey. Thus the honey produced in Kalakamati village appears to be unheated and unadulterated.

CONCLUSIONS

In Kalakamati village, honey from stingless bees is collected from the bush and stingless bees are not kept by

farmers. The stingless bees produce honey underground and occasionally nest on tree trunks. *Vachellia*, *Senegalia* and *Grewia* species are the dominant vegetation in the area that are used as forage by the stingless bees. According to the respondents, honey from stingless bees is used as is, substitute for sugar in tea and for its medicinal properties to treat cough, stomach ache and flu. The results of the laboratory analyses indicate that honey produced by stingless bees in Kalakamati village is of good quality and complies with international quality standards for honey.

Many people in Botswana utilize honey from stingless bees as a food and for medicinal purposes. To date, no effort has been made in Botswana to study and promote the production and utilization of honey from stingless bees. Moreover, currently there is no legislation in the country that specifies quality criteria for honey produced by stingless bees. The present work, therefore, can serve as baseline information for future studies aimed at promoting meliponiculture in the country.

ACKNOWLEDGEMENTS

The authors would like to thank the respondents from Kalakamati village who participated in the survey study. This study was funded by the Department of Tertiary Education of Botswana.

CONFLICT OF INTEREST

None.

REFERENCES

- Alves, R.M.O., Carvalho, C.A.L., Souza, B.A., Sodre, G.S. and Marchini, L.C. (2005). Physico-chemical characteristics of honey samples of stingless bee *Melipona mandacaia* Smith (Hymenoptera: Apidae). *Food Science and Technology (Campinas)* 25(4): 644–650.
- Bradbeer, N. (2009). Bees and their role in forest livelihoods. Non-Wood Forest Products 19. Food and Agriculture Organization of the United Nations, Rome, Italy. pp. 51–53.
- Bogdanov, S. and Kilchenmann, V. (1994). Sugar profiles and conductivity of stingless bee honeys from Venezuela. *Apidologie* 25: 271–288.
- Bogdanov, S., Lullmann, C., Mossel, B.L., D'Arcy, B.R., Russmann, H., Vorwohl, G., Oddo, L., Sabatini, A.G., Marcuzzan, G.L., Piro, R., Flamini, C., Morlot, M., Lheretier, J., Borneck, R., Marioleas, P., Tsigouri, A., Kerkvliet, J., Ortiz, A., Ivanov, T., Vit, P., Martin, P. and von der Ohe, W. (1999). Honey quality, methods of analysis and international regulatory standards: review of the work of the International Honey Commission. *Mitteilungen Aus Dem Gebiete Der Lebensmittel-untersuchung Un Hygiene* 90: 108–125.
- Bogdanov, S., Martin, P., Lullmann, C., Borneck, R., Vorwohl, G., Russmann, H., Persano, L., Sabatini, A.G., Marcuzzan, G.L., Morlot, M., Heretier, J., Flamini, C.H., Marioleas, P., Tsigouri, A., Kerkvliet, J., Ortiz, A. and Ivanov, T.Z. (1997). Harmonized methods of the European Honey Commission. *Apidologie* (Extra Issue): 1–59.
- Chuttong, B., Chanbang, Y., Sringarm, K. and Burgett, M. (2016). Physicochemical profiles of stingless bee (Apidae: Meliponini) honey from South East Asia (Thailand). *Food Chemistry* 192: 149–155.
- Codex Alimentarius Commission (2001). 24th Session, July 2001, adopting the draft revised standard for honey. Alinorm 01/25, Appendix II: 22–24.
- Eardley, C.D. (2004). Taxonomic revision of the African stingless bee (Apoidea: Apidae: Apinae: Meliponini). *African Plant Protection* 10(2): 63–96.
- FAO (2005). Beekeeping development. Government of the Republic of Botswana. Food and Agriculture Organization of the United Nations (FAO), Rome, Italy.
- Garedew, A., Schmolz, E. and Lamprecht, I. (2003). The Antimicrobial activity of honey of stingless bee *Trigona* spp. *Journal of Agricultural Science* 47(1): 37–49.
- Gobessa, S., Seifu, E. and Bezabih, A. (2012). Physicochemical properties of honey produced in the Homesha District of Western Ethiopia. *Journal of Agricultural Science* 56: 33–39.
- Halcroft, M., Spooner-Hart, R. and Dollin, A. (2013). Australian stingless bees. In: Pot-honey: A legacy of stingless bees. pp. 1–54. Vit, P., Pedro, S.R.M. and Roubik, D.W. (Ed) Springer, New York.
- Hussein, M.H. (2001). Beekeeping in Africa II-Central, Southern African countries and islands. In: Proceedings of the 37th International Apicultural Congress 28 October – 1 November 2001, Durban, South Africa.
- IHC (2009). Harmonized methods of the International Honey Commission. http://www.apis.admin.ch/english/host/pdf/honey/HMF_abstract.pdf (accessed 27 May 2017).
- Issaro, N., Weerakul, T., Machana, S., Ornnim, P., Phanudulkitti, C., Srijan, T., Laiwattanaphaisal, J. and Pattarapanich, C. (2013). Stingless bee honey II: Qualitative and quantitative studies on honey produced by three stingless bee species collected from a Mangosteen garden in Chantaburi Province, Thailand. *Thai Journal of Pharmaceutical Sciences* 38 (Suppl.): 16–18.
- Jalil, M.A.A., Kasmuri, A.R. and Hadi, H. (2017). Stingless bee honey, the natural wound healer: A review. *Skin Pharmacology and Physiology* 30: 66–75.
- Lane, J. and Eynon, L. (1923). Determination of reducing sugars by means of Fehling's solution with methylene blue as internal indicator. *Journal of Society of Chemistry of India* 42: 32–36.
- Moo-Huchin, V.M., González-Aguilar, G.A., Lira-Maas, S.D., Pérez-Pacheco, E., Estrada-León, R., Moo-Huchin, M.I. and Sauri-Duch, E. (2015). Physicochemical properties of *Melipona beecheii* honey of the Yucatan Peninsula. *Journal of Food Research* 4(5): 25–32.
- Nascimento, A.S., Marchini, L.C., Carvalho, C.A.L., Araújo, D.F.D., Olinda, R.A. and Silveira, T.A. (2015). Physical-chemical parameters of honey of stingless bee (Hymenoptera: Apidae). *American Chemical Science Journal* 7(3): 139–149.
- Nkoba, K. (2012). Distribution, behavioral biology, rearing and pollination efficiency of five stingless bee species

- (Apidae; Meliponae) in Kakamega forest, Kenya. *PhD Thesis*, Kenyatta University, Kenya.
- Nkoba, K., Raina, S.K. and Langevelde, F. (2016).** A vertical compartmented hive design for reducing post-harvest colony losses in three afro tropical stingless bee species (apidae: meliponinae). *International Journal of Development Research* 6(8): 9026–9034.
- Pearson, D. (1971).** The chemical analysis of foods. 6th Edition. Chemical Publishing Company Inc., New York, pp. 123–126.
- QSAE (2005).** Ethiopian honey standard ES 1202:2005. Quality and Standards Authority of Ethiopia, Addis Ababa, Ethiopia, 17 pp.
- Ramalho, M., Kleinert-Giovannini, A. and Imperatriz-Fonseca, V.L. (1990).** Important bee plants for stingless bees (Melipona and Trigoniini) and Africanized honeybees (*Apis mellifera*) in neotropical habitats: a review. *Apidologie* 21: 469–488.
- Roubik, D.W., Yanega, D., Aluja, S.M., Buchmann, S.L. and Inouye, D.W. (1995).** On optimal nectar foraging by some tropical bees (Hymenoptera: Apidae). *Apidologie* 26(3): 197–211.
- Sommeijer, M.J. (1999).** Beekeeping with stingless bees: a new type of hive. *Bee World* 80(2): 70–79.
- Souza, B., Roubik, D., Barth, O., Heard, T., Enríquez, E., Carvalho, C., Villas-Boas, J., Marchini, L., Locatelli, J., Persano-Oddo, L., Almeida-Muradian, L., Bogdanov, S. and Vit, P. (2006).** Composition of stingless bee honey: setting quality standards. *Interciencia* 31(12): 867–875.
- Souza, M.C.L., Jesus, E.F.O., Lopes, R.T., Lemos, C.S., Borges, V.B., Assis, J.T., Vital, H.C., Vila, A.J.M. and Gomes, S.R. (2008).** Physicochemical and sensory characterization of bees spent blooms wild honey. *Food Hygiene* 22(160): 89–92.
- TBS (2006).** Stingless bee honey – specification. TBS/AFDC 17(3908) P3. Tanzanian Bureau of Standards, Dar es Salaam, Tanzania. pp. 1–5.
- Terrab, A., Recamales, A.F., Hernanz, D. and Heredia, F.J. (2004).** Characterization of Spanish thyme honeys by their physicochemical characteristics and mineral contents. *Food Chemistry* 88(4): 537–542.
- Turner, Q. and Makhaya, B. (2014).** Beekeeping in Botswana. Paper presented at the ApiExpo Africa 2014 held in Harare Zimbabwe, 6–11 October 2014. http://www.apitradeafrica.org/Documents/CS_Papers-ApiExpoAfrica2014/BotswanaApiculture_Situation_Paper_2014.pdf (accessed 13 June 2017).
- Wille, A. (1983).** Biology of the stingless bees. *Annual Review of Entomology* 28: 41–64.